NUTRIENT IMPACT OF SWINE CAFO WASTE ON SURFACE WATER AND GROUNDWATER IN EASTERN NORTH CAROLINA

By

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ABSTRACT

Eastern North Carolina (ENC) is a significant pork producer in the United States, with most swine raised in concentrated animal feeding operations (CAFOs). Swine wastewater typically contains elevated nutrient concentrations and is stored in lagoons (LG) until land application occurs. Periodically, nutrient-rich wastewater is applied to spray fields (SFs) to facilitate plant uptake by crops; however, nitrogen and/or phosphorus may leach into groundwater or run off into streams, potentially degrading water quality located hydraulically downgradient of SFs. The goal of this study was to analyze nutrient concentrations, transport, and physiochemical parameters of wastewater and water resources located hydraulically downgradient from SFs at a swine farm in ENC. Piezometers were installed to monitor groundwater quality in the surficial aquifer upgradient and downgradient of spray fields. Additionally, surface water quality was evaluated for streams upgradient and downgradient of the CAFO. Monthly samples were collected between July 2023 and February 2024, which were analyzed for nitrogen and phosphorus. Results revealed that wastewater irrigation from the LG influenced nutrient concentrations in water resources. Wastewater in the LG had a median Total Dissolved Nitrogen, TDN (1651.12 mg/L) and a phosphate concentration (7.48 mg/L), which was greater than all other groundwater sampling locations. Groundwater beneath the SF contained the next highest nutrient, and background concentrations were low. Streams located hydraulically downgradient from SF also tended to contain nutrient concentrations that were significantly greater than stream nutrient concentrations upstream of the farm (median TDN and phosphate of 1.25 mg/L and < 0.001, respectively). Stream discharge was substantially lower than expected, likely due to drought conditions that occurred before and during the study period. Median discharge at the downstream sampling location was 54.4 L/min, while the streams that drain the farm (East Creek and Seep) had a median discharge ranging from 11.29 to 163.37 L/min. Thus, drought conditions also likely influenced nutrient mass transport, particularly for TDN. The median mass transport for nitrogen and phosphate was 1.01 kg-N/day and 0.04 g-P/day respectively. The mass transport at the UP and DW locations was not statistically significant ($p > 0.05$) for nitrogen and phosphorus, respectively. These results differed from a previous study at the same farm during normal conditions, where stream discharge (median: 2,150 L/min) and nutrient exports were substantially greater (median TDN: 21.4 kg-N/day; median phosphate: 3 g-P/day) than the current study. Results from this study indicated that nutrient concentrations and mass transport typically decreased as distance from the CAFO increased. However, nutrient concentrations suggest that eutrophication could still occur, but mass transport was likely underestimated due to drought. Thus, CAFOs can be a significant source of nutrients to nearby water resources, possibly contributing to environmental and/or public health challenges.

NUTRIENT IMPACT OF SWINE CAFO WASTE ON SURFACE WATER AND GROUNDWATER IN EASTERN NORTH CAROLINA

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LIST OF ABBREVIATIONS

BG	Background
BMP	Best Management Practice
CAFO	Concentrated Animal Feeding Operation
Cl	Chloride
DOC	Dissolved Organic Carbon
DO	Dissolved Oxygen
DTW	Depth to Water
DW	Downstream
EC-D	East Creek Downstream
EC-U	East Creek Upstream
LG	Lagoon
NH_4^+	Ammonium
NO_3^-	Nitrate
NO_2^-	Nitrite
N_2O	Nitrous oxide
NMP	Nutrient Management Plan

ORP	Oxidation–Reduction Potential
P	Phosphorus
PO_4^{3-}	Phosphate
RB	Riparian Buffer
SC	Specific Conductance
SD	Seep Downstream
SF	Spray Field
SU	Seep Upstream
DN	Total Dissolved Nitrogen
TN	Total Nitrogen
TP	Total Phosphorus
UP	Upstream
USDA	United States Department of Agriculture
USGS	United States Geological Survey
USEPA	United States Environmental Protection Agency

CHAPTER 1 : INTRODUCTION AND BACKGROUND

1.1 Nutrients: Environmental Concerns and Sources

Nutrients, like nitrogen and phosphorus, are critical for life since they are fundamental components in biomolecules, like nucleic acids and proteins. These nutrients play a crucial role in supporting life on Earth by providing the raw materials necessary for the complex network of biomolecules required for the growth, maintenance, and functioning of living organisms. Nitrogen is essential due to its role in various biological processes, supplying the energy and structural components essential for life (Canfield et al. 2010). Phosphorus, like nitrogen, is also a critical nutrient for all life forms. It is indispensable for forming nucleic acids and is involved in cell division and growth (USEPA, 2024). While nutrients are necessary for survival, excessive nutrients pose a significant environmental and public health risk.

Elevated nutrient loads to surface water can degrade water quality, potentially impairing water use and endangering human health and/or aquatic ecosystems. Consumption of elevated concentrations of nitrate can pose a direct threat to human health. Health impacts like methemoglobinemia and cancer have been associated with increased nitrate consumption; thus, the US EPA requires that nitrate-nitrogen in drinking water must not exceed 10 mg/L (USEPA, 2023; Weyer et al. 2001). Ward et al. (2005) linked nitrate exposure through contaminated drinking water to health issues, such as birth defects and colorectal cancer. Unlike nitrate-nitrogen, there is no drinking water standard for any phosphorus species, indicating that it does not pose a direct health risk (USEPA, 2024). Ammonium, nitrate, and phosphate are classified as plant-available nutrients, and in excess, these nutrients can contribute to eutrophication and algal blooms. Elevated nutrient concentrations in surface waters can result in algal blooms, some of which produce toxins harmful to humans, pets, and wildlife (Townsend et al. 2003). Thus, ammonium and phosphate can also pose an indirect threat to human health (Mueller and Helsel, 2024). Additionally, eutrophication has also been linked with other socioeconomic challenges that damage human well-being. A study by Dodds et al. (2009) highlighted that the combined costs of eutrophication in U.S. freshwaters were approximately $2.2 billion annually. Potential economic losses can be associated with social, ecological, and policy-related concerns. Taste and odor issues increase in frequency and intensity when eutrophic conditions initiate algal blooms in drinking water supplies. Eutrophication-driven macrophyte growth and algal blooms can hinder recreational boating, aesthetic benefits, and angling activities (Fig. 1) (Dodds et al. 2009). Furthermore, eutrophication threatens aquatic ecosystems by changing their structure, function, and water quality (Smith et al. 2006). Some algal bloom events may also lead to fish kills, which typically occur due to limited dissolved oxygen (DO) in the water, potentially causing hypoxic or anoxic conditions (Diaz and Rosenberg, 2008). Thus, hypoxia or anoxia can significantly disrupt aquatic ecosystems, potentially resulting in loss of biodiversity.

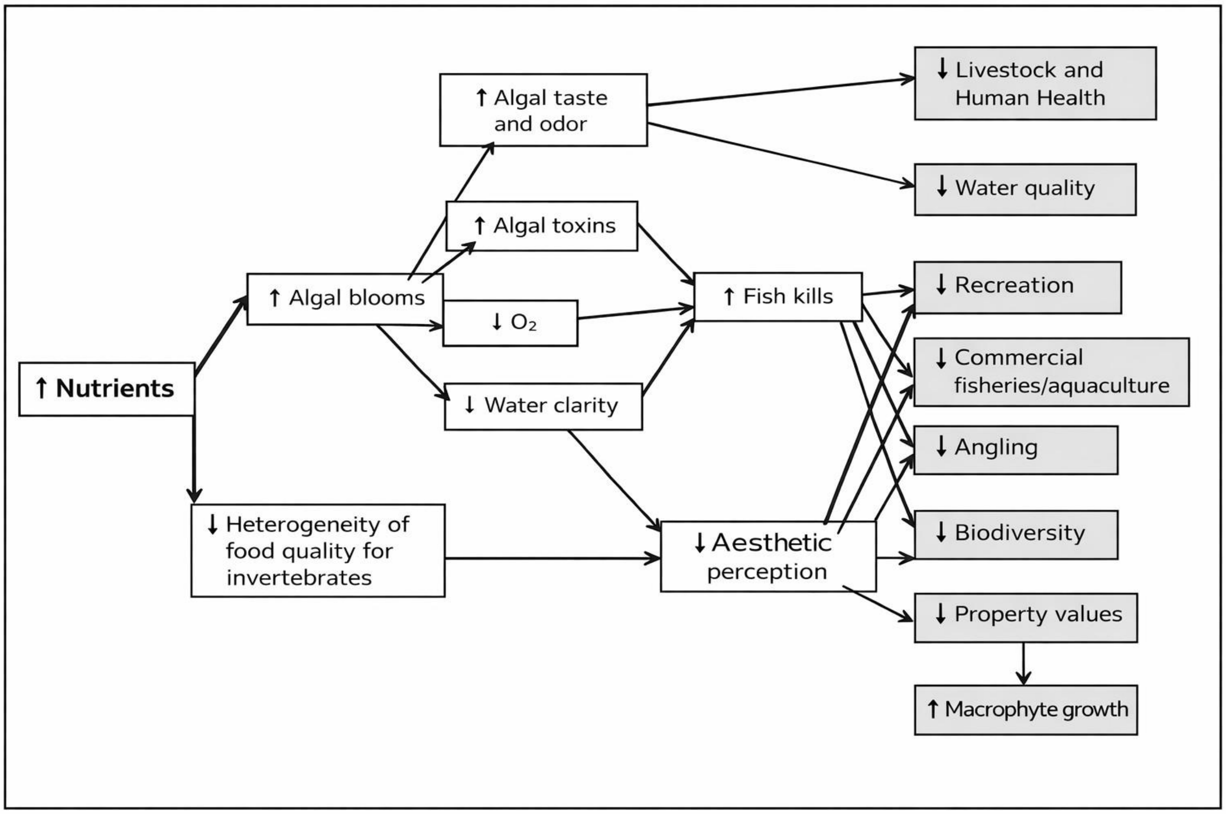

Figure 1.1: Socioeconomic and environmental effects from nutrient pollution. Nutrient pollution can contribute to algal blooms that may degrade water quality, which can degrade aquatic habitat, endanger public health, and cause billions of dollars in damage to recreation, tourism, real estate, and/or municipal services. The up and down arrows in each box indicate increase or decrease (modified from Dodds et al. 2009).

Animal wastewater from concentrated animal feeding operations (CAFOs) may be a significant source of nutrients to water resources in some settings (Rabalais et al. 2002; Turner et al. 2008). CAFOs are industrialized animal farms that can house hundreds to tens of thousands of livestock that generate considerable volumes of wastewater. Animal wastewater tends to contain elevated total dissolved nitrogen (TDN) and total phosphorus (TP) concentrations, typically ranging from 200 to 2500 mg/L and 22 to 329 mg/L, respectively (Ham and DeSutter, 2000). Wastewater from the lagoon can influence the environment via leakage from lagoons without liners and/or those with damaged liners, and during extreme weather events (e.g., intensive rainfall, cyclonic storms, or other natural disasters) that can result in overflows or runoff from the lagoon to nearby waters (Burkholder et al. 2007).

Swine wastewater is periodically land applied to increase soil fertility and allow plant uptake of nutrients. However, there remain concerns about the environmental impacts of this practice, such as odors, eutrophication, and emission of hydrogen sulfide, ammonia, methane, and other air pollutants (Brown et al. 2020; Paerl et al. 2016). The extent of these impacts from CAFO wastewater depends on the type of nutrient, soil properties, and proximity to waterways (Huddleston, 1996; Burkholder et al. 2007). Subsurface nutrient transport to groundwater and/or surface water varies based on the nutrient species, soil texture, permeability, and organic matter (McDowell and Sharpley, 2003). Wastewater may move quickly through the soil profile, especially in sandy horizons with high effective porosity, while organic matter can enhance denitrification and cation exchange (Huddleston, 1996; Burkholder et al. 2007; Ham and DeSutter, 2000). Additionally, antecedent weather conditions can influence the fate and transport of nutrients during wastewater irrigation events. For example, applying wastewater to saturated soils or irrigating SF during inclement weather can promote leaching into groundwater or overland flow to adjacent streams (Amini et al. 2017; Burkholder et al. 2007). Improperly constructed or malfunctioning lagoons and waste lagoons without liners can also be a source of water pollution via leakage (Burkholder et al. 2007). The proximity of CAFOs to streams and floodplains increases the risk of contamination from animal wastewater, especially during episodic events, such as cyclonic storms that can result in lagoon overflow (Wing et al. 2002; Martin et al. 2018).

1.2 Fate and Transport of Nutrients from Swine CAFOs

Many CAFOs in the United States utilize liquid waste management systems that include flushing fecal waste into pits (or lagoons) and spraying the liquid component on fields. North Carolina (NC) is ranked 3rd in hog production by receipts, generating $3.1 billion in 2022 (USDA Economic Research Service, 2024). In Eastern NC, over 2400 CAFOs are permitted to use liquid waste management systems, posing potential environmental and health threats in the future (Wing et al. 2002, NCDEQ, 2024). These operations also yield over 130 million tons of manure annually in the United States (Burkholder et al. 2007). The flushed animal waste from swine houses is held in a lagoon for temporary storage at these CAFO facilities. Upon entering the lagoon, swine wastewater undergoes primary treatment.

1.2.1 Nutrient Cycling in Swine Lagoons

Primary treatment facilitates the settling of solids and the breakdown of organic matter through enhanced microbial processes, such as mineralizing organic nutrients (He et al. 2021; Mallin and Cahoon, 2003). Anaerobic lagoons create an environment conducive to the production of high concentrations of ammonium nitrogen (NH_4^+) through the microbial deamination of organic nitrogen. In these systems, microbial communities degrade organic matter via anaerobic digestion, releasing biogas (methane and carbon dioxide) and converting complex organic nitrogen into simpler forms, primarily NH_4^+. Key processes such as ammonification, driven by decomposer bacteria and fungi like *Bacillus*, *Pseudomonas*, and *Fusarium*, transform organic nitrogen from manure into NH_4^+. These processes (deamination and ammonification) play a central role in nitrogen transformation within anaerobic lagoons (Mallin and Cahoon, 2003; He et al. 2021;

Martínez-Espinosa et al. 2011). Although lagoons are predominantly anaerobic, localized areas with oxygen may allow for limited nitrification, converting NH_4^+ into nitrate (NO_3^-). However, this process is generally minimal due to the low oxygen levels in lagoons (He et al. 2021). Volatilization is a significant challenge associated with uncovered swine wastewater lagoons. Research by Ducey et al. (2019) and Sousan et al. (2021) has shown that covering these waste lagoons can help reduce ammonium volatilization, minimize gas exchange, and decrease odors.

Primary treatment also influences phosphorus (P) concentrations in swine wastewater. while anaerobic conditions transform organic P into dissolved orthophosphate (henceforth referred to as phosphate). Over time, organic P is mineralized, making it bioavailable and converting it into inorganic P compounds (USEPA 2001; Michalopoulos et al. 2014; USEPA 2016). Raw animal waste consists mostly of organic P (>70% of total P), with the rest composed of inorganic P (primarily orthophosphate/phosphate) formed through microbial decomposition (Sharpley and Moyer, 2000; USEPA, 2016). Key transformations include mineralization, where microbes convert organic P into bioavailable orthophosphates (Sharpley et al. 2003). P-solubilizing/mineralization microorganisms include 34 genera of bacteria, notably *Bacillus*, *Pseudomonas*, *Escherichia*, and *Burkholderia*, with *Bacillus* and *Pseudomonas* extensively studied. *Aspergillus* and *Penicillium* are prominent among fungi, with *Aspergillus* being the most frequently reported genus (Divjot et al. 2021; Pang et al. 2024). Aneja et al. (2008) reported that the pH range for a swine CAFO is 7.7 to 8.5, indicating a high pH value. Michalopoulos et al. (2014) and Burkholder et al. (2007) observed that CAFO waste contains metals such as iron, magnesium, and calcium that accumulate in the sludge.

These metals can form ligands with phosphate, resulting in the precipitation of phosphate; however, this condition is dependent on the pH of the lagoon. Calcium, when added to the lagoon, may recover phosphate as amorphous calcium phosphate under neutral to alkaline conditions in lagoons (Szogi et al. 2018). One challenging aspect of managing anaerobic swine lagoons is the accumulation of sludge in the bottom of the lagoon. Eventually, the excessive accumulation of sludge reduces the liquid storage volume of the lagoon and the ability of the lagoon to treat waste. (Szogi et al. 2018; Hamilton, 2010)

1.2.2 Nitrogen Cycling in Spray Fields

Wastewater from the lagoon is periodically land-applied to SF for soil amendment and to increase storage capacity in the lagoon (Harden, 2015). As wastewater infiltrates the SF, it should encounter aerobic soils that facilitate biological processes that reduce nutrient concentrations. NH_4^+ typically undergoes additional transformations in the subsurface, including volatilization, anaerobic ammonium oxidation (anammox), adsorption, plant/microbial uptake, or nitrification. Volatilization of NH_4^+ to ammonia gas may also occur in aerobic soils after irrigation under suitable soil pH (e.g., pH > 8) (Rochette et al. 2013). Anammox occurs when specific bacteria convert NH_4^+ and nitrite (NO_2^-) into nitrogen (N_2) in oxygen-deprived environments, contributing to N_2 production without generating nitrous oxide (Whalen and DeBerardinis, 2007; Mallin and Cahoon, 2003). Zhang and Okabe (2020) noted that these anammox bacteria consist of 19 species from 6 Candidatus genera (*Brocadia, Kuenenia, Jettenia, Scalindua, Anammoxoglobus,* and *Anammoximicrobium)*. NH_4^+ can also be reduced via adsorption, which occurs when NH_4^+ binds with the soil surface (which tends to be negatively charged) (Ham and DeSutter 2000). Adsorption may only be a temporary reduction mechanism as ammonium can be desorbed from fluctuating water levels or ammonium can be nitrified. Nitrification commonly occurs in aerobic soils, where aerobic bacteria oxidize NH_4^+, converting it to NO_3^-. Nitrification is a two-phase biological process, nitrification by oxidizing bacteria (e.g., *Nitrosomonas*) and nitrite-oxidizing bacteria (e.g., *Nitrobacter*). The efficiency of nitrification is strongly influenced by soil pH, with the optimal range being 6.5 to 8.5 (Whalen and DeBerardinis, 2007; Kowalchuk and Stephen, 2001; Mallin and Cahoon, 2003). NO_3^- is also biologically available and can be reduced via plant/microbial uptake or denitrification. More than 50% of the inorganic nitrogen (NO_3^- and NH_4^+) applied to SF is assimilated by plants (Whalen and DeBerardinis, 2007). However, NO_3^- has a high leachability potential due to its mobility in the subsurface. NO_3^- tends to be highly mobile in most soils due to its negative charge that repels soil surfaces that are also typically negatively charged (Whalen and DeBerardinis, 2007; Ham and DeSutter 2000). NO_3^- can also be removed by denitrification, which typically occurs in anaerobic environments. Denitrification occurs when denitrifying microbes (e.g., *Pseudomonas*, *Azosprillium, and Paracoccus*) convert NO_3^- to dinitrogen gas (N_2) or nitrous oxide (N_2O) (Whalen and DeBerardinis, 2007; Rich et al. 2003). Denitrification occurs under anoxic conditions in the presence of nitrate (NO_3^-) and carbon (Mallin and Cahoon, 2003). It is primarily controlled by oxygen concentration and carbon availability. When groundwater is anaerobic and/or saturated, carbon is typically unavailable, thereby inhibiting denitrification, especially in areas with deep groundwater systems. However, groundwater may migrate through environments with an abundance of carbon (e.g., riparian buffers, some riverine sediments), and these areas can serve as hotspots for denitrification and/or plant uptake due to abundant vegetation (Hefting et al. 2004; Vymazal, 2007). If riverine sediments lack sufficient carbon or riparian buffers are degraded or absent, then nitrate transport from CAFOs to water resources will likely increase.

1.2.3 Phosphorus Cycling in Spray Fields

Wastewater irrigation can also increase the concentration of P in groundwater. Several biogeochemical processes influence P mobility in the subsurface (Dubrovsky et al. 2010; Michalopoulos et al. 2014). Wastewater from CAFOs contains organic P, inorganic P, and colloid-bound P. These P forms behave differently depending on the soil characteristics (mentioned above in Section 1.2.2), affecting their mobility and bioavailability (Gall et al. 2015). P retention in SF occurs through adsorption, precipitation, and plant and microbial assimilation. Soils with high clay content or significant amounts of aluminum, calcium, magnesium, and iron oxides exhibit an enhanced capacity for phosphate sorption (Kleinman et al. 2011; Schelde et al. 2006). This is attributed to the strong binding of phosphate to aluminum, iron oxides, and clay particles. Clay soils, characterized by high surface area and low permeability, are particularly effective in facilitating phosphate retention and treatment. Phosphate removal through precipitation reduces environmental loading and enables nutrient recovery. Optimizing conditions for precipitation (e.g., pH and presence of binding agents) can enhance the efficiency of this removal process (Jordaan et al. 2010). Adsorption and precipitation may not be permanent removal mechanisms of phosphate. Phosphate desorption may occur if exchange sites become flooded due to water table fluctuations or prolonged flooding from extreme weather, P application on saturated soil, tillage management, and runoffs (USEPA, 2016; Kleinman et al. 2011; Schelde et al. 2006). Dissolution of phosphate-bearing minerals is possible if environmental factors (oxygen levels and pH) change.

The internal loading of phosphate (from legacy P) into the environment is significantly high under aerobic conditions and alkaline pH. Similarly, in acidic, anaerobic soils, the internal loading of phosphate remains notably elevated (Nguyen and Maeda, 2016). However, several factors can influence the dissolution of phosphate precipitates, including the sources of phosphate, erosion, soil characteristics, and soil tillage practices. Managing phosphate dissolution is essential, as it can help reduce eutrophication and improve water quality (Kleinman et al. 2011; Sharpley et al. 1994). In addition to these biogeochemical processes, fertilization practices by farmworkers can also influence P availability in the environment. If land application rates of P exceed the crop's need, then P may leach to the groundwater, especially if the soil phosphorus index (P-I) is high (McDowell and Sharpley, 2001). The excessive application of wastewater can overwhelm the soil's capacity to retain phosphate, resulting in leaching and runoff. Liquid effluent from CAFOs enhances the movement of P through preferential flow pathways and colloidal transport, especially during periods of intense rainfall or irrigation (McDowell and Sharpley, 2001; Sharpley and Moyer, 2000). P moves from the soil to water in dissolved and particulate forms through surface and subsurface runoff. Particulate phosphate consists of phosphate bound to soil particles and organic matter, which is eroded during surface runoff. This form accounts for over 75% of the phosphate transported from cultivated land (McDowell et al. 2001; Reddy et al. 1999). However, the risk of groundwater contamination by phosphate is generally low because it strongly binds to soil particles.

Nutrient treatment in the soil is typically influenced by soil properties (e.g., soil composition, texture, and structure), the length of flow paths, and the depth of groundwater. In the North Carolina Coastal Plain, permeable soils and a shallow water table increase the risk of nutrient leaching (Mallin et al. 2015; USGS, 2019). In the soil environment, phosphate interacts with clay, organic matter, and mineral surfaces through adsorption, which reduces its mobility. Despite its generally low mobility, phosphate concentrations in groundwater can increase significantly under certain conditions, such as accumulation of wastewater P in the soil profile, fractured aquifer, and increased contamination of storm runoff (Michalopoulos et al. 2014). Excessive P applications can exceed the adsorption threshold, particularly in areas with a high P-I, which increases the risk of leaching. Over the decades, the amount of P applied has often exceeded the crop need, resulting in a higher soil P-Index (Kleinman et al. 2011; USEPA, 2016; Michalopoulos et al. 2014). Soil properties such as pH (Sharpley et al. 1994) and organic content can reduce the availability of phosphate by adsorbing ions or clay particles. For example, phosphate is more likely to precipitate as calcium phosphate in alkaline soils, and neutral pH increases its dissolution and transport potential (Schoumans et al. 2014; Sharpley et al. 1994; DeRouchey et al. 2001).

1.3 Influence of Swine CAFOs on Water Resources

Nutrients not treated in the soil or groundwater can be transported to adjacent surface waters via groundwater discharge (Harden, 2015). Nutrient leaching is especially challenging to manage in areas with shallow water tables, limited availability of carbon

(Harden and Spruill, 2008; Harden, 2015; Korom, 1992; Tesoriero et al. 2000), and the presence of riparian buffers (Seitzinger et al. 2006). Spruill et al. (2000) found that NO_3^- concentrations in groundwater underlying agricultural fields in the North Carolina Coastal Plain ranged from 2 to 20 mg/L, whereas other studies reported NO_3^- concentrations exceeding 20 mg/L in groundwater downgradient of SFs (Karr et al. 2001; Israel et al. 2005) compared to the background concentrations (<1 mg/L). Two other studies found similar results to Spruill et al. (2000). One of the studies by Burow et al. (2010) reported median NO_3^- concentrations of 3.10 mg/L, while the other study by Nolan and Stoner (2000) reported 3.40 mg/L on agricultural land, which is higher than the urban and undeveloped locations. Historically, phosphate was regarded as immobile in the soil, leading to the application of P fertilizers and manures based primarily on the nitrogen requirements of crops. This often resulted in excessive P application (McDowell et al. 2001; Reddy et al. 1999).

In areas with CAFOs, where soil P levels are already elevated, research has demonstrated increased phosphate concentrations in groundwater (Michalopoulos et al. 2014). Specifically, Michalopoulos et al. (2014) reported that phosphate concentrations in groundwater influenced by swine manure ranged from 0.55 mg/L to 2.58 mg/L. Additionally, Smith et al. (2001) found that reactive P concentrations in runoff varied from 0.87 mg/L (in samples pretreated with alum) to 5.50 mg/L (in untreated wastewater), with a background concentration of 0.55 mg/L. In contrast, Krapac et al. (2002) reported a low detection limit for phosphate concentration in groundwater influenced by swine manure, measuring less than 0.3 mg/L. Like NO_3^-, the leaching potential of phosphate increased in areas with shallow water tables and permeable soils, which can increase phosphate concentrations in groundwater (USGS, 2019).

Multiple studies highlight the significant influence of CAFOs on nutrient concentrations in surface water. CAFOs can be a significant source of total nitrogen, TN (mostly inorganic forms) to surface water (Harden 2015; Mallin et al. 2015; Karr et al. 2001; Harden and Spruill 2008;), particularly during storms (Harden 2015). Mallin et al. (2004) documented elevated NH_4^+ and TN in watersheds with CAFOs, while Harden (2015) observed NH_4^+ and NO_3^- spikes during stormflows. Karr et al. (2001) and Harden and Spruill (2008) also found high NO_3^- concentrations near swine operations and waste application fields, with tile drainage increasing NO_3^- transport. Collectively, these findings indicate that CAFOs and associated drainage systems can contribute to persistently high NO_3^- and NH_4^+ concentrations in nearby streams, especially during storm events. Past studies in North Carolina have found that groundwater comprises approximately 35 - 60% of streamflow; thus, groundwater can be a conduit of nutrients from SFs to adjacent surface water (Spruill et al. 2005; Harden et al. 2013). Streams draining watersheds with swine CAFOs contained significantly greater concentrations of nutrients compared to the streams upgradient of the watershed (Harden 2015; Mallin et al. 2015). Mallin et al. (2015) reported that total nitrogen (TN) concentrations in surface water locations within the downgradient of the Cape Fear River Basin ranged from 0.11 to 46.7 mg/L. Harden (2015) also noted significantly higher TN values, ranging from 0.20 to 17.0 mg/L, in stream samples collected downstream from nearby CAFOs. These concentrations were higher than the background concentrations. Harden (2015) reported a background TN concentration range of 1.0 to 1.3 mg/L. Excessive nutrient loading and insufficient treatment capacity often exacerbate elevated P levels. High discharge events, such as storms, can significantly increase phosphate transport due to the greater water volume and force moving through streams, raising P concentrations in receiving waters (Reddy et al. 1999). In the soil, both inorganic and organic forms of P are present, and their mobility can differ significantly. Organic forms of P tend to be more mobile than their inorganic counterparts and can leach further down into the soil profile. This can raise concerns about the bioavailability of organic P (Schelde et al. 2006; McDowell et al. 2001).

Runoff and erosion are primary pathways for P loss from agricultural land, transporting dissolved and particulate phosphate into streams (Sharpley et al. 1994). Heavy rainfall can flood SFs, causing the desorption of NH_4^+ and phosphate, which may migrate into groundwater or surface waters. Large storms further intensify runoff and erosion, mobilizing dissolved and particulate nutrients into nearby water. Surface runoff occurs when rainfall flows over the land surface and can carry both dissolved and particulate phosphate. Subsurface runoff occurs when water moves through the soil profile and can transport phosphate via matrix flow or preferential flow (Schelde et al. 2006; McDowell et al. 2001). Certain agricultural practices increase the risk of P loss, including excessive fertilizer and manure application, plowing, and the timing of manure application to rainfall events. Plowing can increase the risk of particle-facilitated and dissolved P leaching shortly after the soil inversion (Kleinman et al. 2011; Schelde et al. 2006; McDowell et al. 2001; Petersen et al. 2004).

Given the impact of wastewater irrigation practices on water resources downstream, it is essential to address the environmental challenges that can arise from these agricultural methods. This research examined the concentration and mass transport of nutrients derived from swine wastewater associated with a nearby CAFO. Both groundwater and surface water act as conduits for these nutrients to flow into larger bodies of water, resulting in considerable environmental repercussions. This study will add to the few studies on evaluating and monitoring the impact of CAFOs on nutrient availability in groundwater and surface water quality. These findings will help pinpoint nutrient-sensitive water resources. Comprehensive evaluations, including spatial analyses, can offer valuable insights into CAFOs impact on nutrient cycles and water quality. This information could ultimately lead to improved management strategies designed to mitigate the environmental effects and water quality challenges associated with nutrient management practices.

1.4 Study Goals and Objectives

The goal of this study was to examine the influence of a swine CAFO on nutrient inputs to groundwater and surface water. This was accomplished through the following objectives:

1. Quantify nutrient concentrations and physicochemical parameters in swine wastewater
2. Compare nutrient concentrations and physicochemical parameters in groundwater and surface water downgradient of SFs to upgradient or background concentrations.
3. Estimate nutrient mass reductions in soil beneath and downgradient from SFs.

4. Estimate nutrient mass transport by streams located upstream of the CAFO, on the CAFO, and downstream of the CAFO.

1.5 Thesis Structure

The thesis consists of 5 chapters, which are organized as follows:

Chapter 1: Introduction and Background

Chapter 2: Methods

Chapter 3: Nitrogen Results and Discussion

Chapter 4: Phosphorous Results and Discussion

Chapter 5: Conclusions and Management Implications

CHAPTER 2 : METHODS

2.1 Study Area

The study was carried out at a swine CAFO in Harnett County, NC. Most of Harnett County is within the Coastal Plain, but the county is situated near the boundary between Piedmont and Coastal Plain (Fig. 2.1). The soil series at the site includes Gilead loamy sand (~61%), Blaney loamy sand (~19%), and Roanoke loam (~17%) (Table 1) (USDA, 2024). Gilead (Fine, kaolinitic, thermic Aquic Hapludults) is typified by a loamy sand topsoil texture with very low to moderately high permeability. This well-drained soil type is characterized by its ability to support agricultural activities while minimizing the risks of surface ponding or waterlogging. Gilead is the most dominant soil series found beneath the SFs, including where most of this study occurred (P1-3 and P8-11) and toward the downstream (Figs. 2.1 and 2.2). This soil series has a high clay content, which increases with depth in the soil profile (Table 2.1). This elevated clay content suggests that waste treatment could be effective. However, during the winter months and periods of high seasonal moisture, it may lead to ponding and an escalation of runoff into adjacent surface water bodies. Blaney (Loamy, siliceous, semiactive, thermic Arenic Hapludults) is typically quartz rich with a thick sandy surface layer and develops in warm climates with high leaching and moderate permeability (0.51 to 1.45 cm/hr). It is moderately well-drained, leading to occasional wetness, particularly after heavy rainfall in depressional features or areas with restricted drainage due to subsurface clay layers (USDA, 2024). This soil series is primarily found in the upland portion of the farm, partially extending into the riparian buffer in the upstream reaches of Horse Pen Branch (Fig. 2 and 3). Roanoke (Fine, mixed, semiactive, thermic Typic Endoaquults) is a silt loam that typically contains greater clay content in shallower soil horizons, resulting in poor drainage and low permeability. Unlike Gilead and Blaney soils, Roanoke soils typically feature shallow water tables, usually within 30 cm of the surface. Thus, these soils are not ideal for land application due to the increased potential for ponding and runoff. At the study site, Roanoke is in the region underlying the streams on the farm, such as East Creek and Horse Pen Branch (Fig. 2.2) (USDA, 2024; USDA, Natural Resources Conservation Service, 1999).

Soil data from a previous study conducted at this site were obtained for each piezometer to determine the specific soil mineralogy and how soil properties could aid in understanding nutrient attenuation and treatment (Appendix A).

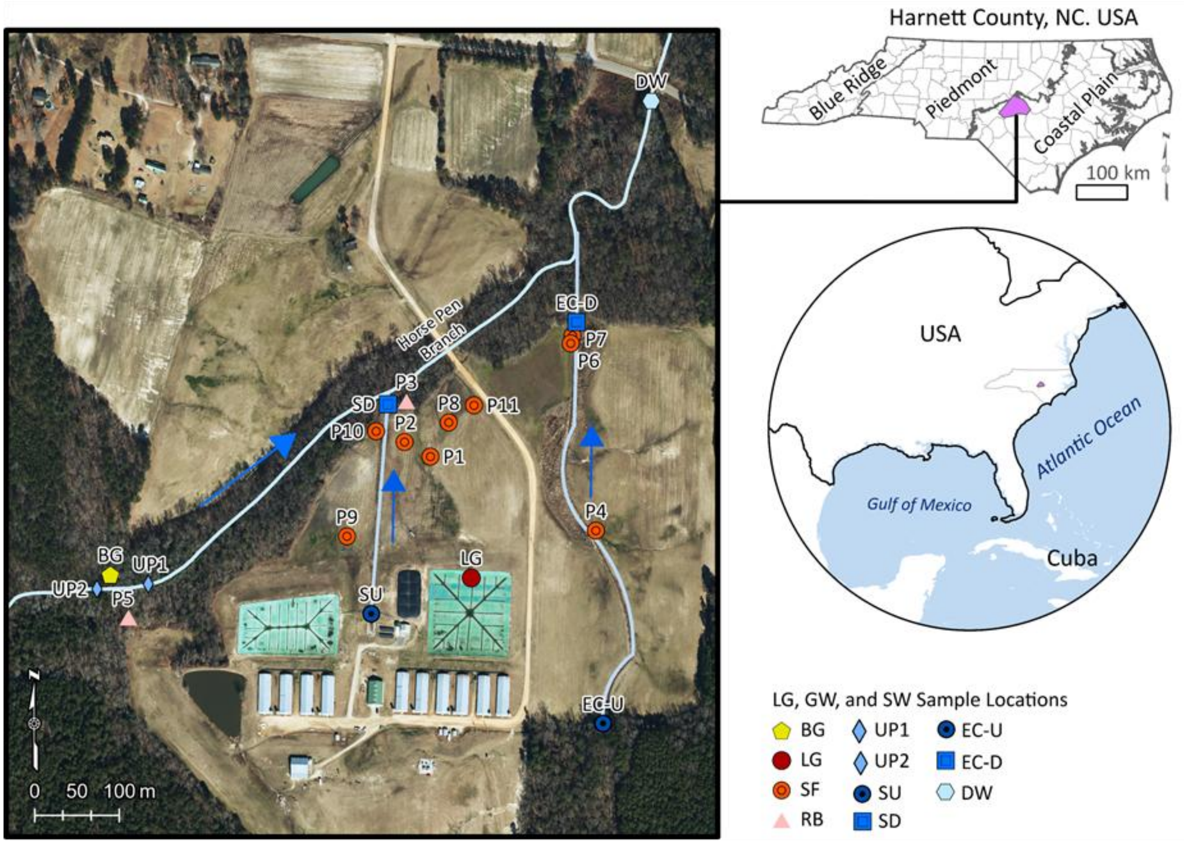

Figure 2.1: A site map illustrating the location of the Background (BG), groundwater (GW), such as riparian buffer (RB), spray field (SF) and surface water (SW) sampling locations, including: Upstream 1 and 2 (UP1 and UP2), Seep Up (SU), Seep Down (SD), East Creek Down (EC-D), East Creek Up (EC-U), and Downstream (DW). The green rectangular features show the location of 2 covered lagoons (LGs), immediately to the north of the hog houses. The blue arrow indicates the direction of the stream flow.

Table 2.1: Summary of soil series and selected soil properties in the study area (USDA, 2024).

Soil Series	Texture	Drainage	Typical Depth to Water	Organic Matter
Gilead	Sandy loam	Moderately Well-drained	46-76 cm (18-30 inches)	Low
Blaney	Sandy loam	Well drained	> 203 cm (>80 inches)	Low
Roanoke	Loam	Poorly drained	0-30 cm (0-20 inches)	High

Table 2.2: Summary of soil series - typical soil profile as described by the USDA (2024)

Soil Series	Depth (cm)	Texture
Gilead	0 - 12.7	Loamy Sand
Gilead	12.7 - 20.32	Sandy Loam
Gilead	20.32 - 106.68	Sandy Clay
Gilead	106.68 - 132.08	Sandy Clay Loam
Roanoke	0 - 17.78	Loam
Roanoke	17.78 - 25.4	Loam
Roanoke	25.4 - 63.5	Clay
Blaney	0 - 10.16	Loamy Sand
Blaney	10.16 - 63.5	Loamy Sand
Blaney	63.5 - 157.48	Sandy Clay Loam

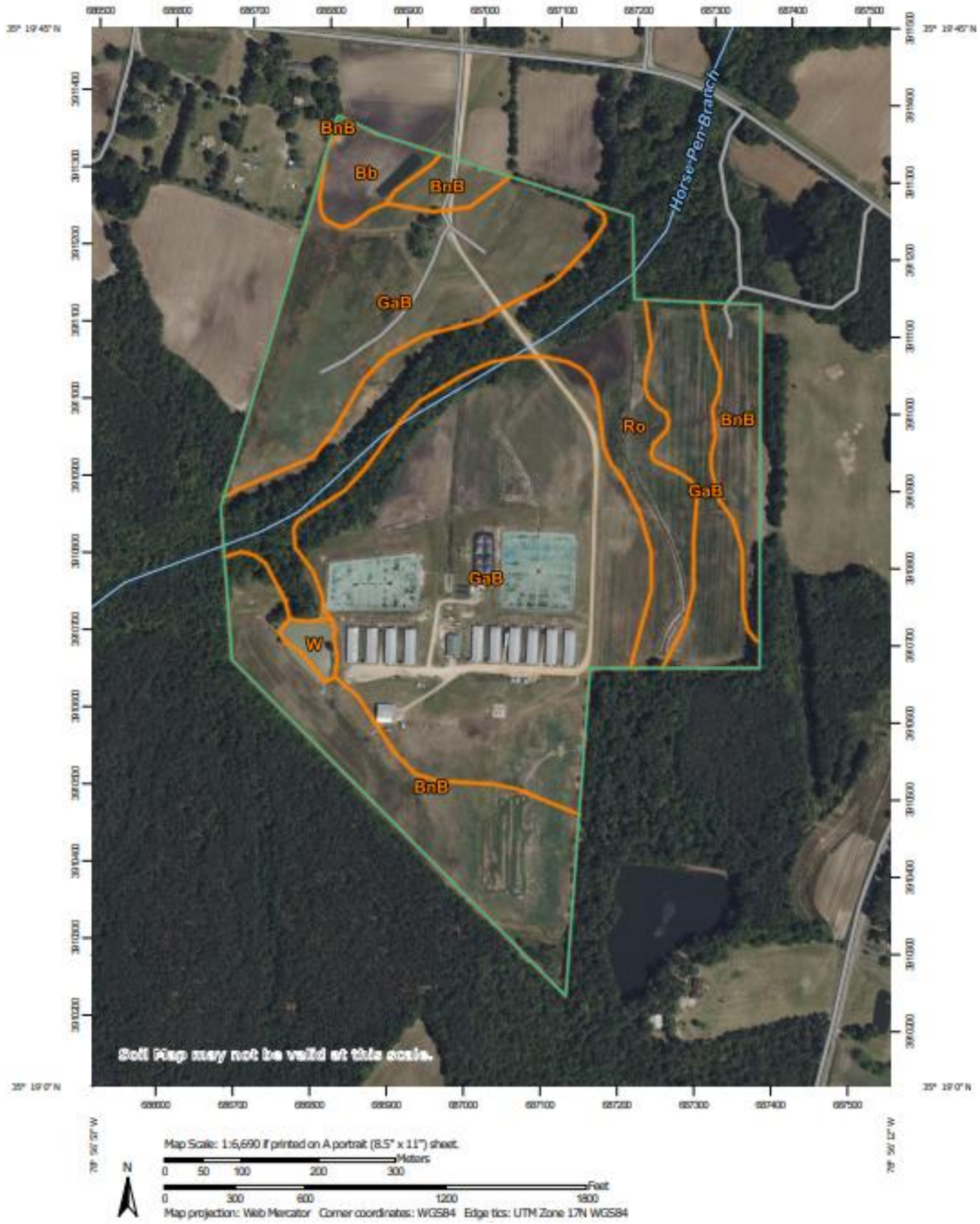

Figure 2.2: This is a soil map indicating the soil types (Gilead – GaB, Blaney – BnB, and Roanoke – Ro) found in the farm location (USDA, 2024). The soil contact boundaries in the figure may not be fully accurate, as they were not mapped at the current scale.

Harnett County experiences a humid subtropical climate typical of the southeastern United States. According to the National Oceanic and Atmospheric Administration (NOAA) and the State Climate Office of North Carolina, the annual average temperature is approximately 16°C (61°F). Winter average temperatures can drop to around 0°C (32°F) on the coldest days, while average summer temperatures can peak at around 32°C (90°F). The county receives an average annual precipitation of about 114 cm (45 inches), with rainfall distributed relatively evenly throughout the year (NOAA, 2023; State Climate Office of North Carolina, 2023).

Horse Pen Branch is a stream that receives drainage from several tributaries located on the farm. However, two major tributaries (East Creek and the Seep) were of research interest, flowing from west-southwest to east-northeast (Fig. 2.1). During winter months, groundwater seeps are often found in low-lying areas due to decreased evapotranspiration and shallower water tables. These seeps tend to create shallow ponds of water near the riparian buffer bordering Horse Pen Branch. In some areas, small channels form that slowly discharge to Horse Pen Branch. Except for the Seep, which daylights between the 2 lagoons (SU in Fig. 2.1), these groundwater seeps tend to be dry during warmer months.

2.2 Site Instrumentation

Groundwater quality was evaluated by installing a piezometer network that facilitated sampling of the surficial aquifer. Hand augers were used to drill to a depth of 0.9 to 1.8 m to the water table or seasonal wetness indicator to ensure piezometers yield a sample. Upon reaching the desired depth, a piezometer was constructed on site using a 5.08 cm diameter (2 inches), schedule-40 polyvinyl chloride (PVC) solid pipe affixed to a section of 5.08 cm, schedule-40 PVC screen. After construction, the piezometer was inserted into the borehole, and sand was poured around the screened interval of the piezometer. Bentonite was poured just below the ground surface. Bentonite acts as an environmental plug that expands when wet and seals the piezometer, thereby preventing surface water from migrating down the side of the pipe. After sealing the piezometers, a disposable PVC bailer was used to purge the piezometers numerous times to remove sediment accidentally incorporated into the piezometers during installation. Finally, piezometers were disinfected using Clorox bleach and purged several times before sampling for the first time. Most of the piezometers were installed within or adjacent to SFs (P1, P2, P6 to P11). An additional 2 piezometers were installed within the riparian buffer adjacent to the Horse Pen Branch (P3 and P5). A piezometer was also installed outside of the influence of SFs to measure nutrient concentrations in background (BG) groundwater (Fig. 2.1). After installing each piezometer, a mobile phone was used to record a latitudinal and longitudinal coordinate.

In addition to the 12 groundwater piezometers, there are 7 surface water monitoring locations along 3 streams that drain the CAFO (Fig. 2.1). The 3 streams include the “Seep” (a surface water feature formed at the point where groundwater discharges to the surface), the “East Creek” (a feature that drains the easternmost SF), and “Horse Pen Branch” (the primary channel that receives drainage from the Seep, East Creek, and other surface water features). There are at least 2 sampling locations for each stream feature, including an “upstream” and “downstream” sampling location. Two upstream locations (UP1 and UP2) were chosen along Horse Pen Branch because the stream was previously observed as dry near UP2 during warmer months. Data from these 2 sampling locations were pooled and categorized as “Upstream”. Staff gauges were installed at 2 locations upstream of the farm (e.g., UP1 and UP2), Seep Down (SD), and 1 location downstream of the farm (e.g., DW). These gauges allow for the determination of the stream stage.

2.3 Sampling Protocol

Wastewater, groundwater, and surface water were sampled approximately monthly between July 2023 and February 2024. During each sampling event, a *Hanna Instruments* (HI) 9829 was used to measure physicochemical parameters such as pH, specific conductance (SC), temperature, DO, oxidation-reduction potential (ORP), and turbidity (streams only). Groundwater samples were collected before sampling the lagoon to reduce contamination potential. Before sampling groundwater from the piezometers, the depth to water (DTW) was measured using a *Solinst* Temperature, Level, and Conductivity meter. The meter was used to measure the DTW and temperature at each piezometer by immersing the probe into the piezometer (Solinst Canada LTs, Georgetown, ON). After measuring DTW, the piezometer was purged by withdrawing at least 2 full bailer volumes to remove stagnant water within the piezometer and allow groundwater to recharge the piezometer. After purging, the same bailer was used to prime a polypropylene sampling bottle and collect a groundwater sample. A portion of the collected groundwater was poured into the calibration cup for the HI-9829 to measure the physicochemical parameters. After all groundwater piezometers were sampled, wastewater from the lagoon was sampled directly from an access point in the lagoon cover using a disposable PVC bailer to collect a sample. A portion of the sample was poured into the calibration cup to measure the physicochemical parameters, and a wastewater sample was collected into a polypropylene bottle for nutrient analysis. Samples were stored in an iced cooler and transported back to the Environmental Research Laboratory at ECU for nutrient analysis.

Surface water samples were also collected on the same day as groundwater and wastewater samples. Grab samples were collected by submerging a primed polypropylene bottle at each surface water location (Fig. 2). Samples were stored using the same protocols as described above for groundwater and wastewater. Physicochemical parameters were measured by submerging the HI-9289 sonde into the stream. Stream discharge (L/s) was calculated at each surface water location to estimate the volume of water that each stream transported. First, the cross-sectional area of the active stream channel was determined by multiplying the cross-sectional width by its average depth, which was calculated by measuring the depth approximately every 15 cm and calculating the average. Streamflow was measured using a *GlobalWater* Probe FP 111 flow meter; however, if the flow was too low to engage the propeller, then either a xanthene dye or a floating object method was used to measure stream velocity. The discharge was calculated by multiplying the stream velocity by the cross-sectional area of the stream.

2.4 Laboratory Analysis

To quantify nutrient concentrations in water samples, samples were initially filtered using a 1.5- and 0.7-micron *Whatman* glass microfiber filter paper. The filtrate was analyzed for NO_3^-, NO_2^-, NH_4^+, phosphate, and chloride (Cl) concentrations using a KPM Analytics SmartChem 170 or 200 discrete auto-analyzer. The SmartChem uses colorimetric analysis to determine concentrations of various ions in water samples by detecting absorbance changes to quantify analyte concentration relative to a standard curve. Additionally, a Shimadzu TOC/TN analyzer was used to measure dissolved organic carbon (DOC) and TDN. The Shimadzu TOC/TN analyzer uses combustion oxidation to convert organic carbon into CO_2 and nitrogen into NO_3^-, which are then detected by infrared or chemiluminescence sensors to measure DOC and TDN, respectively.

2.5 Statistical Analysis

Microsoft Excel was used to organize data, conduct the mixing model, calculate stream nutrient mass transport, generate tables, and calculate summary statistics. Sampling locations were categorized into comparison groups to determine if spatiotemporal patterns existed in nutrient concentrations. Wastewater nutrient concentrations were compared to groundwater to evaluate nutrient concentration reductions. Groundwater piezometers were located in SF and RB downgradient of the farm, and the upgradient location had BG piezometers. Streams were subdivided into 3 comparison groups: upstream of the farm, streams on the farm (e.g., Seep and East Creek), and downstream of the farm. Comparison groups were analyzed using the R statistical framework and the RStudio interface (RStudio Team, 2023). Before conducting any statistical testing, the data were tested against the assumptions of parametric statistics. If the data upholds the assumptions of parametric statistics, then the data was summarized with mean ± standard deviation, and parametric statistics was employed. Otherwise, the median and range with non-parametric statistics were used.

Google Earth Pro was utilized to calculate the areas of three sampling fields: the one located upgradient of the farm (7.89 ha), the one situated towards East Creek (9.67 ha), and the main sampling field containing piezometers (4.85 ha). The site map (Fig. 2.1) displaying sampling locations was developed using ArcGIS Pro. A geodatabase incorporated publicly available datasets, including a raster shapefile of the farm derived from NC OneMap orthoimagery (North Carolina Department of Information Technology, 2024) and a stream shapefile from the National Hydrography Dataset (U.S. Geological Survey, 2024).

The mass input was calculated using the farm irrigation schedule for the period from May to October 2024. The volume of wastewater applied over time (in minutes) was determined by multiplying the flow rate (wastewater irrigation rate) by the duration of application. By multiplying this volume by the median LG concentration, we obtain the mass of nutrients applied during irrigation. This mass of nutrients represents the input from wastewater to SF. The mass budget, which is discussed in chapters 3 and 4, was derived from both the mass input during spray events and the mass exports leaving the farm via stream discharge.

$$Nutrient\ Mass\ Transport = C * Q \qquad \text{Eq 1}$$

Nutrient export (g/day) from each stream was estimated using Eq. 1, where C = nutrient concentration (mg/L), and Q = discharge on a sampling day (L/min). The product of concentration and discharge is then converted g/day or kg/day. Mass reductions of nutrients on the farm were estimated using a two-component mixing model (Eq. 2). This model was used with chloride concentrations in wastewater and groundwater to account for dilution effects and estimate nutrient mass reductions. The fraction of chloride in wastewater and groundwater was estimated using Eq 2.

$$Fraction_{\text{BG-GW}} = \frac{Cl_{lagoon} - Cl_{GW}}{Cl_{lagoon} - Cl_{BG}} \quad \text{Eq 2}$$

$$Fraction_{\text{Wastewater}} = 1 - Fraction_{\text{BG-GW}}$$

The mathematical expression above is the mixing model equation. Where Cl_{lagoon} is the concentration of Cl in the lagoon, Cl_{GW} is the concentration of Cl in the groundwater, and Cl_{BG} is the concentration of Cl in the background (Pinay et al. 1998; Humphrey et al. 2016). The wastewater fraction is multiplied by the nutrient concentration in wastewater (e.g., TDN concentration in the lagoon) to calculate the predicted TDN concentration. This concentration estimates the TDN concentration in the groundwater sampling location if dilution was the only treatment mechanism. If the predicted value is similar to the observed value, it indicates that dilution was a dominant factor contributing to nutrient concentration reduction. The model uses chloride because it typically is not influenced by biological or chemical removal processes like labile nutrients (Li et al. 2019). Furthermore, since this study occurred in a freshwater environment, there is limited Cl that naturally occurs within water resources. Thus, reductions in chloride concentrations from wastewater and groundwater beneath and downgradient of SFs are assumed to represent dilution. The predicted nutrient concentration in groundwater assumes that dilution is the only reduction mechanism. A percentage difference calculation was conducted between the predicted and observed nutrient concentration to estimate groundwater mass reduction percentages. Thus, larger differences in predicted and observed concentrations indicate greater mass reduction.

After a series of diagnostic plotting, analysis, and checking for normality, the dataset did not uphold assumptions of normality. Thus, non-parametric statistics were used to test the hypotheses for this study. The Kruskal-Wallis test was used to detect if significant differences existed in the nutrient concentrations and mass load between the comparison groups in wastewater (LG) and groundwater (e.g., BG, SF, and RB) and surface water (e.g., UP, SU, SD, ECU, ECD, and DW). If a significant difference is detected, a post-hoc Wilcoxon rank sum test was used to identify which groups differed. If a significant difference was found, a post-hoc pairwise Wilcoxon rank sum test with the Bonferroni p-adjustment factor was used to determine statistical significance between comparison groups (Wilcoxon, 1945; Kruskal and Wallis, 1952). A Wilcoxon Rank Sum test was also used to evaluate if the upstream and downstream sampling locations for the Seep and East Creek differed significantly. A significance level of $p \leq 0.05$ was used for all statistical testing.

CHAPTER 3 : NITROGEN RESULTS AND DISCUSSION

3.1 Nitrogen Concentration in Wastewater

The wastewater in the LG had a median concentration of TDN of 1651.07 mg/L, which was 118.86, 705.56, and 1,543 times greater than the median values found in SF, RB, and BG, respectively (Table 3.1; Fig. 3.1). Differences between the TDN concentration in the LG and all other comparison groups were statistically significant ($p < 0.01$). The concentrations of TDN in the LG ranged from 414.39 to 2801.05 mg/L, and TDN concentrations were greater than all other sampling locations 100% of the time. Wastewater contained TDN concentrations similar to those found in past research studies. Previous studies found that TDN ranged from 200 to 2000 mg/L (Ham and DeSutter, 2000), 327 to 756 mg/L (Miner et al. 2003), and 564 to 1783 mg/L (Ducey et al. 2019). A prior study conducted at the same site reported similar TDN concentrations, ranging from 304.8 to 1824.9 mg/L (Richardson 2023). Wastewater TDN concentrations were several orders of magnitude greater than BG, indicating that land application of swine wastewater may be a substantial source of nutrients to water resources if high removal efficiencies of nutrients by soil and cropping systems are not observed.

Nitrogen concentrations in wastewater consisted mostly of ammonium (NH_4^+) and organic nitrogen. The predominant species of TDN was NH_4^+, accounting for 99% of the TDN (Table 3.1; Fig. 3.2). This finding aligns with past studies, as animal waste primarily consists of organic nitrogen and NH_4^+. NH_4^+ is a byproduct of the mineralization of organic nitrogen that typically occurs in anaerobic settings (He et al. 2021; Mallin and

Cahoon, 2003). Ham and DeSutter (2000) noted that LGs are anaerobic environments where nearly all nitrogen exists in the form of NH_4^+. Open CAFO LG facilitates direct exchanges of gas and water, while NH_3 volatilization increases with elevated pH levels (neutral to alkaline), temperature, and wind conditions. This process converts aqueous NH_4^+ into gaseous NH_3, thereby reducing the amount of aqueous nitrogen available for leaching (Rochette, 2013; Szögi et al, 2005). Concurrently, the NH_3 released from CAFO barns and LG can be transported downwind and deposited as reactive nitrogen (NH_3/NH_4^+), contributing CAFO-derived nitrogen to downstream watersheds through both dry and wet deposition (Baker et al. 2020). In the current study, LG is capped by an impermeable synthetic cover, likely limiting volatilization. Ducey et al. (2019) reported that open and closed lagoons had a mean TKN of 473 mg/L (range: 276 mg/L to 630 mg/L) and 1009 mg/L (range: 906 mg/L to 1110 mg/L). Furthermore, the use of covered LG may increase TDN loads to SFs, groundwater, and/or streams if application rates are not guided by nutrient management efforts (e.g., crop nutritional needs, soil fertility, analysis of TDN concentration in wastewater).

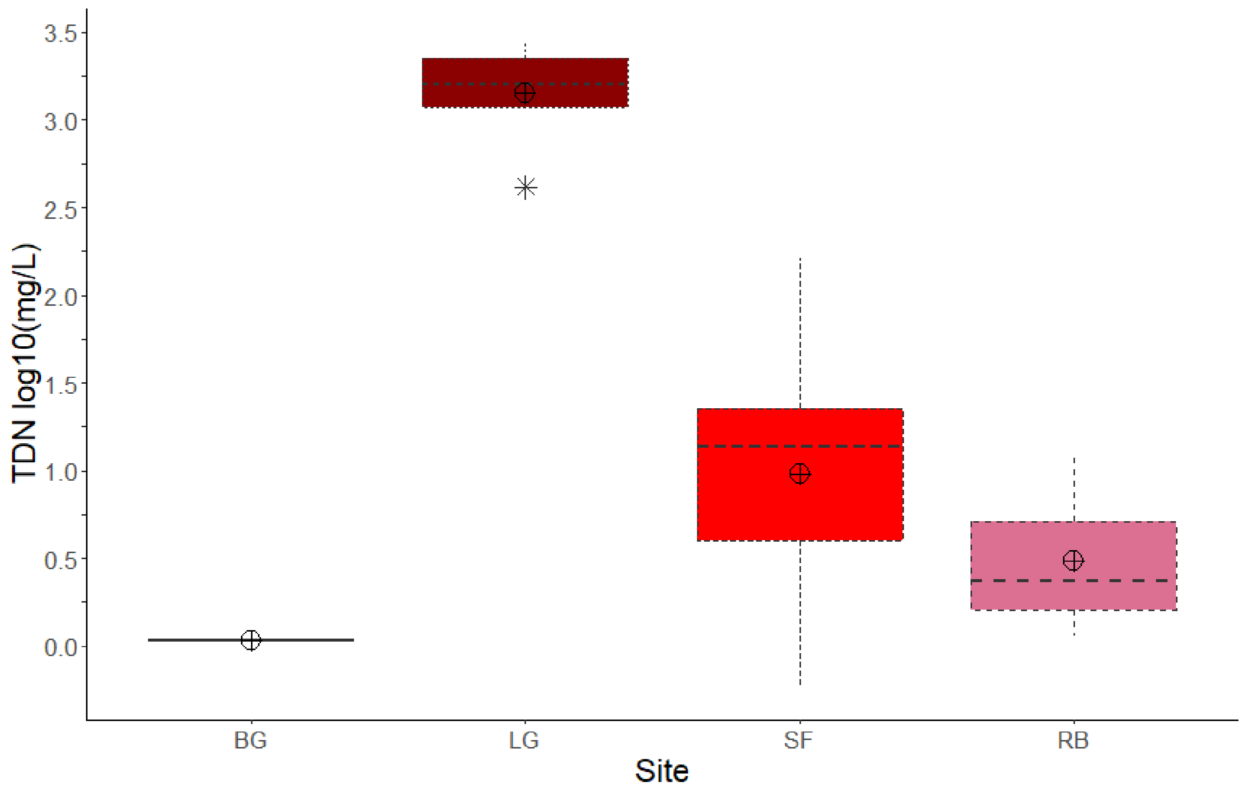

Figure 3.1: Boxplot showing median concentrations of total dissolved nitrogen (TDN) for the lagoon (LG) and groundwater locations, including background (BG), spray field (SF), and riparian buffer (RB).

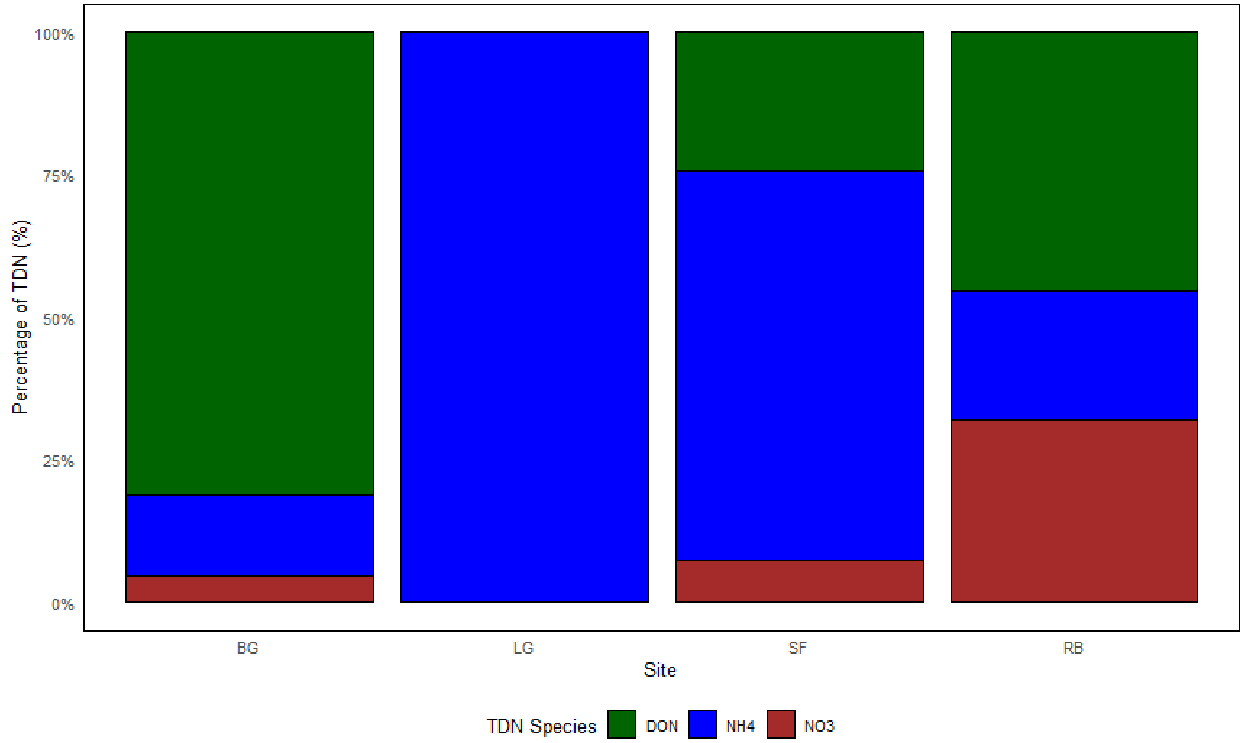

Figure 3.2: A chart showing the percentage of nitrogen species such as dissolved organic nitrogen (DON), ammonium (NH_4^+), and nitrate (NO_3^-) in the lagoon (LG) and groundwater sampling locations, such as background (BG), spray field (SF), and riparian buffer (RB).

Table 3.1: Median (range) of concentrations of DOC= dissolved organic carbon, total dissolved nitrogen (TDN), and its species grouped by sampling location. BG= background, SF= spray field, RB= riparian buffer, and lagoon= LG. The nitrogen species include DON= dissolved organic nitrogen, NH_4^+= ammonium, and NO_3^-= nitrate. (DOC).

Site	NH_4^+	NO_3^-	DON	TDN	DOC
LG	1651.02 (414.39-2801.05)	<0.01 (<0.01-0.84)	<0.01[a]	1651.02 (414.39-2801.05)	1333.00 (704.60-1451.00)
BG	0.15	0.05	0.88	1.07	3.44
SF	10.03 (0.11-162.42)	1.04 (<0.01-20.60)	1.41 (<0.01[a]-6.87)	13.89 (0.58-163.43)	9.06 (2.44-177.40)
RB	0.53 (0.08-3.95)	0.75 (<0.01-8.65)	1.27 (<0.01[a]-5.51)	2.34 (1.14-12.78)	4.70 (1.78-45.58)

[a]= DON was incalculable due to dissolved inorganic species representing 100% of TDN.

3.2 Nitrogen Concentration in Groundwater

Groundwater beneath or near SFs tended to have greater TDN concentrations (Table 3.1; Fig. 3.2). The median concentration of TDN in groundwater beneath the SF was 13.89 mg/L (Table 3.1), which represents a 99% reduction from wastewater. Despite a substantial decrease in concentration, groundwater underlying SFs typically contained elevated TDN concentrations compared to the RB and BG. The median TDN concentration in SF was approximately 6 and 13 times higher than the RB (median TDN: 2.34 mg/L) and BG (median TDN: 1.07mg/L) groundwater, respectively. Groundwater TDN concentrations differed significantly between SF and RB ($p = 0.02$), but not between BG ($p = 0.57$). The lack of statistical significance was due to the limited sample size from the BG ($n = 1$) location. Sampling for the study overlapped with a drought; thus, the BG piezometer was routinely dry. Data from the same piezometer in a previous study were included to supplement water quality data (Richardson, 2023). These additional data decreased the median TDN concentration to 0.43 mg/L, and differences between groundwater beneath the SF and BG were statistically significant ($p < 0.01$; $n = 10$ [9 supplemental + 1 from current study]).

Median TDN concentrations in groundwater beneath the SF were highly variable (range: 0.58 to 163.43 mg/L), but due to the high TDN concentrations of wastewater, the treatment efficiency was between 97.6 to 99.9% (Table 3.1). The highest TDN concentrations were observed in P1, which had a median TDN of 39.92 mg/L (range: 20.98 to 163.43 mg/L; Table 3.2). The median nitrogen treatment efficiency that occurred at P1 was approximately 98% reduction. Of the remaining SF piezometers, P2 consistently had the lowest median TDN concentrations of 1.69 mg/L (range: 1.36 to

3.28 mg/L; Table 3.2). Groundwater TDN concentrations tended to decline from SFs to RB location (Table 3.1). The median TDN concentration in RB piezometers was 2.34 mg/L, representing an 83.1% reduction from the median SF piezometer (Table 3.1 and Fig. 3.1). Nitrogen concentrations were variable in RB piezometers. P3 had a greater median TDN concentration (median: 3.55 mg/L, range: 1.41 to 6.37 mg/L) relative to P5 (median: 1.87 mg/L), but the range was greater at P5 (range: 1.14 to 12.78 mg/L) (Table 3.2). TDN concentrations declined by >99% when comparing the LG and RB piezometers. Despite the large concentration reduction between the RB and the LG, TDN concentrations in groundwater underlying the RBs were about double the concentrations in the BG groundwater. However, this difference was not statistically significant ($p = 1$) due to the drought ($n = 1$ for BG). Statistical analysis was repeated after supplementing with additional BG data from a prior study (as discussed above), yielding a statistically significant difference ($p < 0.01$). The SF and DW TDN concentrations in the current and previous studies vary slightly, respectively. The previous study had an average median TDN for SF of 11. 30 mg/L (range of median value: 1.67 to 27.2 mg/L) while the current study had a TDN median of 13.89 mg/L for the SF location. These data show that land application of swine wastewater can significantly elevate groundwater TDN concentrations relative to the BG. Elevated concentrations in groundwater beneath the RBs suggest that waste-derived TDN may reach adjacent surface waters, potentially contributing to eutrophication issues in receiving streams.

Findings from this study indicated that the land application of swine wastewater can serve as a significant source of nitrogen to groundwater, aligning with previous research

(Burkholder et al. 2006; Spruill et al. 2002; Israel et al. 2005; Richardson et al. 2023). Groundwater TDN concentrations in the current study aligned with a previous study by Spruill et al. (2000) conducted in the North Carolina Coastal Plain. They reported NO_3^- concentrations in groundwater underlying agricultural fields ranged from 2 to 20 mg/L. Two other studies found similar results to Spruill et al. (2000): Burow et al. (2010) reported median NO_3^- concentrations of 3.10 mg/L, while Nolan and Stoner (2000) reported 3.40 mg/L on agricultural land, which is higher than the urban and undeveloped locations. Other studies found NO_3^- concentrations exceeded 20 mg/L in groundwater downgradient of SFs compared to the background concentrations (<1 mg/L) (Karr et al. 2001; Israel et al. 2005). Elevated TDN concentrations were reported in groundwater beneath SFs (Stone et al. 1998a; Karr et al. 2001; Spruill et al. 2002), which were between 4 and 65 times greater than background concentrations (Richardson et al. 2023). Additionally, RB and SF concentrations (Table 3.1) exceed the nutrient reference criteria for Ecoregion IX established by the US EPA, which range from 0.7 to 1.0 mg/L of total nitrogen (TN) (US EPA, 2000). The nutrient reference criteria established by the U.S. Environmental Protection Agency (EPA, 2000) for Ecoregion IX (Southeastern Temperate Forested Plains and Hills) represent baseline or reference conditions for nutrient concentrations (TN) in surface waters that are likely minimally impacted by human activities.

Past studies, along with results from the current study, suggest that CAFOs can significantly influence groundwater nitrogen concentrations. Thus, implementation of best management practices (BMPs) is suggested to enhance nitrogen treatment by facilitating nitrogen transformations. Examples of these BMPs, including subsurface bioreactors, extended buffer zones, or wetlands can enhance denitrification and facilitate nitrogen removal in surface water and groundwater. A study by Messer et al. (2012) in North Carolina found that riparian buffers in the Conservation Reserve Enhancement Program significantly reduced NO_3^- levels in groundwater. Past studies found that RBs can reduce groundwater NO_3^- concentrations by about 2.5 to 11.5 mg/L (approximately 61 to 99%) at the site scale (Messer et al.; Hyatt et al.; Harden and Spruill, 2008). Richardson (2023) reported reduced mean TDN concentration in RB (range: 2.85 mg/L to 9.10 mg/L) compared to the SF location (range: 1.67 mg/L to 27.2 mg/L). Comparable results have been observed in non-CAFO agricultural systems, where cover crops lowered NO_3^- leaching by nearly 70%, and denitrifying bioreactors and controlled drainage systems achieved 20 to 50% and 15 to 40% reductions, respectively (Nouri et al. 2022; Rosen and Christianson, 2017; Bodrud-Doza et al. 2025). Factors such as land slope, soil layers, water table depth, dissolved organic carbon, the duration of groundwater residence, bioreactors, extended buffer zones, and wetlands all influenced the effectiveness of the buffers in reducing NO_3^-. Furthermore, the dominant species of nitrogen is an important consideration in BMP selection. In the current study, NH_4^+ was prevalent in several groundwater piezometers. Thus, subsurface denitrification-based BMPs will not be as effective due to the increased NH_4^+ concentrations. Implementing these BMPs would require pretreatment to aerate NH_4^+-rich groundwater to maximize denitrification.

Table 3.2: Median (range) of DON= dissolved organic nitrogen, NH_4^+= ammonium, NO_3^-= nitrate, total dissolved nitrogen (TDN), and DOC= dissolved organic carbon. The groundwater (GW) sample locations/piezometers (P), BG= background, and LG= lagoon. The BG piezometer had one sample of data because it is often dry during the sampling regime.

Site	Location	NH_4^+	NO_3^-	DON	TDN	DOC
LG	Wastewater	1651.018 (414.39-2801.05)	0.02 (0.0001-0.84)	<0.01[a]	1651.02 (414.39-2801.05)	1333.00 (704.60-1451.00)
BG	Background	0.15	0.045	0.88	1.067	3.45
P1	Spray Field	32.84 (12.78-162.41)	5.63 (1.01-8.89)	<0.01[a] (<0.01a-5.83)	39.92 (20.98-163.43)	21.14 (17.45-177.40)
P2	Spray Field	0.58 (0.11-3.24)	0.05 (0.02-0.09)	0.79 (<0.01[a]-1.43)	1.69 (1.36-3.28)	6.08 (3.98-18.56)
P3	Riparian Buffer	0.24 (0.080-3.94)	0.80 (0.12-3.11)	0.37 (<0.01[a]-3.33)	3.55 (1.41-6.37)	10.45 (4.60-45.58)
P5	Riparian Buffer	0.71 (0.12-1.11)	0.44 (0.01-8.65)	0.66 (<0.01[a]-5.51)	1.87 (1.14-12.78)	3.85 (1.78-5.78)
P6	Spray Field	10.05 (3.34-12.95)	1.11 (1.07-4.90)	0.14 (<0.01a-5.25)	11.15 (4.55-23.10)	24.62 (22.05-46.48)
P7	Spray Field	3.62 (0.34-16.54)	0.21 (0.0001-6.44)	0.63 (<0.01[a]-6.87)	9.67 (3.84-16.54)	12.31 (8.69-33.42)
P8	Spray Field	1.02 (0.11-11.66)	2.23 (0.0001-7.68)	0.57 (<0.01[a]-4.00)	4.54 (0.58-18.43)	5.03 (3.48-7.97)
P9	Spray Field	13.52 (10.00-28.15)	3.35 (0.03-8.89)	<0.01a (<0.01[a]-4.61)	19.51 (10.04-37.02)	5.53 (3.78-14.48)
P10	Spray Field	10.04 (8.89-11.18)	18.41 (7.73-20.60)	5.59 (<0.01a-14.66)	28.45 (22.40-31.78)	2.81 (2.44-3.94)
P11	Spray Field	13.79 (4.30-21.31)	0.14 (0.03-2.16)	1.29 (<0.01[a]-5.26)	13.83 (5.59-21.45)	44.36 (32.58-54.90)

[a]= DON was incalculable due to dissolved inorganic species representing 100% of TDN

3.3 Mass Reduction Estimates

Ratios of Cl/N and a two-component mixing model suggest that TDN mass reductions occurred in the vadose zone and/or the surficial aquifer system. Ratios of Cl/N increased from SF to the RB locations, suggesting that nutrient treatment may be occurring. The ratio of the median Cl/N concentration was 0.65 in the LG. The ratio of Cl to N in SF groundwater was 2.38 (about 4 times greater than the LG), while groundwater beneath RBs exhibited the highest Cl/N ratio (17.01). This increasing trend indicates that TDN concentrations are decreasing more than Cl concentrations in groundwater. Reductions due to dilution alone would decrease TDN and Cl at a 1:1 ratio; thus, the increased reduction of TDN relative to Cl suggests that nitrogen mass reduction occurred. This supposition is also supported by mixing model results. The observed TDN concentration (i.e., laboratory quantified TDN) was consistently lower than the predicted TDN concentration (i.e., TDN concentration if dilution were the sole reduction mechanism), regardless of sampling location (Table 3.3). The estimated mass reduction of TDN was > 90% for groundwater underlying SFs (91%) and RBs (96%) (Table 3.3). The most likely processes that explain TDN mass reduction include volatilization, denitrification, anaerobic ammonium oxidation (anammox), plant and microbial uptake, or adsorption. The overall trend in the Cl/N ratios indicates an increase from LG and BG locations toward SF and RB. This trend is consistent with findings by Richardson (2023), who also reported that the Cl/TDN ratio in groundwater piezometers exceeded that of LG wastewater (1.53). The overall mass reduction estimates suggest that biogeochemical processes are effective (> 90%); however, mass treatment exhibited more variability when evaluating individual piezometers.

Table 3.3: Two-component mixing model using Cl= chloride and TDN= total dissolved nitrogen to estimate mass reduction of TDN. Cl, fraction WW= wastewater, fraction GW= groundwater, predicted TDN, observed TDN, and TDN mass reduction are shown for sites LG= Lagoon, SF= Spray field, RB= Riparian Buffer, and BG= Background.

Site	Cl (mg/L)	Fraction WW	Fraction GW	Predicted TDN (mg/L)	Observed TDN (mg/L)	Cl/TDN Ratio	Mass reduction (%)
LG	1076.00	1.00	0.00		1651.12	0.65	
BG	3.93	0.00	1.00		1.07	3.68	
SF	77.13	0.07	0.93	112.74	10.03	7.69	91.10
RB	39.81	0.03	0.97	55.26	2.34	17.01	95.77

The differences in concentrations of TDN and Cl/TDN ratios in groundwater samples from the piezometers and wastewater samples from the lagoon suggest nitrogen attenuation processes occurred in the soil and groundwater beneath the spray fields. Median TDN mass reductions across the groundwater piezometers ranged from approximately 8% to 96% (Table 3.4). SF piezometers (Fig. 2.2) exhibited the greatest variability, with reductions ranging from 8% at P2 to 96% at P8. Only two SF piezometers (P8 and P9) exceeded 85% reduction, while most others showed moderate reductions between 30% and 75%, excluding P2 and P10, with lower values (~15%). The limited mass removal at P2 likely reflects its position outside the primary influence of wastewater irrigation, as indicated by low median concentrations of both TDN (1.69 mg/L) and chloride (5.16 mg/L), values similar to the background (BG) site. For other SF piezometers with relatively low reductions (P6, P7, P10, P11), low Cl concentrations suggest that dilution, rather than active biogeochemical processing, played a larger role in reducing TDN concentrations. In contrast, the mass removal observed at the RB piezometers was notably consistent, with both demonstrating approximately a 95% reduction. This trend indicates that the subsurface conditions within the riparian buffers characterized by abundant vegetation, organic matter, and extended residence time facilitate enhanced nitrogen removal through mechanisms such as plant uptake, microbial denitrification, and adsorption. Collectively, these findings underscore the effectiveness of natural and vegetated systems, like SFs and RBs, as nature-based treatment zones that mitigate nitrogen levels before they reach surface waters.

Table 3.4: Two-component mixing model using Cl= chloride and TDN= total dissolved nitrogen to estimate mass reduction of TDN. Cl, fraction WW= wastewater, fraction GW= groundwater, predicted TDN, observed TDN, and TDN mass reduction are shown for sites P= Piezometers, LG= Lagoon, and BG= Background

Site	Location	Cl (mg/L)	Fraction WW	Fraction GW	Predicted TDN (mg/L)	Observed TDN (mg/L)	Cl/TDN Ratio	TDN Mass Reduction (%)
LG	Wastewater	1076.00	1.00	0.00		1651.02	0.65	
BG	Background	3.93	0.00	1.00		1.07	3.68	
P1	Spray Field	104.35	0.09	0.91	154.65	39.92	2.61	74.19
P2	Spray Field	5.12	0.00	1.00	1.83	1.69	3.03	7.78
P3	Riparian Buffer	49.43	0.04	0.96	70.07	3.55	13.92	94.93
P5	Riparian Buffer	30.95	0.03	0.97	41.61	1.87	16.55	95.51
P6	Spray Field	11.78	0.01	0.99	12.09	7.86	1.50	34.98
P7	Spray Field	19.88	0.02	0.98	30.50	9.67	2.06	68.30
P8	Spray Field	80.52	0.07	0.93	117.95	4.54	17.74	96.15
P9	Spray Field	96.18	0.09	0.91	142.07	19.51	4.93	86.27
P10	Spray Field	25.65	0.02	0.98	33.45	28.45	0.90	14.95
P11	Spray Field	26.94	0.02	0.98	35.44	13.83	1.95	60.97

3.4 Physicochemical Parameters in Wastewater and Groundwater

The LG had the highest median SC of 13105.00 µS/cm, representing an increase of about 30, 62, and 423 times compared to the median SC in the SF, RB, and BG piezometers, respectively (Table 3.5). These differences were statistically significant ($p < 0.01$). The median SC in the groundwater beneath the SF site was 432 µS/cm, which is approximately 2 and 14 times higher than that observed in the BG and RB sites, respectively (Table 3.5). These differences were also statistically significant ($p < 0.01$). Median SC values varied from 48 µS/cm (P2) to 1,025 µS/cm (P1) in SF groundwater (Table 3.5). The difference between the SF and RB was statistically significant ($p = 0.0007$). The overall median SC concentration for RB ranged from 58 µS/cm to 319 µS/cm (median: 213.00 µS/cm). Despite elevated SC values in RB groundwater, SC was not significantly different from the BG ($p = 0.2$), which likely occurred due to drought in the BG location during the sample period. Supplementing BG groundwater with prior data (Richardson 2023) resulted in a similar median (29 µS/cm) and a statistically significant difference from all other groundwater comparison groups ($p < 0.01$). The most likely explanation for this difference was due to the application of swine wastewater (Michalopoulos et al. 2014). Groundwater beneath SFs (e.g., P1, P11, P6, P8, P9, P7, and P10) tended to contain elevated SC (300 to 700 µS/cm) relative to RB (P3 and P5) and BG groundwater. A study by Michalopoulos et al. (2014) found that groundwater from monitoring wells adjacent to a swine CAFO showed increased SC values (ranging from 814 to 1,965 µS/cm) due to wastewater application. This finding was congruent with previous studies showing elevated SC in water resources influenced by wastewater (Michalopoulos et al. 2014; Richardson 2023; Burkholder et al. 2007).

Like SC, pH varied between sampling locations. The highest pH was observed in the LG (median: 8.5), which was expected since swine wastewater tends to be alkaline (Mallin and Cahoon, 2003). Groundwater beneath SFs had median pH values that ranged from about 4.7 to 6.7, whereas median pH in RB groundwater was < 5 (Table 3.7). The decline in pH between the LG and SFs suggests that nitrification may be occurring. Nitrification results in the loss of hydrogen ions from NH_4^+, thereby reducing the pH value of the environment (Le et al. 2019). BG groundwater had a near neutral pH (median: 6.8), which was similar to groundwater in predominantly forested land (Liu et al, 2021). The DTW values indicated that the farm had some shallow wells, with P8 and P10 having the lowest minimum range and median DTW, while P1 and P11 had the highest DTW. This reflects substantial spatial variability in topography and subsurface hydrology (DTW range for all groundwater locations was 3.35 to 233.17 cm). The DTW is also crucial because it accounts for the vadose zone necessary for nitrification to occur under suitable conditions. DO concentrations tended to be low in most groundwater samples (median values < 2 mg/L for most piezometers. ORP values were negative for the LG, which was expected due to the high concentrations of organic matter creating a reducing environment. Overall, groundwater tended to have a positive ORP value, although some piezometers (P11, P6, P7) had a median ORP value that was negative, ranging from -71 to -110 (Table 3.7). These reducing environments may likely limit nitrification in the overlying vadose zone and incomplete nitrogen transformation, which may explain why these same piezometers exhibited the lowest mass reduction estimates (34.98, 68.30, and 14.95% respectively; Table 3.4). The temperature did not differ much between sampling locations.

Table 3.5: Summary of median and range for physiochemical parameters: Temperature (Temp); specific conductance (SC); dissolved oxygen (DO); oxidation-reduction potential (ORP); depth to water (DTW) in groundwater and wastewater locations such as lagoon (LG); background (BG); spray field (SF); riparian buffer (RB).

Site	DTW (cm)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
LG		21.83 (11.52-30.70)	13105.00 (5735.00-14970.00)	8.50 (6.75-8.74)	-213.50 (-315.90- 5.40)	0.31 (0.01-1.00)
BG	137.16[a]	21.10	31.00	6.84	219.00	2.95
SF	82.92 (3.35-233.17)	22.33 (12.70-27.70)	432.00 (48.00-1025.00)	5.80 (4.56-6.93)	56.60 (-153.00-226.00)	1.06 (0.01-5.97)
RB	74.83 (24.99-145.69)	20.02 (12.10-24.10)	213.00 (58.00-319.00)	4.90 (4.6-6.90)	106.35 (-32.30-287.10)	1.81 (0.46-6.70)

[a]= DTW measured in July 2023, all other sampling events were dry, thus DTW was > 232 cm (total piezometer depth).

Table 3.6: Summary of median and range for physical and chemical parameters in groundwater (GW) piezometers. pH, temperature (Temp), specific conductivity (SC), dissolved oxygen (DO), depth to water (DTW), and oxidation reduction potential (ORP). The groundwater groups include Lagoon (LG), spray field (SF), and riparian buffer (RB).

Site	Location	DTW (cm)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
LG	Wastewater		21.83 (11.52-30.70)	13105.00 (5735.00-14970.00)	8.50 (6.75-8.74)	-213.50 (-315.90-5.40)	0.31 (0.01-1.00)
BG	Background	137.16[a]	21.1	31.00	6.84	219	2.95
P1	Spray Field	158.50 (121.92-195.99)	20.10 (13.20-25.70)	668.50 (360.00-1025.00)	5.66 (5.20-6.03)	72.00 (-107.80-153.60)	1.40 (0.01-2.77)
P2	Spray Field	104.85 (63.53-121.01)	23.25 (13.30-27.30)	85.00 (48.00-150.00)	5.65 (5.08-6.74)	103.25 (36.00-183.40)	1.77 (1.00-5.20)
P3	Riparian Buffer	64.31 (24.99-87.48)	20.57 (12.70-24.10)	220.00 (173.00-278.00)	4.81 (4.60-6.90)	106.35 (-32.30-228.00)	2.01 (0.95-6.70)
P5	Riparian Buffer	125.27 (33.83-145.69)	19.70 (12.10-22.30)	163.00 (58.00-319.00)	4.95 (4.75-6.88)	118.80 (51.00-287.10)	1.65 (0.46-3.30)
P6	Spray Field	103.02	19.60	400.00	6.66	-109.80	0.95

		(70.71-233.17)	(13.40-26.00)	(269.00-1568.00)	(6.50-6.81)	(-136.00- -83.60)	(0.90-1.00)
P7	Spray Field	81.99 (74.98-91.44)	21.40 (13.50-25.80)	690.50 (651.00-804.00)	6.35 (5.30-6.93)	-71.05 (-149.00-69.30)	0.65 (0.01-1.13)
P8	Spray Field	60.20 (27.43-88.39)	21.90 (12.90-25.70)	353.50 (307.00-439.00)	5.47 (4.78-6.73)	107.20 (15.70-194.70)	2.32 (0.33-5.97)
P9	Spray Field	78.18 (36.27-167.94)	21.26 (13.00-26.10)	525.50 (370.00-546.00)	5.15 (4.57-6.90)	94.60 (-91.00-226.00)	1.71 (0.03-2.88)
P10	Spray Field	92.66 (3.35-145.69[a])	21.94 (12.70-26.80)	383.35 (307.00-609.00)	4.73 (4.56-5.98)	175.25 (82.00-211.60)	1.42 (0.80-2.44)
P11	Spray Field	125.27 (57.30-143.87)	22.40 (13.30-27.70)	441.00 (192.00-487.00)	6.38 (5.60-6.73)	-92.00 (-153.00-56.60)	1.00 (0.44-2.30)

[a]= DTW measured in July 2023, all other sampling events were dry thus DTW was > 232 cm (BG) and >145.87 (P10) (total piezometer depth).

3.5 Nitrogen Concentration in Surface Water

Concentrations of TDN were elevated in streams that drained the farm. The median TDN concentration of the pooled farm streams (SU, SD, EC-U, EC-D) was 25.41 mg/L, and TDN concentrations ranged from 3.84 to 82.56 mg/L. This median value was approximately 2 and 20 times greater than nitrogen concentrations in the DW (15.17 mg/L) and UP (1.25 mg/L) sampling locations, respectively. These differences were statistically significant ($p < 0.01$). The highest TDN concentrations among all surface water sampling locations were recorded at the Seep, while the lowest concentration was observed at the UP location, which was unaffected by wastewater irrigation (Table 3.7; Fig. 3.3). The median TDN concentration in SU (76.70 mg/L) was greater than SD (60.14 mg/L), but this difference was not statistically significant ($p = 0.25$). Although the median TDN concentration for the farm streams (SU, SD, EC-U, and EC-D), UP, and DW was statistically significant ($p < 0.01$). Median TDN concentrations at SU and SD were approximately 3 to 60 times greater than median concentrations observed in other streams. Median TDN concentrations in East Creek were 12.75 mg/L and 17.18 mg/L for EC-U and EC-D, respectively. Like the Seep, nitrogen concentrations did not significantly differ ($p = 0.5$) between EC-U and EC-D. Median TDN at EC-D was greater than UP and DW, although EC-U was only greater than UP (Fig. 3.3; Table 3.7). Groundwater discharge from 2 SFs (1 north of the Seep and another that the Seep dissects) likely discharges to the Seep. Collectively, the area of these SFs is 12.74 ha, which is approximately 1.32 times greater than the SF that East Creek dissects. Thus, nitrogen loads to the Seep are likely greater than those to East Creek, which would explain the difference in TDN concentrations. Both the Seep and East Creek eventually discharged to Horse Pen Branch and likely contributed to the elevated stream TDN concentrations observed DW of the farm. The median TDN concentration DW of the farm (15.17 mg/L) was 12 times higher than the concentration UP (1.25 mg/L) of the farm, and the difference was statistically significant (p = 0.03).

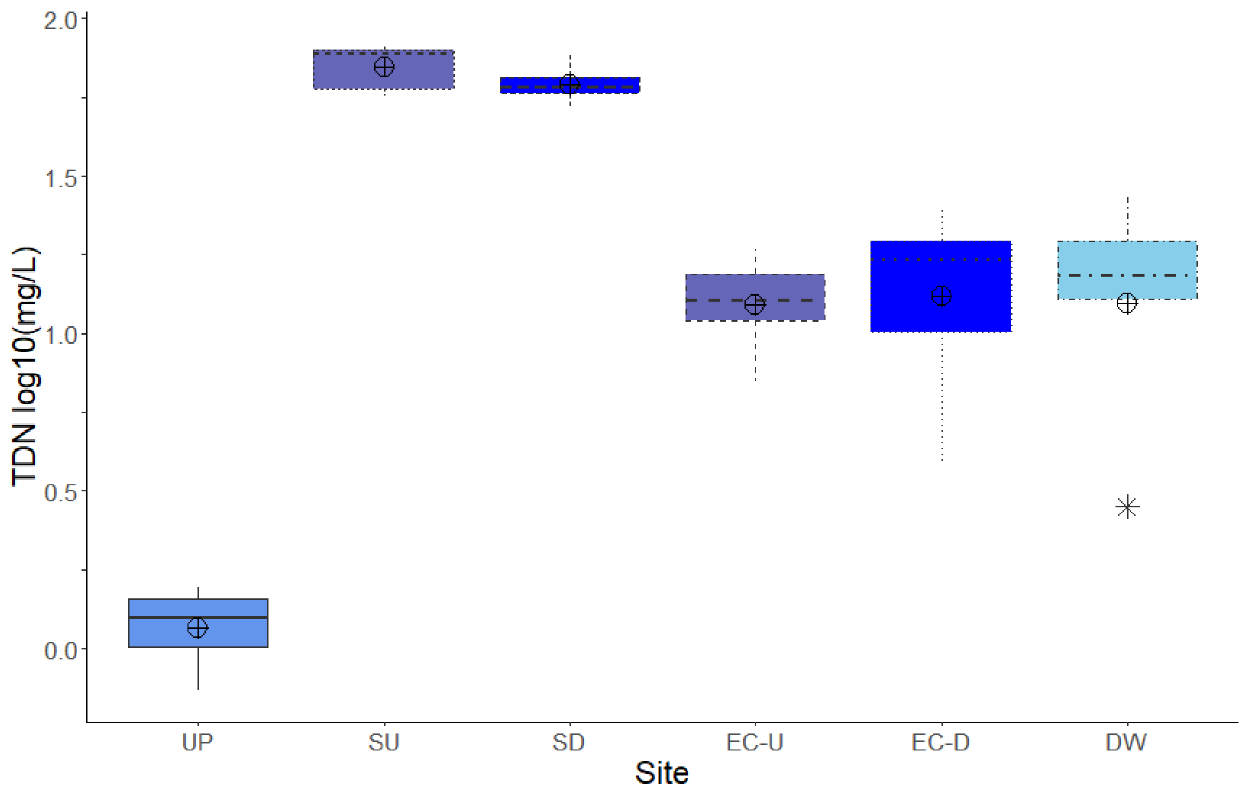

Figure 3.3: Boxplot showing concentrations of total dissolved nitrogen (TDN) for Upstream (UP), Seep Up (SU), Seep Down (SD), East Creek Down (EC-D), East Creek Up (EC-U), and Downstream (DW).

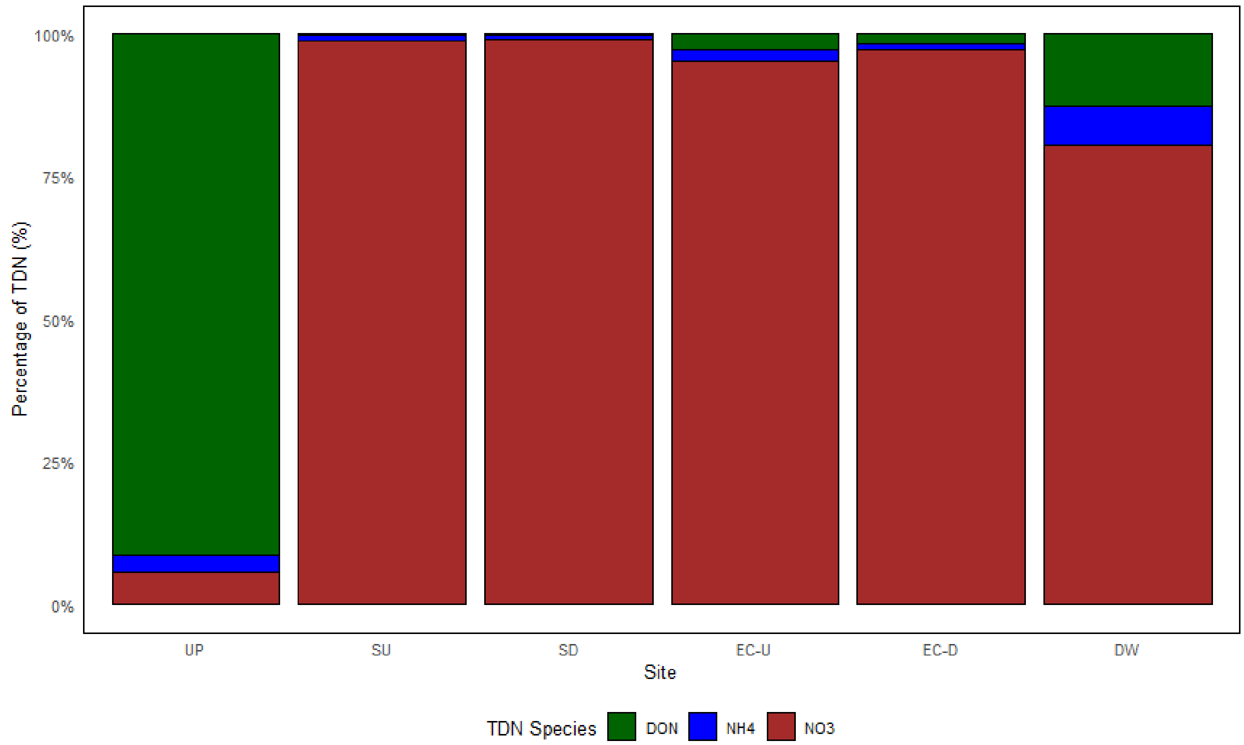

Figure 3.4: A chart showing the percentage of nitrogen species which includes dissolved organic nitrogen (DON), ammonium (NH_4^+), and nitrate (NO_3^-) in the surface water sampling locations such as the Upstream (UP), Seep Up (SU), Seep Down (SD), East Creek Up (EC-U), East Creek Down (EC-D), and Downstream (DW).

Table 3.7: Summary of UP= Upstream, SU= Seep Up, SD= Seep Down, East Creek Up= EC-U, East Creek Down= EC-D, and Downstream= DW showing median and range (in parentheses) concentrations for NH_4^+= ammonium, NO_3^-= nitrate, TDN= total dissolved nitrogen, and DOC= dissolved organic carbon.

Site	NH_4^+	NO_3^-	DON	TDN	DOC
UP	0.04 (0.03-0.44)	0.07 (0.04-0.3)	1.17 (<0.01[a]-1.49)	1.25 (0.74-1.56)	8.31 (5.96-13.44)
SU	0.27 (<0.01[a]-8.89)	75.80 (55.65-81.99)	<0.01[a] (<0.01[a]-4.10)	76.70 (56.77-82.71)	2.90 (2.35-6.54)
SD	0.48 (0.10-0.83)	59.53 (52.39-68.61)	<0.01[a] (<0.01[a]-3.91)	60.14 (52.66-76.62)	5.13 (2.17-8.94)
EC-U	0.24 (0.10-0.83)	12.15 (6.17-18.13)	<0.01[a] (<0.01[a]-1.43)	12.75 (7.00-18.48)	6.53 (2.85-13.65)
EC-D	0.17 (0.09-2.57)	16.73 (1.28-25.32)	<0.01[a] (<0.01[a]-1.83)	17.18 (3.84-25.42)	18.41 (3.77-72.81)
DW	1.05 (0.21-27.26)	12.22 (0.31-19.34)	0.3 (<0.01[a]-2.07)	15.17 (2.82-27.56)	22.67 (5.58-37.55)

[a]= DON and NH_4^+ were incalculable due to dissolved inorganic species representing

100% of TDN

Past studies have consistently identified that CAFOs are major contributors to elevated nutrient concentrations in surface waters, particularly for TN and its inorganic forms (NH_4^+ and NO_3^-) (Karr et al. 2001; Harden, 2015; Mallin et al. 2004; Karr et al. 2001; Harden and Spruill, 2008; Bauwe and Lennartz, 2024; Ma et al. 2023; Aneja et al. 2024). These studies have documented higher nitrogen levels in watersheds influenced by CAFOs, especially during and following storm events. Additionally, tile drainage systems further facilitate NO_3^- movement into streams (Karr et al. 2001; Harden, 2015; Mallin et al. 2004; Karr et al. 2001; Harden and Spruill, 2008; Ma et al 2023; Bauwe and Lennartz, 2024; Ma et al. 2023). Comparative assessments of nitrogen concentrations across various studies show a marked difference between background and CAFO-influenced surface water. Mallin et al. (2015) and Harden (2015) reported TN concentrations in surface waters impacted by CAFO ranged from 0.11 to 46.7 mg/L (median: 8.6 mg/L) and 0.20 to 17.0 mg/L (median: 1.5 mg/L), respectively. These studies also reported TDN concentrations that were substantially greater than background concentrations, which Harden (2015) noted as ranging from 1.0 to 1.3 mg/L. These studies mentioned above had a low nitrogen concentration compared to the current study. The range of TDN concentrations observed in the current study (DW: 2.82 to 27.56 mg/L) was comparable to the range reported in the studies mentioned above, indicating that both studies captured similar nitrogen conditions in streams near a CAFO. However, the elevated median TDN concentration observed in this study was likely due to the proximity of the sampling location to the SFs, where nutrient loading is typically higher. Richardson (2023) also recorded an elevated range of 2.28 to 40.69 mg/L. In contrast, Harden (2015) reported lower median TDN concentrations (1.5 mg/L) at downstream locations farther from direct wastewater influence, although the overall range (0.20 to 17.0 mg/L) still encompassed values similar to those in this study

In the current study, the stream sampling locations were all within 100 m of SFs, which explains the tendency for higher TDN concentrations (especially for streams dissecting SFs). However, Mallin et al. (2015) and Harden (2015) sampled streams that were approximately 3 km downstream from the nearest CAFO. These studies indicate that wastewater-derived TDN can substantially influence water quality in receiving waters. However, it is possible that elevated nitrogen concentrations could be attenuated depending on the DW aquatic habitat. In the current study, the DW location drains to an extensive wetland that is within 0.5 km of the sampling point. Thus, there is potential for biogeochemical processes to reduce inorganic nitrogen; however, additional work is necessary to conduct a longitudinal survey to evaluate water quality as the distance from the farm increases.

3.6 Discharge and Nutrient Mass Transport in Surface Water

Stream discharge was variable at all sites, and prolonged drought conditions occurred during the study. Median discharge at SU, SD, EC-U, and EC-D were 151.75 L/min, 128.89 L/min, 11.29 L/min, and 163.37 L/min, respectively (Table 3.8). Generally, discharge ranged from 0 (dry) to 485.00 L/min. When pooling the farm streams, the median discharge was 84 L/min, which was approximately 2 and 84 times higher than the median value at the DW and UP locations, respectively (Table 3.8). Differences in discharge between the farm streams and UP were statistically significant ($p = 0.04$), but not between the farm streams and DW ($p = 0.66$). Median discharge decreased by about 15% between SU and SD, which was congruent with findings by Richardson

(2023). The opposite trend was observed at East Creek, where discharge increased by more than an order of magnitude between EC-U and EC-D (Table 3.8). This difference likely occurred due to the differences in land cover (Fig. 2.1). There was a pond and a forest immediately upstream of EC-U, which likely influenced stream discharge. The forest likely retained most of the discharge from the pond due to drought. Despite these tendencies, discharge did not yield any statistically significant differences between comparison groups ($p = 0.1$). The drought conditions strongly influenced stream discharge trends at the UP and DW sampling locations. Median discharge at the DW location was 54 L/min, whereas it was 0 L/min UP of the farm. Thus, upstream reaches of Horse Pen Branch tended to be dry. In a previous study at the same farm, Richardson (2023) reported median discharge values of approximately 1,000 and 2,150 L/min at UP and DW, respectively, and these differences were statistically significant ($p < 0.01$). These findings suggest that swine wastewater can be a significant source of water for stream recharge. Interestingly, the SU and SD median discharge values were only slightly higher at 240 and 150 L/min (Richardson 2023), respectively. Wastewater irrigation likely recharged the Seep, insulating the stream from drought impacts. A similar phenomenon was also likely for the East Creek; however, Richardson (2023) did not evaluate stream discharge at this site. Additionally, median discharge at SD and EC-D typically exceeded DW (even after removing dry sampling days), which was unexpected considering that SD and EC-D drain to the DW sampling location and DW serves as the watershed outlet. Thus, prolonged dry conditions likely resulted in reduced discharge due to increased evapotranspiration. Past studies reported that prolonged drought reduced soil moisture and streamflow due to evapotranspiration and high temperature. This may have resulted in intermittent or zero discharge, particularly at the UP and DW locations (Lake, 2003; Vicente-Serrano et al. 2014).

Table 3.8: Median and range (in parentheses) for discharge= Q, mass transport, and total dissolved nitrogen= TDN in surface water sample locations such as Upstream (UP), Seep Up (SU), Seep Down= SD, East Creek Down= EC-D, East Creek Up= EC-U, and Downstream= DW.

Site	TDN mg/L	Discharge (Q) L/min	Mass Transport Kg-N/day
UP	1.25 (0.74-1.56)	0.00 (0.00-282.39)	0.00 (0.00-0.56)
SU	76.70 (56.77-82.71)	151.75 (30.00-284.00)	9.97 (2.47-25.45)
SD	60.14 (7.00-18.48)	128.89 (31.60-215.74)	10.72 (2.82-16.36)
EC-U	12.75 (7.00-18.48)	11.29 (0.00-308.68)	0.20 (0.00-7.03)
EC-D	17.18 (3.84-25.42)	163.37 (2.69-370.09)	3.71 (0.03-13.54)
DW	15.17 (2.82-27.56)	54.44 (0.00-485.97)	1.01 (0.00-4.66)

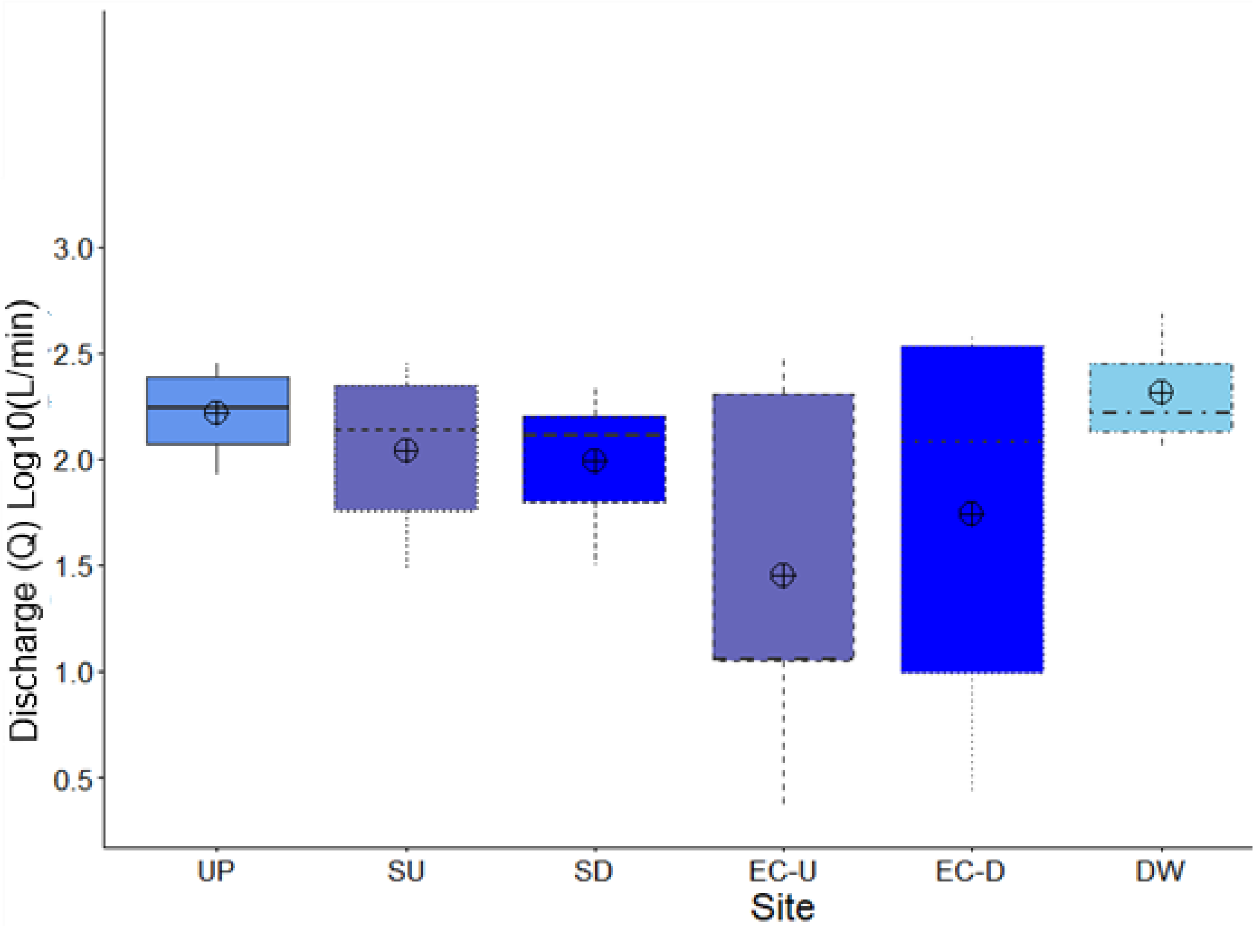

Figure 3.5: Boxplot showing discharge (Q) for upstream (UP), Seep Up (SU), Seep Down (SD), East Creek Up (EC-U), East Creek Down (EC-D), and Downstream (DW).

Streams draining the farm had the greatest nitrogen mass transport relative to UP and DW. The overall median TDN export from these streams was 6.0 kg-N/day, which was approximately 6 and 19 times greater than the DW and UP locations, respectively (Table 3.8; Fig. 3.5). However, the masses did not significantly differ ($p = 0.06$). This likely occurred because TDN export from EC-U was much lower (median: 0.2 kg-N/day) than the other farm streams, where median TDN mass export exceeded 3.5 kg-N/day. The large discrepancy in TDN mass export between these streams occurred due to discharge differences discussed previously. If EC-U is excluded from the analysis, then the difference between the farm stream group, the UP, and DW was statistically significant ($p = 0.01$). Mass loadings were greatest at SU and SD, with median values of approximately 10 and 11 kg-N/day, respectively (Table 3.8). TDN mass export did not significantly differ between SU and SD ($p = 1$). However, Richardson 2023 indicated elevated median TDN at the DW location. Median TDN export at EC-D was approximately 4 kg-N/day, indicating that the East Creek is another potentially significant TDN source to Horse Pen Branch. The median TDN export at DW was approximately 1 kg-N/day, which was about 3 times greater than the UP of the farm (0.3 kg-N/day). This difference was not statistically significant ($p = 1$). Mass transport DW of the farm was highly variable, ranging from 0 to 4.7 kg-N/day. Since both UP and DW were substantially influenced by drought, these streams were supplemented with data from Richardson (2023), which included 13 months of surface water sampling. Median mass export of TDN was approximately 14 and 1 kg-N/day for DW and UP, respectively, and this difference was statistically significant ($p < 0.01$). These findings indicated that irrigation of swine wastewater was a source of nitrogen to Horse Pen Branch.

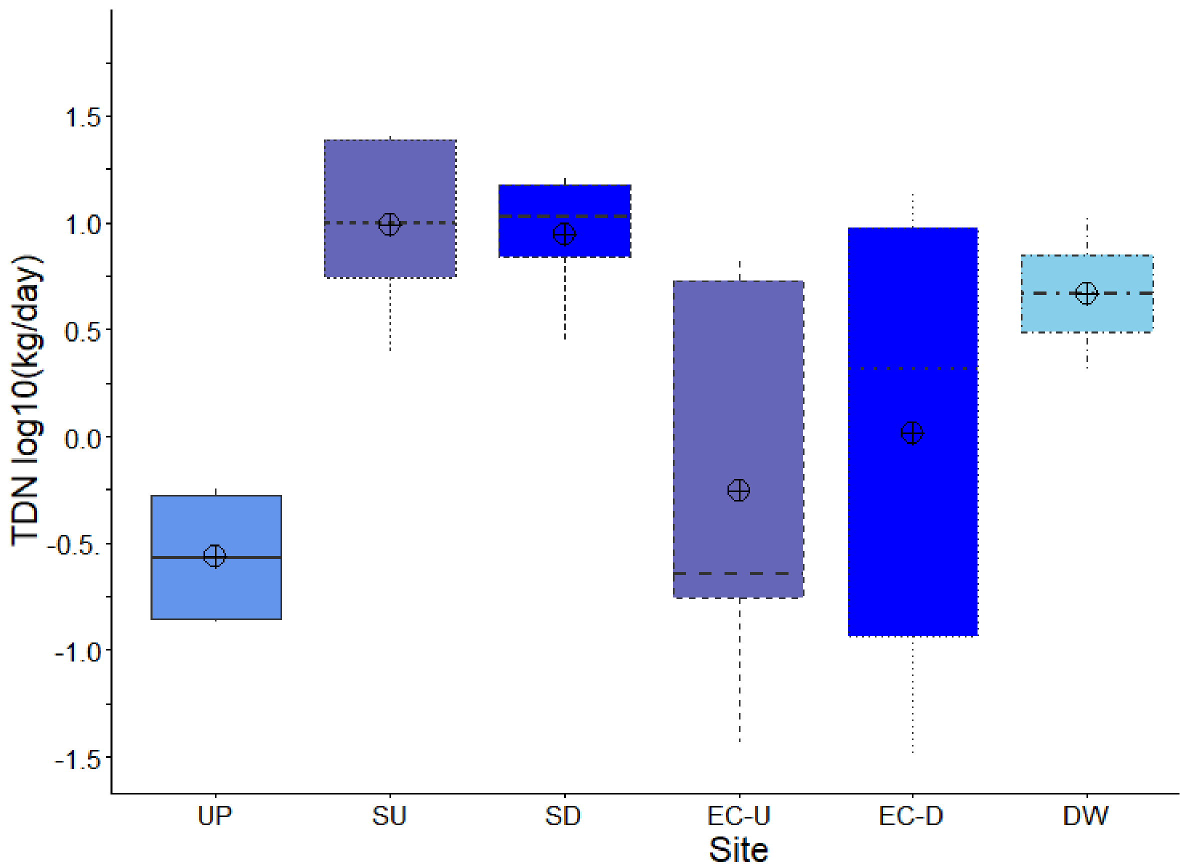

Figure 3.6: Boxplot showing mass transport for Upstream (UP), Seep Up (SU), Seep Down (SD), East Creek Down (EC-D), East Creek Up (EC-U), and Downstream (DW).

The current study also found that drought conditions influenced nitrogen transport from swine CAFOs to receiving water. Data from the NC State Climate Office (NCSCO) indicates that annual precipitation between 2019 and 2020 was 115.12 cm and 169 cm, respectively. Richardson (2023) reported a total precipitation of 194 cm for the previous study. The current study was carried out between 2023 and 2024, which recorded an annual precipitation of 112.12 cm and 134.65 cm, respectively. The current study also reported a total precipitation of 44.81 cm during the duration of the study. These findings reveal a decrease in annual precipitation, suggesting an impact associated with drought conditions in this study. The reduced rainfall rates leading up to and during the current study's sampling period likely reduced stream discharge, thereby reducing nutrient mass transport. The monthly review of the precipitation data (from NCSCO) indicated that the precipitation values for months in which this current study was carried out were among the lowest (Table 3.9; Fig. 3.7)

Table 3.9: A table showing precipitation and evapotranspiration measured in cm from January 2023 to September 2024.

Date	Precipitation cm	Evapotranspiration (ET) cm
1-Jan-23	8.3058	4.75234
1-Feb-23	7.0866	6.44398
1-Mar-23	6.2992	8.88238
1-Apr-23	16.129	10.76452
1-May-23	5.715	13.25626
1-Jun-23	12.2682	14.3383
1-Jul-23	16.4084	15.37462
1-Aug-23	12.2174	14.69644
1-Sep-23	7.5184	10.53592
1-Oct-23	3.1242	8.08482
1-Nov-23	3.175	5.30606
1-Dec-23	13.8684	3.95986
1-Jan-24	13.6906	4.79298
1-Feb-24	2.3622	6.26872
1-Mar-24	10.0584	9.0678
1-Apr-24	2.6924	12.02182
1-May-24	13.4112	12.83716
1-Jun-24	6.858	16.18996
1-Jul-24	35.9918	14.02842
1-Aug-24	14.1224	12.71524
1-Sep-24	27.1018	8.98144

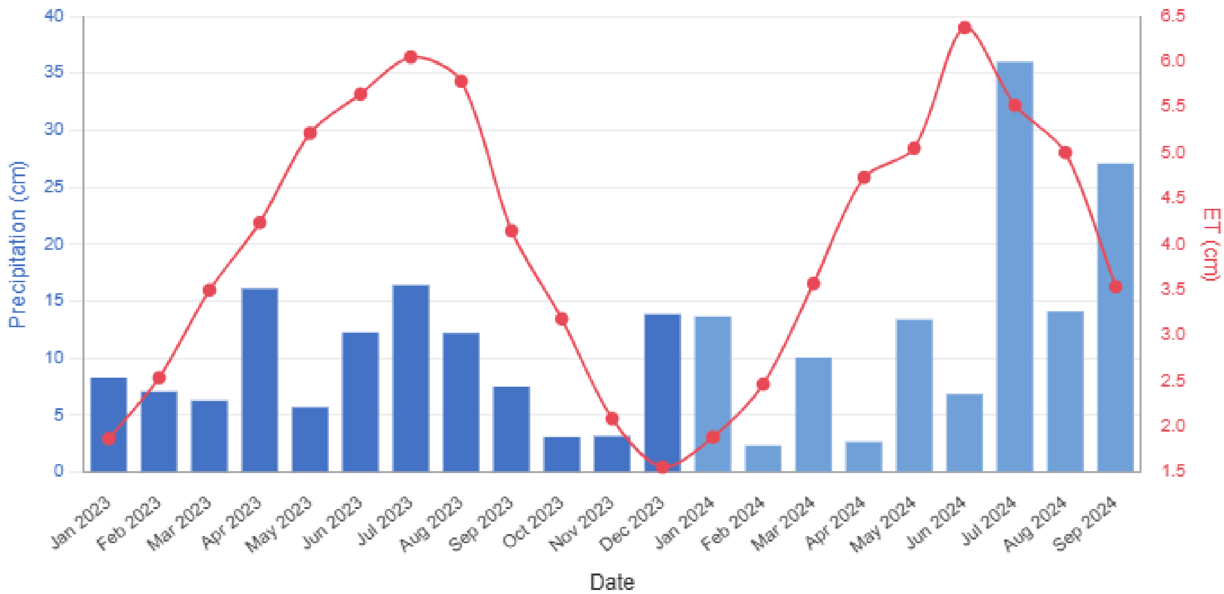

Figure 3.7: A chart showing precipitation and evapotranspiration between January 2023 and September 2024.

Richardson (2023) found that the UP and DW sampling locations transported a median TDN mass of 0.9 and 21.4 kg-N/day, respectively. These masses were between 3 and 21 times greater than the masses observed in the current study, UP and DW of the farm, respectively. Richardson (2023) estimated that the annual area-normalized export of TDN was 29.5 kg-N/ha/yr at the DW location. This estimate was nearly 5 times greater than a study by Haggard et al. (2003), who reported an average TN export of 6.0 Kg-N/ha/year from streams draining pastures that are irrigated by poultry wastewater. This value was approximately 4 to 19 times more than the current location (1.52 kg-N/ha/year). This further suggests that the drought conditions in this current study may have an impact on nitrogen exports. When the maximum mass transport (4.66 kg-N/day; Table 3.8) value for the DW location was adopted, the annual export estimate was approximately 7 kg-N/ha/year. This maximum export estimate was in the range reported by Haggard et al. (2003) and Richardson (2023), further suggesting that drought conditions may have affected the mass export from DW.

3.7 Nitrogen Mass Budget

The nitrogen mass budget suggested that the farm was highly effective at attenuating TDN masses. There are 5 SFs on the farm where wastewater was occasionally used for irrigation from May to October. The average wastewater application rate across all 5 SFs was 76,106.61 L/day. The total volume of wastewater applied to the SFs was 7,686,767 L in a year. Assuming the median TDN in the LG was representative, the daily TDN mass load to SFs was 125.65 kg-N, and the total annual load was 12,691 kg-N. The discharge DW of the farm was ~ 54 L/day and 19,870.6 L/year. Median TDN mass export from DW was approximately 1 kg-N/day (Table 3.10). The estimated annual export was approximately 369 kg-N/year (Table 3.10). Thus, there was a >99% reduction between the annual TDN load to SFs relative to the annual TDN export from the DW sampling location. These results indicate that the studied CAFO was highly effective in attenuating TDN mass transport. However, the nutrient mass budget was likely influenced by drought conditions that substantially decreased stream discharge and watershed nutrient transport. In a previous study at the same farm during normal conditions, Richardson (2023) estimated an annual TDN export from DW sampling location of 7,818.4 kg-N, which was more than 20 times greater than the current study. Some of this estimate also includes non-wastewater-derived TDN originating from soil organic matter, other animal waste, and plant decay. Assuming that TDN export at UP represents these contributions (median: 316.7 kg-N/yr), then the CAFO contributes

7,501.7 kg-N/yr to the downstream (Richardson 2023). Thus, there was a 40% difference between watershed TDN export at DW compared to the TDN load from wastewater application. If Richardson (2023) data are representative of watershed TDN exports during normal conditions, then these findings suggest that CAFOs are less effective at reducing TDN masses and can be significant TDN sources to downstream water resources.

Table 3.10: Daily and annual discharge and mass budget estimate in the farm location. The number of spraying and sampling events for the period was also indicated.

Location	Spraying/sampling events	Discharge Budget (L)		Mass Budget (kg)	
		Day	Year	Day	Year
SF 5	21	109678.5	2303248.2	181.08	3802.71
SF 4	19	61998.3	1177967.7	102.36	1944.85
SF 3	27	66973.47	1808283.75	110.58	2985.51
SF 2	16	68456.46	1095303.3	113.03	1808.37
SF 1	18	72331.35	1301964.3	119.42	2149.57
DW	6	54.44	19870.6	1.01	368.65

3.8 Physicochemical Parameters in Surface Water

Like groundwater physicochemical data, streams downgradient from SFs had elevated SC relative to UP (Table 3.10). The Seep (SU and SD) had the highest SC (range: 710 to 969 µS/cm) compared to the other surface water locations (Table 3.10). Median SC at the Seep (SU and SD) and East Creek (EC-U and EC-D) locations was approximately 5 to 19 times higher than SC recorded at the UP location. The East Creek (EC-U and EC-D) location also recorded elevated SC concentration with a range of 200 to 762 µS/cm (Table 3.10). The SC at the DW (418.00 µS/cm) was approximately 8 times greater than the UP 48.15 µS/cm) location (Table 3.10). The elevated SC values suggest that wastewater influenced water quality in the streams draining the farm and the DW sampling location of Horse Pen Branch (Michalopoulos et al. 2014; Harden 2015). Past studies indicate that watersheds subjected to irrigation from CAFOs generally demonstrate higher SC levels compared to the ambient background SC (Harden, 2015; Richardson, 2023). The elevated SC suggests the influence of wastewater irrigation (Harden, 2015).

Streams contained elevated ORP and DO values relative to groundwater data, which was expected. The median ORP (330.02 mV) and DO (6.45 mg/L) were highest at the UP location, suggesting that this was an aerobic environment supporting oxidation. DO > about 1.0 mg/L is considered a threshold for effective aerobic microbial oxidation (Li and Bishop, 2002). DO concentrations tended to be low (< 4 mg/L) in most of the streams adjacent to or downgradient of SFs. Drought may have also influenced this since stream discharge was low, and there was a tendency for streams to stagnate, which could have inhibited oxygen mixing with the water column. Median temperature ranged from approximately 17 to 20°C. The lowest temperature tended to occur UP of the farm, which was expected due to the difference in tree cover. The UP sampling locations are within extensive forests whose canopy likely shaded the stream.

Measured pH values across the surface waters were acidic to near neutral, ranging from approximately 4.2 to 6.7, with the lowest median (4.19) at the UP location and the highest (6.72) at EC-D. The generally low pH values, particularly upstream and within seep areas, may reflect the influence of organic-rich soils, atmospheric deposition, and limited buffering capacity in the watershed. In contrast, slightly higher pH values at the East Creek (EC-U and EC-D)and DW locations indicate a buffer effect as wastewater effluent is diluted along the flow path. Turbidity showed substantial spatial variability, with median values ranging from 0.05 to 52.6 NTU. The greatest turbidity was recorded at EC-U and EC-D, suggesting higher suspended solids and particulate transport near the effluent-receiving reaches, while the lowest turbidity at the Seep locations points to limited surface disturbance. The high turbidity patterns may have resulted in the low discharge values recorded at the East Creek location.

Table 3.91: Physical and chemical parameters collected at the farm through the sample events for upstream= UP, Seep Up= SU, Seep Down= SD, East Creek Down= EC-D, East Creek Up= EC-U, and downstream= DW. Median (range) of pH, Temp= temperature, specific conductivity= SC, dissolved oxygen= DO, turbidity= NTU, and oxidation reduction potential= ORP.

Site	Temp (°C)	SC (uS/cm)	Turb (NTU)	pH	ORP (mV)	DO (mg/L)
UP	17.45 (10.30-24.45)	48.15 (40.80-89.00)	5.12 (3.33-80.00)	4.19 (3.60-5.82)	330 (164.00-384.20)	6.45 (5.00-9.10)
SU	18.80 (10.93-28.48)	919.00 (846.00-969.00)	0.05 (0.00-5.3)	4.91 (4.82-5.13)	294.50 (278.00-421.00)	5.72 (3.80-8.50)
SD	18.93 (13.12-26.02)	870 (710.00-930.00)	20.35 (1.40-112.00)	6.10 (4.64-6.59)	281.75 (198.00-326.90)	3.90 (1.44-12.50)
EC-U	19.57 (10.42-26.20)	264.50 (200.00-379.00)	52.60 (5.10-689.00)	6.07 (5.41-7.01)	199.05 (146.60-218.00)	6.54 (4.70-9.14)
EC-D	20.05 (10.38-26.70)	507.50 (348.00-762.00)	52.60 (5.10-689.00)	6.72 (6.00-7.01)	147.20 (-8.00-188.40)	3.51 (0.01-10.14)
DW	20.27 (11.20-26.00)	418.00 (255.00-481.00)	26.10 (9.50-850.00)	6.57 (5.92-7.10)	158.00 (73.90-210.50)	3.67 (1.45-9.70

3.9 Fate and Transport of Nitrogen at the Farm

3.9.1 Fate and Transport of Nitrogen in Groundwater

Groundwater nitrogen concentrations tended to decrease within RB (Table 3.1; Fig. 3.1). Additionally, estimates of TDN mass reduction also increased as distance downgradient from SFs increased (Table 3.3). These data suggest that biogeochemical processes in the soil and groundwater influenced nitrogen treatment. Median TDN concentration reduced by >99% between the LG and SF piezometers. There was an additional 83% reduction in TDN concentrations between SF and RB piezometers. Mass reduction estimates were more variable in SF piezometers (approximately 8 to 96%) but were high (≥ 95%) at the RB. In addition to concentration and mass reductions, nitrogen speciation also shifted relative to wastewater. There was no clear trend in groundwater nitrogen speciation across all piezometers. Most SF piezometers consist primarily of NH_4^+ (P1, P2, P6, P7, P9, and P11), but the percentage of NH_4^+ comprising TDN reduced from >99% in wastewater to approximately 70% in groundwater. NO_3^- was dominant in the remaining piezometers, accounting for 49% and 65% of TDN in P8 and P10, respectively. Nitrogen speciation trends once again shifted between SF and RB piezometers. DON was the dominant species of TDN (45.3%) in groundwater beneath RBs, followed by 32.05% NO_3^- and 22.7% NH_4^+, respectively (Table 3.1). Two possible explanations for this shift could be due to natural sources of DON and/or reduction of dissolved inorganic nitrogen (DIN) species. Decomposition of leaves or other organic matter and animal waste may be a source of DON to groundwater underlying RBs (Panno et al. 2006). NO_3^- and NH_4^+ are labile forms of TDN; thus, biogeochemical processes preferentially reduce DIN relative to DON. Several nitrogen transformation processes may have occurred to reduce nitrogen, including volatilization, adsorption, denitrification, dilution, anaerobic ammonium oxidation, and plant and microbial uptake.

Some combination of these processes likely occurred in the vadose zone between the ground surface and water table. In the subsoils of the vadose zone, the most likely processes included adsorption, plant and microbial uptake, volatilization, and nitrification (not a net reduction in TDN). Adsorption occurs within aerobic soils and can reduce NH_4^+ concentrations. Soil surfaces tend to be negatively charged, which attracts the NH_4^+ cation (if there are sufficient cation exchange sites available). While adsorption reduces bioavailability of NH_4^+, sorbed NH_4^+ cations can be permanently removed via nitrification or plant and/or microbial uptake (Ham and DeSutter 2000). However, if the adsorption capacity of the soil is saturated, groundwater contamination may occur (Kadyampakeni et al. 2018). In the current study, soil properties and mineralogy (Appendix A) revealed elevated CEC and clay content down the soil horizon. This suggests that adsorption can enhance NH_4^+ immobilization due to soil mineralogy. However, when the soil is saturated, groundwater may be prone to NH_4^+ contamination. Plant and microbial uptake are other possible removal mechanisms that likely occurred within the SFs. CAFOs discharge swine wastewater to SFs to increase storage capacity in the LG and to fertilize crops growing in SFs. Plants and microbes utilize NH_4^+ and NO_3^- as nutrient sources to enhance growth and cell metabolic activities (Zhu et al. 2021; Xu et al. 2012). Plants take up NH_4^+ and NO_3^- through their root systems because nitrogen is a vital component of amino acids, nucleic acids, and chlorophyll, which are required for protein synthesis, photosynthesis, and overall growth (Xu et al. 2012). Microbes, in turn, assimilate NH_4^+ and NO_3^- to produce biomolecules such as enzymes and nucleotides needed for cell maintenance and proliferation (Kuypers et al. 2018).

Volatilization can also reduce NH_4^+ by converting it to gaseous ammonia if soil pH and temperature are supportive. As temperature rises, the fraction of total ammoniacal nitrogen present as free ammonia increases at a neutral or alkaline pH (>7), increasing potential for volatilization (Sommer and Hutching 2001; Rochette et al. 2013). Nitrification can also reduce NH_4^+ by converting it to NO_3^- through a two-phase biological process where oxidizing bacteria (e.g., *Nitrosomonas*) convert NH_4^+ to nitrite and nitrite-oxidizing bacteria (e.g., *Nitrobacter*) convert nitrite to NO_3^-. The main environmental factors controlling nitrification are pH (6.5 to 8.5) and an aerobic environment (Whalen and DeBerardinis, 2007; Kowalchuk and Stephen, 2001; Mallin and Cahoon, 2003). While direct soil pH was not measured in the current study, using groundwater pH as a proxy for soil pH indicated that P11, P6, and P7 had the highest pH (median: 6.4 to 6.7), but values were still slightly acidic. Groundwater in all other piezometers was more acidic, with a pH < 5. Both volatilization and nitrification result in protonation, which decreases pH. These processes likely explain why the median pH in SF groundwater was about 1.8 to 3.8 pH units lower than swine wastewater (Table 3.5). Dilution (mixing swine wastewater with BG) may also influence pH, although most SF piezometers contained a pH that was also lower than BG. It is difficult to differentiate volatilization and nitrification without additional research. Since nitrification increases NO_3^- concentration (Whalen and DeBerardinis, 2007; Ham and DeSutter, 2000), evaluating NO_3^- concentrations can further understanding. Relative to wastewater, NO_3^- concentrations increased in all groundwater piezometers (Table 3.5). However, NO_3^-

concentrations were lower than expected in most piezometers. Cocco et al. (2018) found that nitrification may be limited when DTW is ≤ 60 cm, whereas nearly complete nitrification can occur at ≥ 60 to 120 cm (Cocco et al. 2018; Humphrey et al 2013). In the current study, all piezometers (apart from P11) had a median DTW that was > 60 cm (Table 3.6), suggesting that there was sufficient vadose zone to support nitrification. However, elevated NH_4^+ concentrations were found in most SF piezometers. One reason for this phenomenon could be nitrification inhibition from the wastewater application rate. If wastewater application rates exceed the assimilatory capacity of the SFs, it is possible that NH_4^+ could leach into the groundwater, particularly if cation exchange sites are filled (Kadyampakeni et al. 2018). Wastewater application rates were discussed in greater detail in Section 3.7. Another possible explanation could be due to an LG or irrigation system malfunction, resulting in an unexpected release of wastewater. If raw wastewater is directly discharged to surficial aquifer, nitrification will be inhibited since this process requires an aerobic environment (Iverson et al. 2024; Humphrey et al, 2013). More research is needed to evaluate this phenomenon. Water levels in the LG could be monitored using pressure transducers and manual measurements. Comparing these data to application records would allow for the calculation of a water budget and could indicate if the LG leaked.

Treatment processes for NH_4^+ are limited to the surficial aquifer. Adsorption is inhibited since exchange sites are flooded, and hypoxic/anoxic conditions within groundwater may impede nitrification. Plant uptake may aid nitrogen removal within SFs; however, its effectiveness is constrained by the depth of groundwater. Bermudagrass (*Cynodon dactylon*), which dominates the SF, typically concentrates its roots within the upper 0 to

40 cm of soil, with only limited penetration to 15 to 20 cm under ideal conditions (Kiniry et al. 2007; Fuentealba et al. 2015). Since the DTW was typically > 60 cm, plant uptake of NH_4^+ from the surficial aquifer may be limited within the SFs. However, if groundwater migrates through an RB zone with a dense population of trees and herbaceous plants, it is possible that their deeper root zones may reduce TDN concentrations via plant uptake of NH_4^+ and NO_3^-. Past studies reported that more than 50% of the inorganic nitrogen (NO_3^- and NH_4^+) applied to SF were assimilated by plants (Whalen and DeBerardinis 2007; NC State Extension 2017). While RB locations were reported to attenuate 60-95% of nitrogen and other nutrients (Hefting et al. 2004; Messer et al. 2012). Another possible reduction of NH_4^+ that could be occurring within SF and RB groundwater is anaerobic ammonium oxidation (anammox), a microbial process that couples NH_4^+ oxidation with NO_2^- reduction to form N_2 under strictly anoxic and low-redox conditions (Canfield et al. 2010). While nitrification was limited in the current study, NO_3^- in groundwater that reaches RB can be removed via denitrification. Denitrification occurs when denitrifying microbes convert NO_3^- to dinitrogen gas (N_2) or nitrous oxide (N_2O) in the presence of carbon and anoxic conditions (Whalen and DeBerardinis, 2007; Rich et al, 2003; Mallin and Cahoon, 2003). Environmental conditions in all SF piezometers support denitrification (Table 3.2). Median DO and ORP values in groundwater beneath SFs were hypoxic/anoxic (median DO ≤ 2.32 mg/L), and ORP tended to fall within the optimal denitrification range -300 to +100mv (Hitchcock et al, 2020). Optimal denitrification generally occurs at a carbon to nitrogen (C/N) ratio of approximately 1.5 to 5 (Fang et al. 2018). This range provides adequate organic carbon to facilitate nitrate reduction without encouraging excessive anaerobic activity. Research

has demonstrated that nitrogen removal efficiency reaches its peak within this range (Fang et al. 2018; Mohan et al. 2016). Ratios below 3 can lead to carbon limitation, while those exceeding 8 to 10 may promote dissimilatory nitrate reduction to ammonium (DNRA) or methanogenesis. In SF groundwater, denitrification appeared to be nitrate-limited in most piezometers (P2, P6, P7, P11) (Table 3.2). Other locations appeared to be carbon-limited (P2, P10). Ratios of C/N are within ideal ranges for P9 (1.5) and P1 (3.8) to support denitrification, suggesting this may be a dominant removal mechanism. The elevated nitrogen concentration at P10 could indicate insufficient residence time and availability of carbon source to support more complete denitrification, but additional work is required to evaluate groundwater discharge in this SF. Environmental conditions in groundwater beneath the RBs also support that denitrification is possible. Median DO concentration in RB piezometers was approximately 2 mg/L or less. Median ORP values in RB groundwater were between 105 and 120 mV (Table 3.5; Table 3.6), which is supportive of denitrification (Hitchcock et al, 2020). C/N ratios in RB groundwater are near the optimal range for P5 (8.8) but are nitrate-limited for P3 (13.1). Groundwater quality suggests that denitrification likely occurred within RBs, and that it may be plausible for some SF piezometers.

3.9.2 Fate and Transport of Nitrogen in Surface Water

Surface waters adjacent to and downgradient from the SFs exhibited elevated nitrogen concentrations and masses compared to the UP location (Tables 3.11; Fig. 3.3). However, both nitrogen concentrations and loads decreased with distance from the farm, indicating attenuation along the flow path. The Seep location recorded the highest median TDN concentration (> 60 mg/L) and mass export (≥ 10 kgN /day). East Creek also had elevated TDN concentration (> 15 mg/L) and mass export (4 kgN /day). Streams draining the farm exhibited higher TDN than the DW (15 mg/L) location, suggesting that in-stream processes such as dilution and biological uptake contributed to TDN reduction.

Nitrogen speciation also shifted notably between groundwater and surface water. In groundwater, NH_4^+ was the dominant nitrogen form, whereas in surface waters NO_3^- predominated in all locations except UP (Table 3.7). This pattern indicates that nitrification likely occurred after groundwater discharged into surface waters, which contained greater DO concentration than groundwater (Table 3.5; Table 3.11). Median NO_3^- concentrations accounted for 80 to 99% of TDN in streams downgradient from the SFs, while NH_4^+ concentrations were typically < 1 mg/L (≤ 2 % of TDN). In contrast, UP was dominated by DON (> 90 % of TDN), likely derived from plant decay, wildlife inputs, or other background sources (Cerling et al. 2004). The compositional difference between UP and the other streams aligns with expectations, since swine wastewater is a known source of dissolved inorganic nitrogen (DIN), primarily as NO_3^- and NH_4^+ (Vanotti et al. 2018; Ham and DeSutter, 2000).

Surface waters demonstrated a higher median NO_3^- concentration compared to groundwater (Tables 3.1, 3.4), with NO_3^- accounting for over 80% of TDN. This finding supports the idea that active nitrification of NH_4^+ is a predominant transformation process (Mayer et al. 2007; Cheng and Basu, 2017), either in the vadose zone or after groundwater discharges to surface waters. While anammox could theoretically take place under hypoxic conditions (Canfield et al. 2010), the elevated concentrations of DO observed in surface waters likely render its contribution negligible. Among the sites studied, the Seep and East Creek locations recorded the highest concentrations of NO_3^-, whereas the UP and DW sites, both characterized by greater vegetation, exhibited lower levels of DIN.

High DO and ORP values (Table 3.11) suggest that aerobic microbial activity drives nitrification in surface waters. The dominance of DIN (> 90 % of TDN) across most sites indicates both microbial uptake and plant assimilation may also contribute to nitrogen removal (Saunders and Kalff, 2001; Cheng and Basu, 2017). Vegetation and well-defined riparian buffers, particularly along the Horse Pen Branch, likely enhance water retention, evapotranspiration, and infiltration, which reduce runoff and nutrient export (Vidon and Hill, 2004; Lake, 2003). These vegetated riparian zones, often characterized by an active hyporheic exchange (Spruill, 2020), are recognized hotspots for nitrogen attenuation and a resultant net loss. Past studies have reported TDN reduction of 49 to 90 % (Gregory et al. 1991; Tesoriero et al. 2005) and significant (> 98%) NO_3^- mass decreases (Harden and Spruill, 2008). The observed decline in NO_3^- and TDN between SD and DW locations suggests that in-stream and biogeochemical processing further reduce nitrogen export (Royer et al. 2004; Udy et al. 2006). Although high DO and ORP values in surface waters suggest that denitrification within the water column is limited, the process may still occur in localized anoxic microzones, particularly in the hyporheic zone or during periods of low streamflow. Reduced discharge can limit oxygen exchange and increase residence time, promoting denitrification if organic carbon is available (Mulholland et al. 2008). Additionally, nitrate-rich water may infiltrate into the hyporheic zone, where interaction between surface and subsurface flows can create redox gradients that favor microbial denitrification (Harvey et al. 2013).

Results in the current study showed that the farm was a significant TDN source to surface waters. Both TDN concentration (median: 15.17 mg/L) and export (median: 1.00 kg-N/day) DW of the farm were substantially greater than UP of the farm (median: 1.25 mg/L and < 0.01 kg-N/day). The median nitrate (NO_3^-) concentrations surpassed the USEPA drinking water limit of 10 mg/L (USEPA, 2002). While these surface waters are not direct sources of drinking water, they may contribute to downstream watersheds that supply municipal systems. The elevated levels of NO_3^- near the SFs pose risks to both ecological integrity and public health (Ruggierio et al 2025; Burkholder et al. 2007; Mallin et al. 2004, 2015). High nitrogen concentrations can lead to eutrophication, algal blooms, and hypoxia, and are linked to adverse human health effects, including methemoglobinemia, birth defects, and colorectal cancer (Weyer et al. 2001; Ward et al. 2005). The resulting cyanobacterial blooms can release harmful microcystins, degrade water quality, and impose significant ecological and economic costs (Dodds, 2002; Diaz and Rosenberg, 2008; Townsend et al. 2003; Jeffrey et al. 2009).

Previous research in North Carolina has shown that groundwater contributes 35 to 60 % of streamflow, allowing nutrients from the subsurface to enter surface systems (Spruill et al. 2005; Harden et al. 2013). The elevated NO_3^- observed in surface waters in this study is consistent with nitrification of NH_4^+ rich groundwater as it discharges through oxic zones with high ORP and DO. These findings mirror other reports showing that CAFO land-application areas are significant sources of NO_3^- to nearby streams, especially within the North Carolina Coastal Plain (Burkholder et al. 2007; Karr et al. 2001; Harden, 2015; Mallin et al. 2004; Harden and Spruill, 2008). Harden (2015) found that streams near CAFOs had median total nitrogen levels ranging from 0.20 to 17.0

mg/L, substantially greater than background sites (1.0 to 1.3 mg/L), supporting the elevated TDN and NO_3^- observed in this study (Table 3.7).

The strong connectivity between groundwater and surface water underscores how nutrient laden discharges from CAFOs can mobilize nitrogen downstream, amplifying eutrophication potential and affecting broader watershed health. Elevated NH_4^+ levels ($\geq$ 0.5 mg /L) have been shown to stimulate algal blooms in coastal plain streams (Mallin et al. 2004), while concentrations up to 37.8 mg/L and total nitrogen as high as 46.7 mg/L have been measured in CAFO-adjacent waters (Mallin et al. 2004). Such conditions promote cyanobacterial dominance and toxin production (Glibert et al. 2015; Siegel et al. 2011), degrading ecosystem function and posing ongoing water quality challenges. The elevated NH_4^+ and TDN concentrations observed in this study, particularly downgradient of SFs, therefore confirm that CAFO operations exert a measurable influence on adjacent and downstream water quality (Harden, 2015; Mallin et al. 2015; Richardson, 2023).

Additional work is recommended to verify which biogeochemical processes are occurring in both groundwater and surface water. Results from this study suggest that adsorption, nitrification, denitrification, and plant and microbial uptake are the primary nitrogen transformation pathways occurring within the LG–SF–stream continuum. However, additional data are required to confirm these processes. Future research should incorporate molecular and isotopic tools to identify the specific microbial communities and pathways involved. Techniques such as PCR and metagenomic sequencing provide a powerful means of confirming the presence and activity of nitrogen-transforming microorganisms. By targeting functional genes such as *amoA* for nitrifiers, *nirK/nirS* and *nosZ* for denitrifiers, and *hzsA/hzo* for anammox bacteria, these molecular tools can identify microbial communities driving nitrogen cycling in groundwater and surface water (Ducey et al. 2011; Guo et al. 2016; Huang et al. 2023). Complementary use of stable isotope analysis ($\delta^{15}N–NO_3^-$ and $\delta^{18}O–NO_3^-$) can trace nitrogen sources and distinguish between denitrification and physical dilution processes (Hosono et al. 2013). Additionally, measuring intermediate nitrogen species such as NO_2^- and N_2O, together with detailed redox profiling, provides direct evidence of ongoing nitrogen transformations and helps quantify pathway contributions under varying redox conditions (Beaulieu et al. 2010; Harvey et al. 2013).

CHAPTER 4 : PHOSPHORUS RESULTS AND DISCUSSION

4.1 Phosphate Concentration in Wastewater

Wastewater in the LG contained the highest median concentration of phosphate (7.48 mg/L), which was about 2 to 3 times greater than groundwater locations (Table 4.1; Fig. 4.1). These differences in phosphate were significant ($p < 0.01$). Phosphate concentrations in the LG ranged from 6.12 to 14.04 mg/L. Past studies found that total P (TP) ranged from 22 to 329 mg/L (Ham and DeSutter, 2000). Ducey et al. (2019) reported a range of 7.9 to 94.0 mg/L for phosphate in swine wastewater from two commercial swine finishing farms in North Carolina housing approximately 1,200 to 1,500 and 2,100 to 2,200 pigs per production cycle. Vanotti et al. (2003b) found liquid swine with mean values of 81 mg/L and 46 mg/L for TP and phosphate concentrations, respectively, from a lagoon near Kenansville in Duplin County, NC, that served around 2,600 hogs. Szogi and Vanotti (2009) reported that ten LGs in North Carolina (a full-scale operation containing approximately 4,360 finishing pigs) had a range of 17.8 to 79.1 mg/L for phosphate (mean: 51 mg/L) and 26 to 85.2 mg/L for TP (mean: 61.2 mg/L) in 10 LGs. Median phosphate concentrations in wastewater in the current study tended to be lower than those reported in previous studies. However, phosphate concentrations did partially overlap with ranges reported in other studies (7.9 to 94.0 mg/L in Ducey et al. [2019]), 17.8 to 79.1 mg/L in Szogi and Vanotti [2009]). Past studies (Vanotti et al. 2003b; Szogi and Vanotti, 2009) found that phosphate consisted of about 57 to 100% of TP. If this speciation trend is representative in the current study, then TP concentrations may range from 8.94 to 15.12 mg/L.

Swine waste is typically rich in organic P (≥ 70% of TP) that originates from animal feces and feed (feed composition, particularly the P content and use of phytase additives) (Rosemarin et al. 2021). Phosphate concentrations in LG undergo similar treatment processes as nitrogen, where organic P mineralizes into inorganic P (mostly as phosphate) via anaerobic microbial decomposition (USEPA 2016). Additionally, LG is a low-energy environment that promotes sedimentation of solid waste particles. Swine wastewater often contains protein and metal elements such as potassium, sodium, calcium, magnesium, iron, manganese, copper, zinc, and nickel. These metals can form ligands with phosphate, reducing phosphate concentration in wastewater (Chakrabarti 1993; Burkholder et al. 2007; Reddy et al. 1999; Michalopoulos et al. 2014). Additionally, sedimentation of particles can increase phosphate retention within the LG, potentially reducing water quality impacts to water resources. Kleinman et al. (2011), Hamilton (2010), and USEPA (2016) reported that particulate forms of P can settle and deposit as sludge, eventually requiring a pump out to restore storage capacity. Therefore, the LG wastewater is land applied intermittently to free up more space for wastewater inflow. The pumped wastewater is irrigated on farmland, which can pose environmental issues. Wastewater within the LG contained elevated SC (Table 4.1), indicating that wastewater contains numerous dissolved ions. Thus, SC can be used as an indirect tracer to evaluate if groundwater or surface water is influenced by wastewater application. Furthermore, trends in SC data support the likelihood of ligand formation occurring between metals (Burkholder et al. 2007; Michalopoulos et al. 2014), proteins (Chakrabarti 1993), or other substances within the LG. The pH in the LG is alkaline, which could encourage the phosphate interactions with ions and form a precipitate (Table 4.2; Szogi et al. 2018; Aneja et al. 2008).

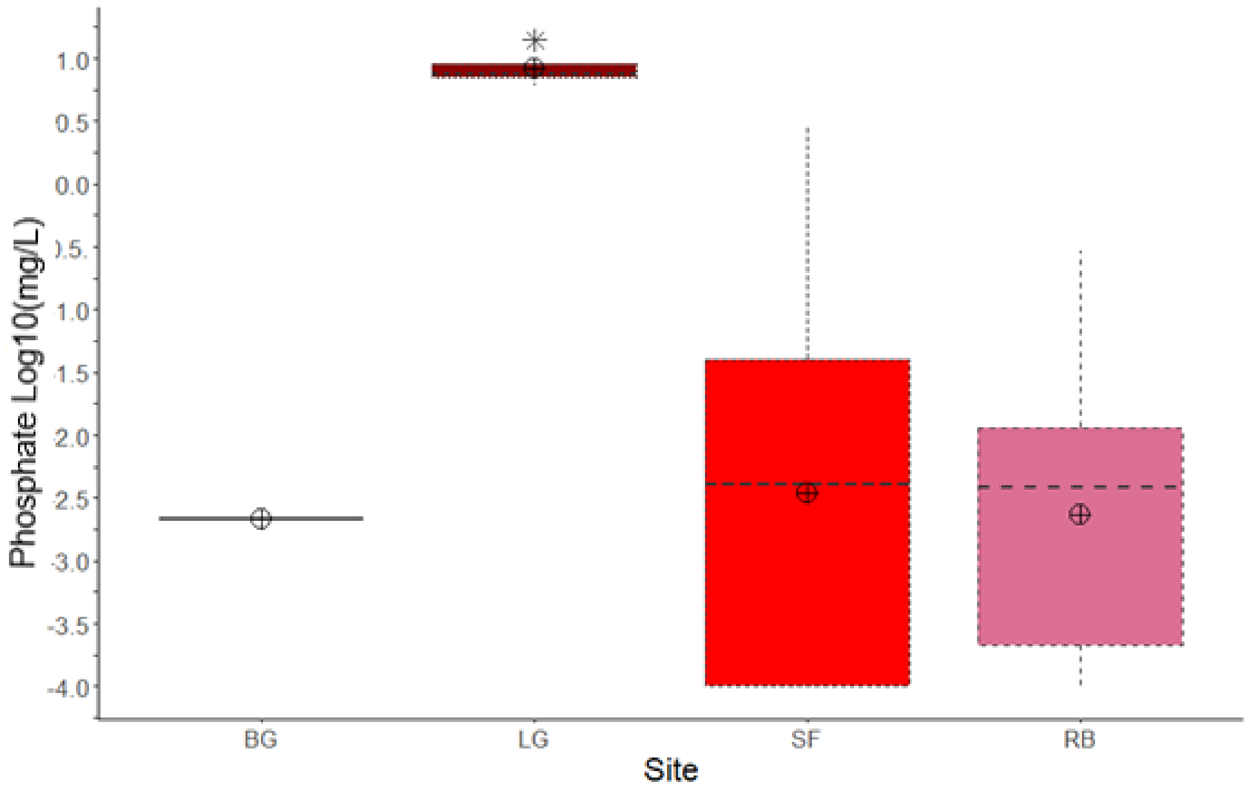

Figure 4.1: Boxplot showing concentrations of Phosphate for background (BG), lagoon (LG), spray field (SF), and riparian buffer (RB).

Table 4.1: Summary of median and range for physicochemical parameters and phosphate concentration in groundwater and wastewater locations. Temperature (Temp); specific conductance (SC); dissolved oxygen (DO); oxidation-reduction potential (ORP); depth to water (DTW) in groundwater and wastewater locations such as lagoon (LG); background (BG); spray field (SF); riparian buffer (RB)

Site	PO_4^{3-} (mg/L)	DTW (cm)	Temp (°C)	SC (µS/cm)	pH	ORP (mV)	DO (mg/L)
LG	7.48 (6.12-14.04)		21.83 (11.52-30.70)	13105.00 (5735.00-14970.00)	8.50 (6.75-8.74)	-213.50 (-315.90- 5.40)	0.31 (0.01-1.00)
BG	0.002	137.16[a]	21.10 (21.10-21.10)	31.00 (31.00-31.00)	6.84 (6.84-6.84)	219.00 (219.00-219.00)	2.95 (2.95-2.95)
SF	0.001 (<0.001-2.94)	82.92 (3.35-233.17)	22.33 (12.70-27.70)	432.00 (48.00-1025.00)	5.80 (4.56-6.93)	56.60 (-153.00-226.00)	1.06 (0.01-5.97)
RB	0.004 (<0.001-0.29)	74.83 (24.99-145.69)	20.02 (12.10-24.10)	213.00 (58.00-319.00)	4.90 (4.6-6.90)	106.35 (-32.30-287.10)	1.81 (0.46-6.70)

[a]= DTW measured in July 2023, all other sampling events were dry, thus DTW was > 232 cm (BG) (total piezometer depth).

4.2 Groundwater Phosphate Concentration and Mass Reduction

Phosphate concentrations in groundwater tended to be low in all sampling locations (Tables 4.1 and Figs 4.1). The groundwater concentration beneath the SF ranges from < 0.001 to 2.94 mg/L, with a median concentration of 0.001 mg/L (Table 4.1; Fig. 4.1). Thus, the median concentration reduction of phosphate was >99% between the LG and SF. P1 was the only SF piezometer where phosphate concentrations were consistently > 0.01 mg/L. This piezometer was also the only one where the maximum phosphate concentration exceeded 1 mg/L. Except for P2, all other SF piezometers had a median phosphate concentration ≤ 0.01 mg/L. Furthermore, some piezometers (P7, P8, P10) had a maximum range of phosphate concentration in SF groundwater that was ≤ 0.01 mg/L. Past studies reported a phosphate range of <0.02 mg/L to 3.8 mg/L from 19 monitoring wells (Collins 1996). Additionally, Michalopoulos et al. (2014) reported phosphate concentrations that overlap with the aforementioned range (< 0.3 mg/L to 2.58 mg/L). Smith et al. (2001) also found that phosphate concentrations in agricultural runoff varied from 0.87 mg/L (in samples pretreated with alum) to 5.50 mg/L (in untreated wastewater), with a background concentration of 0.55 mg/L

Groundwater phosphate concentrations beneath the studied SFs tended to be similar to BG groundwater. Apart from P1, the median phosphate concentration in SF groundwater was similar to BG groundwater (Table 4.4). The current study was only able to yield one BG sample due to drought conditions, which caused the piezometer to be dry during most sampling events. Data collected from a previous study (Iverson, unpublished data) revealed that BG groundwater contained a median phosphate concentration of 0.01 mg/L (range: <0.01 to0.04 mg/L). These data also support the similarity in phosphate concentration between SF and BG groundwater. Additionally, SC in groundwater beneath the SFs (Table 4.1) was elevated relative to BG (current study: 32 μS/cm; Iverson, unpublished data: median 29 μS/cm [21 to 145 μS/cm]). These data (median phosphate: <0.01 mg/L) indicate that SF groundwater was influenced by wastewater applications. However, the low phosphate concentrations in groundwater beneath and downgradient from SFs suggest that soil processes within the vadose zone were likely effective at removing phosphate before swine wastewater recharged groundwater. Similarly, groundwater beneath RBs also contained low phosphate concentrations (median: 0.01 mg/L, range: <0.01 to 0.29 mg/L; Table 4.1). This was expected based on phosphate trends in SF groundwater. It is possible that dilution could influence concentration reductions; therefore, mass reduction estimates are necessary.

Table 4.2: The table shows the range and median concentrations of phosphate (PO_4^{3-}) and physiochemical parameters for the groundwater sample piezometers and the lagoon (LG). pH, Temp= Temperature; SC= specific conductance, DO= dissolved oxygen, ORP= oxidation-reduction potential, DTW= depth to water. The groundwater groups (BG= background, SF= spray field, and RB= riparian buffer), and Lagoon= LG.

Site	PO_4^{3-}	DTW (ft)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
LG	7.48 (6.12-14.04)		21.83 (11.52-30.70)	13105.00 (5735.00-14970.00)	8.50 (6.75-8.74)	-213.50 (-315.90-5.40)	0.31 (0.01-1.00)
BG	0.002	137.16[a]	21.10	31.00	6.84	219.00	2.95
P1	0.76 (0.03-2.94)	158.50 (121.92-195.99)	20.10 (13.20-25.70)	668.50 (360.00-1025.00)	5.66 (5.20-6.03)	72.00 (-107.80-153.60)	1.40 (0.01-2.77)
P2	0.03 (0.01-0.05)	104.85 (63.53-121.01)	23.25 (13.30-27.30)	85.00 (48.00-150.00)	5.65 (5.08-6.74)	103.25 (36.00-183.40)	1.77 (1.00-5.20)
P3	0.01 (<0.001-0.29)	64.31 (24.99-87.48)	20.57 (12.70-24.10)	220.00 (173.00-278.00)	4.81 (4.60-6.90)	106.35 (-32.30-228.00)	2.01 (0.95-6.70)
P5	0.002 (<0.001-0.01)	125.27 (33.83-145.69)	19.70 (12.10-22.30)	163.00 (58.00-319.00)	4.95 (4.75-6.88)	118.80 (51.00-287.10)	1.65 (0.46-3.30)
P6	0.004 (<0.001-0.16)	103.02 (70.71-233.17)	19.60 (13.40-26.00)	400.00 (269.00-1568.00)	6.66 (6.50-6.81)	-109.80 (-136.00- -83.60)	0.95 (0.90-1.00)
P7	<0.001 (<0.001- <0.01)	81.99 (74.98-91.44)	21.40 (13.50-25.80)	690.50 (651.00-804.00)	6.35 (5.30-6.93)	-71.05 (-149.00-69.30)	0.65 (0.01-1.13)
P8	<0.001 (<0.001-0.002)	60.20 (27.43-88.39)	21.90 (12.90-25.70)	353.50 (307.00-439.00)	5.47 (4.78-6.73)	107.20 (15.70-194.70)	2.32 (0.33-5.97)
P9	<0.001 (<0.001-0.07)	78.18 (36.27-167.94)	21.26 (13.00-26.10)	525.50 (370.00-546.00)	5.15 (4.57-6.90)	94.60 (-91.00-226.00)	1.71 (0.03-2.88)
P10	<0.001 (<0.001-0.01)	92.66 (3.35-145.69[a])	21.94 (12.70-26.80)	383.35 (307.00-609.00)	4.73 (4.56-5.98)	175.25 (82.00-211.60)	1.42 (0.80-2.44)
P11	0.01 (<0.001-0.06)	125.27 (57.30-143.87)	22.40 (13.30-27.70)	441.00 (192.00-487.00)	6.38 (5.60-6.73)	-92.00 (-153.00-56.60)	1.00 (0.44-2.30)

[a]= DTW measured in July 2023, all other sampling events were dry, thus DTW was > 232 cm (BG) and >145.87 (P10) (total piezometer depth).

Results from the two-component mixing model indicated that mass removal processes accounted for most of the phosphate treatment observed in the current study (Tables 4.3 and 4.4). The ratio of Cl to phosphate increased by several orders of magnitude between the LG and groundwater beneath the SFs and RBs (Tables 4.3 and 4.4). These data suggest that dilution was not a dominant reduction mechanism since dilution would reduce both Cl and phosphate concentrations at a 1:1 rate (Humphery et al, 2013). If dilution were the sole reduction mechanism, the mixing model predicted phosphate concentrations of 0.20 and 0.25 mg/L in SF and RB piezometers, respectively. However, the measured median concentration of the pooled SF and RB piezometers was 0.002 and 0.004 mg/L, respectively. Assuming the percentage difference between predicted and observed concentrations represents mass removal, mass reduction of phosphate was >98% between the LG and groundwater. The individual piezometers suggested that mass removal occurred in all the piezometers except for P1 and P2. The highest recorded mass removal was observed in P7 to P10, which had over 99%, suggesting that the soil was an effective treatment medium for phosphate (Table 4.4).

Negative mass reductions typically indicate that the observed phosphate concentration was greater than predicted, suggesting that mass removal processes were inhibited at these sites. The estimated mass reduction at P2 was very low (about -261%); however, the predicted (0.01 mg/L) and observed (0.03 mg/L) concentrations of phosphate only differed by 0.02 mg/L (Table 4.4). Based on the low concentrations of phosphate and Cl, it is unlikely that wastewater application significantly influenced water quality at this site. At P1, the mass reduction percentage was slightly negative (-8.47%), which was likely due to a high outlier in the dataset. During the July sampling event, this piezometer could not be located since it had been damaged. It was later located and repaired in the following sampling event. The damaged piezometer was a direct conduit of swine wastewater to the surficial aquifer, resulting in an elevated groundwater phosphate concentration in August (2.94 mg/L). If this outlier was removed, the median concentration at P1 decreased to 0.62 mg/L, and the estimated mass reduction increased to about 12%. Another possible explanation for the negative reduction estimates could be due to the relatively low phosphate concentrations in the LG relative to other studies. If the median phosphate concentration was similar to past studies (Szogi and Vanotti 2009), then the predicted phosphate concentration would increase to 4.7 and 0.06 mg/L for P1 and P2, respectively. Consequently, the mass reduction estimates would also increase to 84% and 47% for P1 and P2, respectively. Thus, if wastewater phosphate concentrations were underestimated, then the mixing model estimate will be negatively affected.

Table 4.3: Two-component mixing model using chloride and phosphate (PO_4^{3-}) to estimate phosphate mass reductions. Chloride concentrations (Cl), fraction of wastewater (fraction WW), fraction of groundwater (fraction BG), predicted phosphate, observed phosphate, Cl /PO_4^{3-} ratio, and phosphate mass reduction are shown for sites Lagoon (LG), Spray field (SF), Riparian Buffer (RB), and Background (BG).

Site	Cl (mg/L)	Fraction WW	Fraction GW	Predicted PO4 (mg/L)	Observed PO_4^{3-} (mg/L)	Cl/PO_4^{3-} Ratio	Mass reduction (%)
LG	1076.00	1	0		7.48	143.78	
BG	3.93	0	1		0.002	1838.83	
SF	32.90	0.03	0.97	0.20	0.002	537997.90	99.01
RB	39.81	0.03	0.97	0.25	0.004	10449.97	98.47

Table 4.4: Two-component mixing model using chloride and phosphate (PO_4^{3-}) to estimate phosphate mass reductions in groundwater piezometers. Chloride concentrations (Cl), fraction of wastewater (fraction WW), fraction of groundwater (fraction GW), predicted phosphate, observed phosphate, Cl /PO_4^{3-} ratio, and phosphate mass reduction are shown for sites BG= background, SF= spray field, RB= riparian buffer, and lagoon= LG.

Site	Location	Cl (mg/L)	Fraction WW	Fraction GW	Predicted PO_4^{3-} (mg/L)	Observed PO_4^{3-} (mg/L)	Cl/PO_4^{3-} Ratio	Mass reduction (%)
LG	Wastewater	1076.00	1.00	0.00		7.48	143.85	
BG	Background	3.93	0.00	1.00		0.0021	1871.43	
P1	Spray Field	104.35	0.09	0.91	0.70	0.76	137.30	-8.47
P2	Spray Field	5.12	0.00	1.00	0.01	0.03	170.67	-261.32
P3	Riparian Buffer	49.43	0.04	0.96	0.32	0.01	4943.00	96.85
P5	Riparian Buffer	30.95	0.03	0.97	0.19	0.00	15475.00	98.94
P6	Spray Field	11.78	0.01	0.99	0.05	0.00	2945.00	92.70
P7	Spray Field	19.88	0.02	0.98	0.14	0.00	19880.00	99.28
P8	Spray Field	80.52	0.07	0.93	0.53	0.00	80520.00	99.81
P9	Spray Field	96.18	0.09	0.91	0.64	0.00	96180.00	99.84
P10	Spray Field	25.65	0.02	0.98	0.15	0.00	25650.00	99.34
P11	Spray Field	26.94	0.02	0.98	0.16	0.01	2694.00	93.77

4.3 Soil Data Impact on Phosphate Attenuation

Soil analyses at the farm site revealed that phosphate concentrations were primarily retained within the upper 0 to 15 cm of the soil profile at the SF locations (Table 4.3). This enrichment in the surface horizon indicates strong adsorption and accumulation within the topsoil, where repeated effluent applications are likely to enhance phosphate buildup. Subsurface horizons (below ~55 cm) showed markedly lower phosphate concentrations, suggesting that most phosphate remains bound near the surface and that downward transport through the soil profile is limited. This pattern aligns with previous findings that phosphate is largely immobile under typical agricultural field conditions due to its strong sorption to soil minerals and organic matter (Sims et al. 2005; Reddy et al. 1999).

Soil analyses revealed elevated concentrations of aluminum (Al), calcium (Ca), magnesium (Mg), and iron (Fe) oxides, elements known to enhance phosphate sorption and promote mineral precipitation. Soils with high clay content, such as those of the Roanoke series underlying the study site, possess abundant reactive surfaces that bind phosphate through ligand exchange and form insoluble P-bearing minerals such as variscite and vivianite (Kleinman et al. 2011; Schelde et al. 2006; USDA NRCS, 2006). Over time, these reactions facilitate the formation of stable inorganic P phases, effectively sequestering phosphate into mineral forms that are resistant to leaching. This precipitation process represents a dominant geochemical pathway for phosphate immobilization within the vadose zone (Reddy et al. 1999). However, phosphate desorption may occur when exchange sites become saturated or inundated due to fluctuating water tables or prolonged flooding associated with extreme weather conditions. Excessive phosphate application on saturated soils, coupled with certain tillage practices and surface runoff, has been documented to increase desorption potential (USEPA, 2016; Kleinman et al. 2011; Schelde et al. 2006). In this study, the consistently low phosphate concentrations observed in groundwater suggest that desorption was minimal.

Trace metals such as zinc (Zn) and copper (Cu), which can compete with phosphate for sorption sites, were generally found to be low (Appendix A). This further indicates that iron and aluminum oxides, along with calcium and magnesium, serve as the primary factors influencing phosphate adsorption in this system. Soil pH values exhibited a decline with depth across most sampling locations, ranging from approximately 4.4 to 6.3 (Table 4.5). Also, concentrations of the soils Zn and Cu were significantly lower than average state background levels (Appendix A), suggesting a minimal risk of phytotoxicity or regulatory exceedance. These results imply that current effluent management practices have not led to significant metal accumulation. However, ongoing monitoring is necessary to ensure compliance with nutrient management and land application standards.

The soils at the SF and RB locations demonstrated elevated levels of clay and Fe/Al oxide content (Iverson, unpublished data), which correlates with the low median groundwater phosphate concentrations observed at these sites. This pattern aligns with previous research that highlighted strong phosphate fixation in soils characterized by high clay and Fe/Al content in the Coastal Plain (Schelde et al. 2006; Sharpley et al. 2013). Collectively, these features suggest that adsorption and mineral precipitation were the primary mechanisms influencing phosphate mobility in the subsurface.

Metal analyses (Appendix A) also revealed measurable but generally low concentrations of cadmium (Cd), chromium (Cr), lead (Pb), nickel (Ni), and arsenic (As) across soil horizons. Lead and nickel concentrations in certain surface samples (e.g., P5B = 8.7 mg/dm^3, P1A = 2.8 mg/dm^3) were slightly above typical background values for North Carolina soils (Hardy et al. 2008) yet remained below regulatory limits for land application (4.8 mg/dm^3 and 0.8 mg/dm^3, respectively). Continued accumulation of these metals, however, could become a concern under prolonged effluent irrigation, potentially leading to exceedances of state standards for land-applied waste.

Table 4.5: The pH and fertility analyses of soil samples measured in mg/dm3 for the following elements P= Phosphorus, K= potassium, Ca= calcium, Mg= magnesium, and S= sulfur.

Location	Site	Depth (cm)	P	K	Ca	Mg	S	pH
SF	P1A	0-15.24	112	265	641	79	27	5.7
	P1B	15.24-55.88	6	420	176	32	37	5
	P1C	55.88-109.22	2	219	144	44	64	4.4
	P1D	109.22-175.26	2	74	115	47	29	4.4
	P2A	0-10.16	135	158	517	67	33	5.1
	P2B	10.16-53.34	17	64	153	29	89	4.4
	P2C	53.34-93.98	4	32	101	35	48	4.4
	P2D	93.98-121.92+	6	25	195	103	70	4.3
	P4A	0-12.7	22	64	493	123	23	5
	P4B	12.7-22.86	2	57	1009	508	19	5.3
	P4C	22.86-53.34	2	66	1054	520	12	6.2
	P4D	53.34-106.68	1	47	571	253	17	6.3
RB	P3A	0-20.32	48	116	310	69	32	4.5
	P3B	20.32-63.5	9	94	131	50	17	4.6
	P5A	0-15.24	4	56	129	94	17	4.5
	P5B	15.24-50.8	2	120	172	191	19	4.4
BG	BG	0-20.32	11	39	132	61	18	4.5

4.4 Stream Phosphate Concentration and Mass Transport

The phosphate concentrations were minimal in all streams regardless of location relative to SFs. The median stream phosphate concentration was < 0.01 mg/L in all streams, and phosphate concentrations did not significantly differ between any locations ($p = 1$). Interestingly, SC and DO values suggest that streams adjacent to and downgradient from SFs were influenced by wastewater, which typically contains elevated SC and low DO (Table 4.2). In contrast, the UP location exhibited higher DO and lower SC, with median values of 6.45 mg/L and 48.15 µS/cm, respectively. Median SC increased in all other streams by at least fivefold, while median DO concentrations declined in all other streams apart from EC-U (Table 4.6). These trends in SC and DO, along with minimal phosphate concentrations, indicate that the CAFO was not a major source of phosphate to surface water. Rather, most SFs appeared effective at removing P within the vadose zone and/or along groundwater flow paths before discharge into nearby streams.

This observation aligns with previous findings in North Carolina's Coastal Plain. Harden (2015) studied 49 stream sites categorized as background (BG), swine only (SW), or swine plus poultry (SP) and reported phosphate concentrations ranging from 0.026 to 0.030 mg/L, while TP ranged from 0.098 to 0.122 mg/L, with no statistically significant differences among groups. Similarly, Harden and Spruill (2008) documented a median phosphate concentration of 0.05 mg/L (range: 0.01 to 0.29 mg/L) in streams adjacent to swine waste application fields during both baseflow and stormflow. Collectively, these findings reinforce that surface waters near swine operations in this region generally exhibit low phosphate concentration, suggesting effective phosphate attenuation before reaching streams if soils have adequate adsorption sites.

In a study of a stream located downstream of intensive swine and poultry operations in North Carolina, Stone et al. (1995) reported mean phosphate concentrations of 0.68 mg/L during baseflow and 1.3 mg/L during stormflow. In a follow-up study within the same watershed, specifically the Herrings Marsh Run (HMR) watershed, a 2,044-ha demonstration watershed located in the Cape Fear River Basin in Duplin County, North Carolina (Stone et al. 1998). The monitored swine operation expanded from approximately 3,300 hogs to more than 14,000 hogs between 1990 and 1997; despite this increase, phosphate mean values ranged only from 0.05 to 0.10 mg/L and remained steady over the study period. This concentration was typically greater than what was observed in the current study.

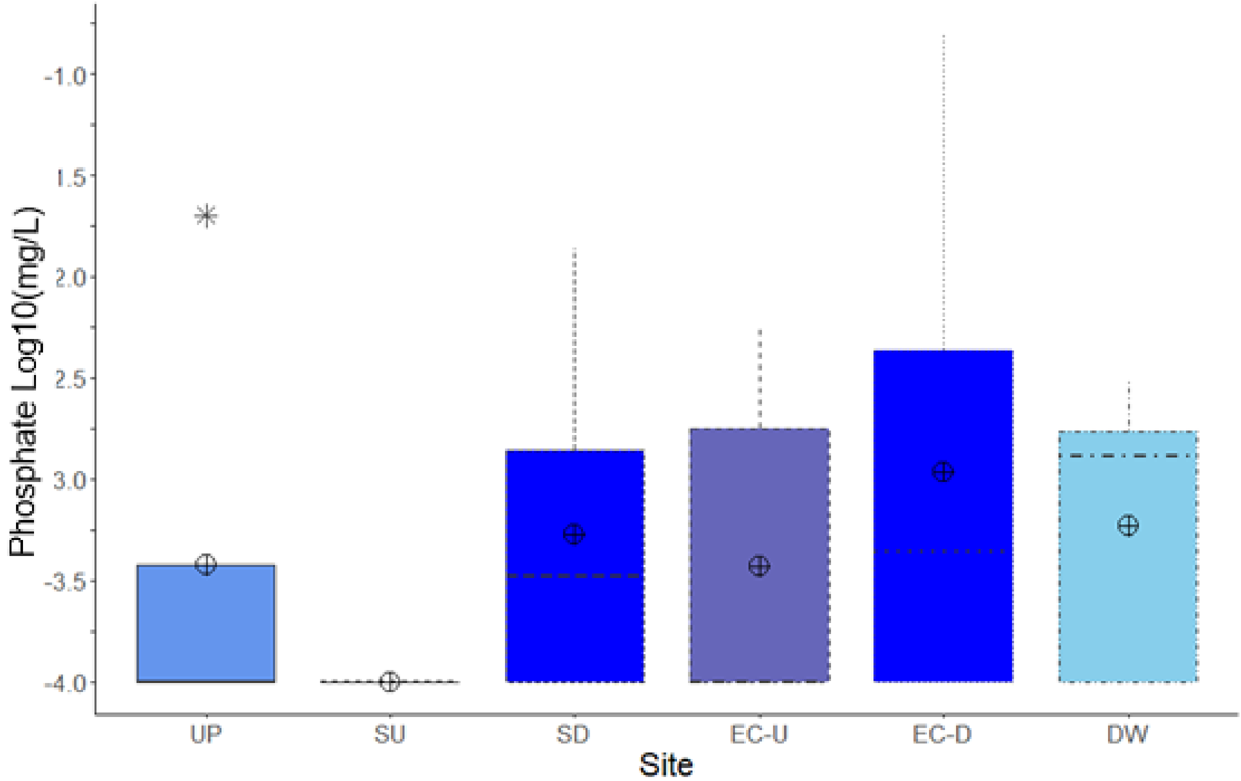

Figure 4.2: Boxplot showing concentrations of phosphate for Upstream (UP), Seep Up (SU), Seep Down (SD), East Creek Down (EC-D), East Creek Up (EC-U), and Downstream (DW).

Table 4.6: Table showing Phosphate concentrations and physicochemical parameters collected at the farm through the sample events for UP= Upstream, SU= Seep Up, SD= Seep Down, EC-D= East Creek Down, EC-U= East Creek Up, and DW= Downstream, median (range) of pH, Temp= temperature, SC= specific conductivity, DO= dissolved oxygen, Turb= turbidity, and ORP= oxidation reduction potential.

Site	PO_4^{3-} (mg/L)	Temp (°C)	SC (µS/cm)	Turb (NTU)	pH	ORP (mV)	DO (mg/L)
UP	< 0.001 (< 0.001 - 0.02)	17.45 (10.30-24.45)	48.15 (40.80-89.00)	5.12 (3.33-80.00)	4.19 (3.60-5.82)	330 (164.00-384.20)	6.45 (5.00-9.10)
SU	< 0.001 (< 0.001 - < 0.001)	18.80 (10.93-28.48)	919.00 (846.00-969.00)	0.05 (0.00-5.3)	4.91 (4.82-5.13)	294.50 (278.00-421.00)	5.72 (3.80-8.50)
SD	0.001 (< 0.001 - 0.01)	18.93 (13.12-26.02)	870 (710.00-930.00)	20.35 (1.40-112.00)	6.10 (4.64-6.59)	281.75 (198.00-326.90)	3.90 (1.44-12.50)
ECU	< 0.001 (< 0.001 - 0.006)	19.57 (10.42-26.20)	264.50 (200.00-379.00)	52.60 (5.10-689.00)	6.07 (5.41-7.01)	199.05 (146.60-218.00)	6.54 (4.70-9.14)
ECD	0.001 (< 0.001 - 0.15)	20.05 (10.38-26.70)	507.50 (348.00-762.00)	52.60 (5.10-689.00)	6.72 (6.00-7.01)	147.20 (-8.00-188.40)	3.51 (0.01-10.14)
DW	0.001 (< 0.001 - 0.003)	20.27 (11.20-26.00)	418.00 (255.00-481.00)	26.10 (9.50-850.00)	6.57 (5.92-7.10)	158.00 (73.90-210.50)	3.67 (1.45-9.70

Discharge data are summarized in Table 4.7, but were previously discussed in Chapter 3, Section 3.6. Refer to this section for the discussion of the stream discharge data.

The median mass transport at the farm streams was higher than at all other stream sites, with both SD recording the highest median phosphate mass transport of 0.05 g-P/day (Table 4.7). The SD, EC-D, and DW locations exhibited the highest median mass exports among all stream locations; however, phosphate mass loadings did not exhibit any statistically significant differences (p = 1). The median mass transport at the UP location was < 0.01 g-P/day, respectively (Table 4.7). The median mass export of phosphate DW of the farm was slightly elevated relative to UP, but this difference was not statistically significant. These findings suggest that the farm was neither a source of phosphate to Horse Pen Branch nor downgradient surface water. It is likely that drought conditions influenced stream phosphate transport UP and DW of the farm. Past studies have recorded elevated phosphate exports compared to the mass export recorded in the current study. Haggard et al (2003) reported an average TP export of 0.34 Kg-P/ha/year (340 g-P/ha/year) for streams draining pastures that are irrigated by poultry wastewater. This current study found that Horse Pen Branch exported 0.06 g-P/ha/year of phosphate, suggesting that the farm was not a significant source of phosphorus to downstream water resources. Drought conditions influenced the results recorded in this study. This drought condition resulted from low precipitation that occurred during the sampling period and was discussed in detail in the previous chapter (section 3.6).

Richardson 2023 reported median discharge values of 2142 L/min (DW) and 1008 L/min (UP) during non-drought conditions. This resulted in phosphate export estimates of 1102.38 g-P/year and 51.88 g-P/year for the DW and UP, respectively (using the median phosphate concentration in this current study [Table 4.6] and the discharge from Richardson 2023). The normalized DW export estimate without drought conditions was 4.54 g-P/ha/year, which was 75 times more than the export (0.06 g-P/ha/year) recorded during the drought conditions. This further suggests that drought conditions affected the mass export from DW. The drought locations (UP and DW) were supplemented with data from previous studies (Iverson, unpublished data), and the supplemented median mass phosphate transport was 3.66 and 1.22 g-P/day for UP and DW, respectively ($p = 0.1$). The Seep and East Creek (farm streams) median mass transport remained the same (0.02 g-P/day). The difference between farm streams and UP was statistically significant ($p = 0.03$), while the difference between farm streams and DW was not significant ($p > 0.3$). The supplemented data further indicate that the farm was not a significant source of phosphate to the DW location.

Despite clear evidence of wastewater influence based on physicochemical indicators (e.g., elevated specific conductance), phosphate concentrations in surface water remained consistently low. This pattern suggests that phosphate was effectively attenuated within the soil and shallow subsurface before reaching surface waters. The observed attenuation is most likely attributable to the strong sorption capacity of the soils, which contain ions, as well as to biological uptake and sediment retention processes within the vadose zone and riparian areas. Collectively, these findings demonstrate that, under the hydrologic conditions observed during this study, the farm functioned as an effective treatment system for phosphate, substantially limiting off-site transport and reducing the potential for downstream eutrophication.

Table 4.7: Median and range (in parentheses) for Discharge (Q), mass transport, and phosphate (PO_4^{3-}) concentration for surface water sample locations: UP= Upstream, SU= Seep up, SD= Seep down, EC-D= East Creek down, EC-U= East Creek up, and DW = Downstream, showing.

Site	PO_4^{3-} (mg/L)	Discharge (Q) (L/min)	Mass Transport (g-P/Day)
UP	< 0.001 (< 0.001-0.02)	0.00 (0.00-282.39)	0.00 (0.00-3.82)
SU	< 0.001 (< 0.001-< 0.001)	151.75 (30.28-283.72)	0.01 (<0.01-0.04)
SD	0.001 (< 0.01-0.01)	128.89 (31.60-215.74)	0.05 (<0.01-3.10)
ECU	< 0.001 (< 0.01-0.006)	11.00 (0.00-308.68)	0.02 (0.00-0.08)
ECD	0.001 (< 0.01-0.15)	163.37 (2.69-370.09)	0.03 (<0.01-0.61)
DW	0.001 (< 0.01-0.003)	54.44 (0.00-485.97)	0.04 (0.00-0.41)

4.5 Fate and Transport of Phosphorus in Groundwater and Surface Water

Groundwater phosphate concentrations were ≤ 0.01 mg/L in all piezometers, apart from P1 and P2. This suggests that phosphate attenuation within the vadose zone was highly effective. The combination of adsorptive soil properties, chemical precipitation, and plant uptake likely contributed to P treatment. The moderately acidic conditions indicated in the soil data are conducive to phosphate sorption by Fe and Al oxides, particularly within the subsoil horizons (Michalopoulos et al. 2014; USDA NRCS, 2000). The acidic profile, therefore, reinforces that phosphate retention mechanisms remain effective throughout much of the soil column, although slightly higher pH near the surface may favor Ca to P precipitation. Soil data also suggested that the presence of Al, Ca, Mg, and Fe (Table 4.5; Appendix A) supported phosphate adsorption and precipitation. This pH behavior also has implications for P mineralization and mobility under fluctuating redox conditions. Phosphate in the environment can desorb under aerobic conditions and alkaline pH. Similarly, during anaerobic conditions and acidic pH, the internal loading of phosphate remains notably elevated (Nguyen and Maeda, 2016). However, this phenomenon was likely limited due to drought conditions, which likely influenced groundwater depth during winter months (i.e., deeper than normal seasonal high water table)

The most significant reduction in phosphate mass was observed in groundwater at the SF and RB locations. These reductions suggest that geochemical processes, in conjunction with plant and soil microorganisms, may play a crucial role in phosphate removal (Reddy et al. 1999; Yadav et al. 2017; Zhu and Wheldan, 2018). The mass reductions observed at both SF and RB locations suggest that these locations may not pose environmental concerns. The median phosphate concentration in groundwater beneath the SF was low and likely does not contribute significantly to eutrophication in receiving waters (Golterman and Oude, 1991; Qi et al. 2022). Conversely, the median phosphate concentration in P1 (0.76 mg/L) exceeded the recommended threshold of 0.05 mg/L (Golterman and Oude 1991; Qi et al. 2022). Thus, if groundwater with similar phosphate concentration as P1 discharged to nearby surface waters (particularly in nutrient-sensitive waters), then the risk of water quality degradation increases. More specifically, adverse environmental impacts including eutrophication, algal blooms, and fish kills may occur. The nearest surface water feature to P1(median concentration: 0.76 mg/L) is the Seep (SD median mass transport: 0.05 g-P/day), which is situated approximately 5486.4 cm (180 ft) away. Reddy et al. (1999) showed that phosphate transport in soils is typically limited to meters to tens of meters due to strong adsorption and precipitation reactions.

Plant uptake is another potentially significant phosphate reduction mechanism at the swine CAFO (Hoffmann et al. 2009). Studies by Reddy et al. (1999) suggest that plants take up phosphate from soil porewater, influencing phosphate distribution and potentially impacting its movement. Vegetation within RB and SF serves as a critical sink for phosphate through root uptake, biomass accumulation, and harvest removal. Studies have quantified these reductions, showing that plant uptake can immobilize up to 15 kg-P/ha/yr in riparian buffers (Hoffmann et al. 2009) and store between 40 to 375 kg-P/ha in emergent macrophyte biomass, with over half in harvestable above-ground tissues (Reddy and DeBusk, 1987).

P-solubilizing microorganisms, such as *Bacillus*, *Pseudomonas*, and *Aspergillus* species, have been documented to enhance P cycling by mobilizing insoluble phosphate and increasing its bioavailability (Divjot et al. 2021; Pang et al. 2024). However, the presence of diverse vegetation and organic matter inputs at the RB location likely supported these microbial communities, fostering both immobilization (through precipitation) and bio assimilation. The median phosphate concentrations across all stream locations were recorded at below 0.01 mg/L, with no significant differences observed among the sites (Table 4.7). The wastewater in the LG indicates that the farm is a source of phosphate to the environment. However, these consistently low concentrations from the farm suggest minimal phosphate contributions to water resources, indicating that the soil was a substantial source of phosphate attenuation. The findings indicate that soil treatment processes, particularly adsorption, precipitation, and microbial immobilization, effectively retain phosphate before it can reach water resources.

The Seep, East Creek, and DW sites demonstrated low levels of phosphate, indicating effective attenuation of phosphate in the soils overlaying the surficial aquifer and/or along RB. The estimated mass transport of phosphate from the DW site was 0.04 g-P/day, suggesting that off-site phosphate loss is negligible compared to the nitrogen export discussed in Chapter 3. This minimal phosphate export implies that soil treatment and biogeochemical processes play a crucial role in mitigating environmental risks to receiving waters. However, should future monitoring reveal increasing P loads in downgradient locations, it may be necessary to implement additional attenuation measures, such as buffer enhancement or controlled effluent loading, to preserve this stability.

Overall, phosphate within this system was primarily immobilized in the soil matrix through processes such as adsorption, precipitation, and biological uptake. The observed reductions in phosphate concentration and mass across the SF and RB locations indicate that both geochemical and biotic controls effectively limit P mobility. Although groundwater and surface water can act as potential pathways for nutrient transport in CAFO systems, the findings suggest that, under the current management practices and hydrologic conditions, the risk of P-induced eutrophication remains minimal.

Nonetheless, continued monitoring of TP, soil mineralogy, and P-index parameters is essential for evaluating the long-term sustainability of swine wastewater irrigation and maintaining protection of adjacent aquatic ecosystems.

4.6 Phosphate Mass Budget

The DW mass export for phosphate was 0.04 g-P/day and 14.6 g-P/year (Table 4.8). This suggests that the farm could attenuate about 99.9% of the wastewater input it receives from LG. Assuming the median phosphate in the LG is representative, the average mass input to SFs was approximately 569.28 g-P on application days. The annual estimated phosphate mass load to all SFs was 57,497 g of phosphate during the growing season that overlapped with the study period.

Table 4.8: Daily and annual discharge and mass budget estimate in the farm location. The number of spraying and sampling events for the period was also indicated.

Location	Spraying/sampling events	Discharge Budget (L)		Mass Budget (g)	
		Daily	Year	Daily	Year
SF 5	21	109678.50	2303248.20	820.40	17228.30
SF 4	19	61998.30	1177967.70	463.75	8811.20
SF 3	27	66973.47	1808283.75	500.96	13525.96
SF 2	16	68456.46	1095303.30	512.05	8192.87
SF 1	18	72331.35	1301964.30	541.04	9738.69
DW	6	54.44	19870.6	0.04	14.60

To calculate the net mass input on the farm, the DW mass export was deducted from the mass export at the UP location. Since the export at the UP location is zero, this indicates that the CAFO wastewater irrigation primarily contributed to the mass export at DW. This suggests that the farm attenuated 99.9% of the wastewater land applied. Median phosphate mass export from DW was approximately 0.04 g-P/day (Table 4.8). The estimated annual export was approximately 14.60 g-P/year (Table 3.9). Thus, there was a >99% reduction between the annual phosphate load to SFs relative to the annual phosphate export from the DW sampling location.

Drought conditions may have influenced P mass attenuation estimates. In a past study at the same farm, stream discharge DW of the farm was estimated at 2150 L/min during normal conditions. Assuming that the median phosphate concentration DW of the farm is representative (0.001 mg/L), then Horse Pen Branch exported 1154 g-P/yr. Some of this estimate also includes non-wastewater-derived phosphate originating from soil organic matter, other animal waste, and plant decay. If the UP exports represent these contributions (median: 52 g-P/year), then 1102 g of phosphate originated from the CAFO. This estimate suggests that the CAFO attenuated 98% of the phosphate mass applied to the SFs (57,496 g-P/yr). The negligible phosphate export observed during drought conditions indicates minimal P transport, reflecting the low mobility in farm soils.

Although non-drought conditions may result in episodic increases in phosphate export driven by runoff and other conditions, an estimated annual export of 1154 g-P/yr (approximately 3 g-P/day) is small and unlikely to cause eutrophication or other water-quality impairments in receiving waters. This export rate is orders of magnitude lower than P loads typically reported for agricultural watersheds and would be substantially diluted under normal streamflow conditions, yielding concentrations below commonly cited eutrophication thresholds (approximately 0.03 to 0.10 mg/L) (Golterman and Oude, 1991; Kleinman et al. 2011; USEPA, 2000). Additionally, median phosphate concentration in surface water locations did not exceed the recommended threshold of 0.05 mg/L (Golterman and Oude 1991; Qi et al. 2022). Phosphate mass transport could episodically increase during severe storms that generate agricultural runoff. However, more research is needed to evaluate mass transport in response to storm activity

CHAPTER 5 : CONCLUSION AND MANAGEMENT IMPLICATIONS

The goal of this study was to evaluate the influence of a swine CAFO on nutrient inputs to groundwater and surface water by quantifying nutrient concentrations in wastewater, assessing nutrient transport pathways, and estimating nutrient reduction efficiencies across the LG, SF, and RB system. Results showed that LG wastewater contained elevated nutrient concentrations, with a median TDN of 1651 mg/L (dominated by NH_4^+) and a median phosphate concentration of 7.48 mg/L. Groundwater beneath SF exhibited substantial nutrient attenuation, with >99% concentration reductions for both TDN (median = 13.89 mg/L) and phosphate (median < 0.01 mg/L), indicating effective subsurface treatment mediated by biogeochemical processes likely occurring within the vadose zone. Two-component mixing models and increasing Cl/N and Cl/PO4 ratios suggested that dilution was not the predominant reduction mechanism; thus, mass removal processes (e.g., volatilization, denitrification, adsorption, and mineral precipitation) likely occurred. Surface water locations (Seep, East Creek, and DW) adjacent to and downgradient from SF exhibited elevated TDN concentrations relative to upstream locations, whereas phosphate concentrations were minimal (≤ 0.01 mg/L). Findings by the current study concur with past studies indicating that CAFOs can be a significant source of nitrogen to water resources, which is particularly concerning for nutrient-sensitive waters located near a greater density of CAFOs.

North Carolina’s prominence in swine production, especially in the ENC, carries important implications, particularly for environmental public health threats. Many swine CAFOs in ENC are disproportionately located in low-income, rural, and historically marginalized communities, where residents rely heavily on private wells and shallow groundwater for drinking water and have limited resources to mitigate contamination risks (Wing et al. 2002). Although the water resources at the sample location are not designated drinking water sources, nitrogen species like NO_3^- are very mobile and can constitute an environmental public health concern (Ward et al. 2018). When nutrient-laden surface waters ultimately drain into water supply watersheds, increased pollutant loads impose measurable economic burdens on drinking water utilities, as elevated nitrogen concentrations require more intensive and costly treatment processes to meet potable water standards (Temkin et al. 2019; Cullmann et al. 2025).

Within this framework, the findings of this study reported that P transport was effectively mitigated and unlikely to pose downstream risks under observed circumstances. However, excess wastewater applications beyond the adsorption capacity can pose environmental challenges. Therefore, the use of the required nutrient ratio is recommended. Elevated nitrogen levels in groundwater beneath SFs suggest an ongoing risk to water resources if management measures fail or hydrological conditions shift. The study reported that drought conditions resulted in low nutrient export; however, during normal conditions, DW could be a source of nutrients to water resources near CAFOs. The DW indicated elevated nitrogen mass transport during normal conditions (absence of drought). Therefore, it is highly recommended to implement BMPs that are suitable for the farm. These practices should aim to promote or enhance attenuation of nitrogen within the CAFO to mitigate TDN mass transport at the watershed scale.

The adoption of BMPs, such as the expansion of vegetated buffer strips, the use of denitrifying bioreactors, and the implementation of controlled drainage systems, can substantially enhance the removal of nitrogen before it enters surface waters (Messer et al. 2012; Rosen and Christianson, 2017). Research has demonstrated that instream and subsurface bioreactors are effective in promoting denitrification in agricultural contexts, particularly when supplemented with an additional carbon source like wood chips. Instream bioreactors have had success at reducing nitrogen concentrations. Robertson and Merkley (2009) found nitrate reductions from 4.8 mg/L to 1.04 mg/L using an instream bioreactor and wood chips to promote denitrification. Increasing RB area can also help reduce nitrogen mass transport from the farm. The results from the farm also indicated the efficiency of RB location in nutrient attenuation. Furthermore, nitrification (of elevated NH_4^+ in the groundwater location) and denitrification can be further optimized by increasing residence time through controlled drainage combined with bioreactors (instream and subsurface bioreactors) (Woli et al. 2010; Christianson et al. 2021). These results suggest and identify management strategies that can improve nutrient attenuation, minimize water quality impacts, and prevent eutrophication.

Extreme weather events such as droughts, floods, and storms can significantly disrupt fate, transport, and management of nutrients. Drought conditions often lead to a marked reduction in microbial activity, resulting in decreased nutrient availability, while extended periods of dryness hinder decomposition and lower nitrogen levels. On the other hand, heavy rainfall and flooding can wash away nutrients like nitrogen and phosphorus into water bodies, leading to eutrophication and the degradation of aquatic ecosystems (Imran et al. 2025). This underscores the importance of adopting BMPs tailored to existing extreme weather conditions.

Climate variability adds another layer of complexity to nutrient management, especially in swine CAFO systems, where liquid waste application is governed by annual permitting schedules that do not necessarily account for year-to-year swings in hydrological conditions. One question that emerges directly from the findings of this study is whether application rates should be adjusted based on prevailing or anticipated climate conditions, given that TDN export in surface waters was substantially lower during drought. At first glance, reduced streamflow and lower nutrient exports during dry periods might seem to suggest that more waste could be applied to the land without meaningfully increasing nutrient loading to nearby streams. However, drawing that conclusion would be premature.

Dry conditions can reduce surface runoff and weaken the hydraulic connection between streams and shallow groundwater, which would suppress what was observed in surface water monitoring. But suppressed surface export does not necessarily mean nutrients are staying put. Subsurface flow pathways may continue moving nutrients laterally through the soil profile and into adjacent aquifers, just more slowly and through routes that standard surface water sampling would not detect (Heeren et al. 2010; Basu et al. 2011). It is not possible to determine whether the low TDN values observed during drought reflect genuine nutrient retention in the soil and crop system, or simply a shift in transport pathway from surface to subsurface. Nutrients stored in shallow groundwater during dry periods could re-enter streams once wetter conditions return, a pattern that has been documented and sometimes referred to as legacy nutrient loading (Jarvie et al. 2013; Van Meter et al. 2018). Allowing higher application rates during drought, without understanding what is happening beneath the soil, introduces a risk that is difficult to quantify but potentially significant.

With that caveat in mind, the findings of this study do support the broader argument that nutrient management strategies should be sensitive to hydrologic conditions rather than relying on fixed annual schedules. During wet years or periods following significant storm events, application rates should be curtailed and timed to avoid windows when soils are already saturated, and runoff risk is elevated. Under typical hydrological conditions, the 4Rs framework (right source, right rate, right time, right place) provides a workable foundation, with application timing synchronized to periods of active crop uptake to minimize losses to the environment (Kabir and Syfullah, 2026; USDA 2017). During dry or drought years, while surface water data may show lower nutrient export, the conservative approach is to maintain rather than increase application rates until more is understood about subsurface nutrient behavior under those conditions. Future work that pairs the dynamics of groundwater and surface water monitoring across hydrologically distinct periods would help resolve whether drought-period reductions in stream nutrient export reflect true immobilization or a temporary shift in pathway. Until that kind of data is available, reduced surface water export during drought should not be read as an indication that the system has additional assimilative capacity to absorb more waste.

Some limitations should be considered when interpreting the results of this study. Prolonged drought conditions during the sampling period reduced stream discharge and limited groundwater availability, which affected nutrient mass transport estimates. Mass transport reported in the current study likely underestimated nutrient transport relative to a normal water year. In addition, the six-month sampling duration yielded a relatively small dataset, which reduced statistical power and limited the ability to fully capture seasonal variability and storm-driven nutrient fluxes. Despite these limitations, the observed data provide important insights into nutrient behavior under baseflow-dominated conditions and suggest that, when properly managed, the LG-SF-RB system can function as an effective treatment train for nutrient attenuation. Treatment of nitrogen and phosphate in the vadose zone overlying groundwater highlights the critical role of soil mineralogy and biogeochemical processes. Additionally, maintaining adequate riparian buffers along streams can also improve nutrient management by reducing transport to downstream water resources. RB location reduces nutrient mass transport by facilitating plant uptake and other biogeochemical processes that remove nutrient masses (such as increasing residence time, microbial uptake, and denitrification) (Messer et al. 2012; Sharpley et al. 2013). Past studies found that RBs can reduce nutrients by approximately 61 to 99% at the site scale (Messer et al.; Hyatt et al.; Harden and Spruill, 2008)., confirming their effectiveness as cost-efficient, hot-spot, and nature-based nutrient treatment systems (Hefting et al. 2004; Vymazal, 2007). Continued monitoring under non-drought conditions, particularly during storm events, is recommended to more fully characterize nutrient export dynamics and to ensure long-term protection of downstream water resources.

Additional work is recommended to confirm the specific biogeochemical processes governing nutrient fate in groundwater and surface water. Results from this study suggest that adsorption, nitrification, limited denitrification, and plant and microbial uptake are the dominant nitrogen transformation pathways; however, confirmation requires the integration of molecular and isotopic tools. Future nitrogen-focused research should combine molecular techniques (e.g., PCR and metagenomic sequencing targeting *amoA*, *nirK/nirS*, *nosZ*, and *hzsA/hzo*) with stable isotope analyses ($\delta^{15}N–NO_3^-$ and $\delta^{18}O–NO_3^-$), intermediate nitrogen species measurements (NO_2^-, N_2O), and redox profiling to distinguish biological transformation from dilution and to quantify pathway contributions under varying redox conditions (Beaulieu et al. 2010; Ducey et al. 2011; Hosono et al. 2013; Guo et al. 2016; Huang et al. 2023). Additionally, adopting the most effective and efficient BMP that is suitable for the farm to attenuate the nutrients from wastewater application. For phosphorus, future work should focus on sequential chemical extractions, phosphorus speciation, and mineralogical analyses to directly quantify sorption capacity, precipitation mechanisms, and long-term phosphorus stability in soils and sediments, particularly under fluctuating hydrologic and redox conditions (Reddy et al. 1999; Kleinman et al. 2011; Sharpley et al. 2013).

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APPENDIX A: SOIL PROPERTIES SUMMARY

The following tables contain soil data for the groundwater (GW) sample locations, organized by soil physical and chemical properties, including humic matter (HM), weight per volume (W/V), cation exchange capacity (CEC), and base saturation (BS) of soil layers. Metal content including cadmium (Cd), chromium (Cr), arsenic (As), lead (Pb), nickel (Ni), selenium (Se), Zinc (Zn), and copper (Cu). Bolded text indicates content higher than the average of soil samples submitted to the NC Department of Agriculture and Consumer Services Lab.

APPENDIX A Table 1: Soil data showing depth range and ion concentrations for the groundwater (GW) sample locations

Groundwater	Sample Location	Depth (cm)	Cd mg/dm3	Cr mg/dm3	As mg/dm3	Pb mg/dm3	Ni mg/dm3	Se mg/dm3	Zn mg/dm3	Cu mg/dm3
SF	P1A	0-15.24	0	0.2	0.8	2.8	0.7	0.1	4.2	1.9
	P1B	15.24-55.88	0	0.2	0.2	1.7	0.6	0.2	0.3	0.4
	P1C	55.88-109.22	0	0.1	0.1	0.6	0.4	0.1	0.3	0.4
	P1D	109.22-175.26	0	0.1	0.3	1.2	**1.8**	0.1	0.4	0.4
	P2A	0-10.16	0.1	0.1	0.1	**4.5**	0.7	0.1	3	1.4
	P2B	10.16-53.34	0	**0.4**	0.1	2.7	0.5	0.1	0.3	0.7
	P2C	53.34-93.98	0	0.2	0.8	4	0.7	0.2	0.3	0.5
	P2D	93.98-121.92+	0	**0.3**	0.1	2.3	0.4	0.1	2.5	2.7
	P4A	0-12.7	0	0.2	0.2	**4.4**	**1.1**	0.1	2	0.5
	P4B	12.7-22.86	0	0.1	0.2	2.2	1	0.1	0.8	0.4
	P4C	22.86-53.34	0	0.2	0.5	1.5	0.7	0.1	1.7	0.7
	P4D	53.34-106.68	0	0.2	0.1	0.3	0.8	0.1	1.9	0.2
RB	P3A	0-20.32	0	0.2	0.3	2.4	0.4	0.1	2.4	0.5
	P3B	20.32-63.5	0	0.2	0.2	0.9	0.4	0.2	0.4	0.3
	P5A	0-15.24	0	**0.5**	0.2	4.2	0.7	0.1	0.9	1.4
	P5B	15.24-50.8	0	**0.8**	0.6	**8.7**	**0.9**	0.1	1.6	6.5
BG	BG	0-20.32	0	0.2	0.1	**5.1**	**1.1**	0.2	1.4	1.1

*Average content of 3,286 soil samples from North Carolina analyzed at the NCDAandCS lab from Hardy et al. 2008.

	Cd mg/dm3	Cr mg/dm3	As mg/dm3	Pb mg/dm3	Ni mg/dm3	Se mg/dm3	Zn mg/dm3	Cu mg/dm3
NC Avg*	**0.1**	**0.2**	**4.5**	**4.2**	**0.8**	**0.2**	**27.2**	**9.2**

APPENDIX A Table 2: Soil data showing depth range and soil parameters for the groundwater (GW) sample locations

Groundwater	Location	Depth (cm)	HM (g/100 cc)	W/V (g/cc)	CEC (meq/100 cc)	BS (%)
SF	P1B	0-15.24	0.71	1.25	5.8	77
	P1B	15.24-55.88	0.09	1.28	3.3	68
	P1C	55.88-109.22	0.04	1.14	5	33
	P1D	109.22-175.26	0.04	1.26	3.5	33
	P2A	0-10.16	0.46	1.25	5.3	67
	P2B	10.16-53.34	0.04	1.24	3.2	36
	P2C	53.34-93.98	0.09	1.26	2.1	42
	P2D	93.98-121.92+	0.04	1.18	3.5	55
	P4A	0-12.7	0.76	1.11	5.7	64
	P4B	12.7-22.86	0.04	1.1	10.7	88
	P4C	22.86-53.34	0.04	1.21	10.3	95
	P4D	53.34-106.68	0.04	1.24	5.3	95
RB	P3A	0-20.32	1.49	0.93	6.2	39
	P3B	20.32-63.5	0.18	1.2	3.7	35
	P5A	0-15.24	0.18	1.19	4.8	33
	P5B	15.24-50.8	1.43	0.83	9.1	30
BG	BG	0-20.32	2.15	1.16	4.3	30

APPENDIX B: RAW FIELD DATA

The following tables contain concentrations and physiochemical data for surface water and groundwater locations organized by date and measured in the field, then lab-analyzed. Blank locations are due to missed sampling events resulting from drought conditions, human and/or laboratory errors, and/or equipment failures. SC=specific conductance (µS/cm); pH; DO=dissolved oxygen (mg/L); Temp=temperature (°C); ORP=oxidation reduction potential (mV); Turb=turbidity (NTU); Q=discharge (L/min); DTW=depth to water (cm); NH_4^+=ammonia (mg/L); NO_3^-=nitrate (mg/L); DON= dissolved organic nitrogen (mg/L); DOC=dissolved organic carbon (mg/L); TDN=total dissolved nitrogen (mg/L); PO4=phosphate (mg/L); Cl=chloride (mg/L).

APPENDIX B Table 1: Riparian buffer raw data

Riparian Buffer (RB)								
Date	Piezometer	NH_4^+	NO_3^-	DON	TDN	PO4	DOC	CL
Jul -2023	P3			<0.01[a]				
Aug -2023	P3	3.94	0.12	<0.01[a]	4.07	0.02	24.02	39.81
Sep -2023	P3	1.54	0.80	<0.01[a]	2.34	0.01	10.45	49.43
Oct -2023	P3	0.17	0.88	0.37	1.41	<0.01[a]	5.89	52.94
Nov -2023	P3	0.24	3.11	3.01	6.37	<0.01[a]	4.60	53.71
Feb -2024	P3	0.08	0.14	3.33	3.55	0.29	45.58	38.02
Jul -2023	P5	0.90	8.65	<0.01[a]	9.54	0.01	2.88	44.17
Aug -2023	P5	0.53	0.75	0.78	2.06	<0.01[a]	4.15	39.64
Sep -2023	P5	1.11	0.01	0.54	1.67	<0.01[a]	5.79	22.25
Oct -2023	P5	1.03	0.11	<0.01[a]	1.14	<0.01[a]	4.70	4.70
Nov -2023	P5	0.12	0.13	1.27	1.52	0.01	1.78	17.55
Feb -2024	P5	0.16	7.11	5.51	12.78	<0.01[a]	3.55	53.32

[a]= DON was incalculable due to dissolved inorganic species representing 100% of TDN, while PO4 was below detection

APPENDIX B Table 2: Spray Field raw data

Spray Field (SF)								
Date	Piezometers	NH_4^+	NO_3^-	DON	TDN	PO4	DOC	CL
Jul -2023	P1			<0.01[a]				
Aug -2023	P1	162.42	1.01	<0.01[a]	163.42	2.94	177.40	64.44
Sep -2023	P1	32.84	7.08	<0.01[a]	39.92	0.03	20.49	104.35
Oct -2023	P1	43.13	8.89	<0.01[a]	52.02	0.76	21.58	106.64
Nov -2023	P1	23.48	4.45	5.83	33.76	0.48	21.41	104.38
Feb -2024	P1	12.78	5.63	2.57	20.98	1.15	17.45	99.06
Jul -2023	P2	3.24	0.04	<0.01[a]	3.28	0.03	18.56	3.78
Aug -2023	P2	1.90	0.07	<0.01[a]	1.97	0.01	8.17	5.24
Sep -2023	P2	0.85	0.02	0.49	1.36	0.01	7.74	5.00
Oct -2023	P2	0.18	0.09	1.10	1.37	0.03	4.43	4.78
Nov -2023	P2	0.31	0.09	1.39	1.79	0.03	3.98	7.23
Feb -2024	P2	0.11	0.04	1.43	1.58	0.05	3.98	6.26
Jul -2023	P8	0.78	7.68	<0.01[a]	8.46	<0.01[a]	6.28	89.83
Aug -2023	P8	1.50	3.57	<0.01[a]	5.07	<0.01[a]	6.87	86.33
Sep -2023	P8	1.25	0.00	1.03	2.18	<0.01[a]	7.97	83.91
Oct -2023	P8	0.19	0.28	0.11	0.58	<0.01[a]	3.48	77.13
Nov -2023	P8	0.34	0.23	1.40	1.98	<0.01[a]	3.57	65.27
Feb -2024	P8	0.11	1.64	2.26	4.01	<0.01[a]	3.77	59.60
Jul -2023	P9	19.37	0.14	<0.01[a]	19.51	<0.01[a]	14.03	103.53
Aug -2023	P9	10.00	0.03	<0.01[a]	10.04	<0.01[a]	14.48	79.81
Sep -2023	P9	13.52	3.35	<0.01[a]	16.87	<0.01[a]	4.63	95.94
Oct -2023	P9	28.15	8.87	<0.01[a]	37.01	<0.01[a]	4.45	116.03
Nov -2023	P9	11.66	2.81	3.96	18.43	0.07	6.45	85.40
Feb -2024	P9	13.14	6.40	4.61	24.16	<0.01[a]	3.78	96.39
Jul -2023	P11	21.31	0.14	<0.01[a]	21.45	0.01	45.92	5.01
Aug -2023	P11	13.79	0.04	<0.01[a]	13.83	0.06	44.36	34.15
Sep -2023	P11	4.30	0.08	1.21	5.59	0.01	42.67	19.67
Oct -2023	P11	17.16	0.23	<0.01[a]	17.39	<0.01[a]	54.90	60.99
Nov -2023	P11							
Feb -2024	P11	1.05	2.16	5.26	7.42	0.06	32.58	16.34

APPENDIX B Table 2: Spray Field raw data

Spray Field (SF)								
Date	Piezometer	NH_4^+	NO_3^-	DON	TDN	PO4	DOC	CL
Jul -2023	P6	10.05	1.11	<0.01[a]	11.16	0.16	46.48	3.95
Aug -2023	P6			<0.01[a]				
Sep -2023	P6	3.34	1.08	0.14	4.56	<0.01[a]	24.62	31.65
Oct -2023	P6							
Nov -2023	P6							
Feb -2024	P6	12.95	4.90	5.25	23.10	<0.01[a]	22.05	11.78
Jul -2023	P7	2.06	0.51	1.26	3.84	<0.01[a]	9.56	13.45
Aug -2023	P7	5.18	0.19	<0.01[a]	5.38	<0.01[a]	9.06	22.00
Sep -2023	P7	16.54	0.00	<0.01[a]	16.54	<0.01[a]	33.42	17.75
Oct -2023	P7	14.14	0.00	<0.01[a]	14.13	<0.01[a]	25.62	25.62
Nov -2023	P7	3.57	0.07	1.49	5.13	<0.01[a]	15.06	61.86
Feb -2024	P7	0.65	6.44	6.87	13.96	<0.01[a]	8.69	10.98
Jul -2023	P10							
Aug -2023	P10	11.18	20.60	<0.01[a]	31.78	<0.01[a]	2.44	18.01
Sep -2023	P10	11.18	16.46	<0.01[a]	27.64	<0.01[a]	3.05	22.87
Oct -2023	P10	8.89	20.37	<0.01[a]	29.26	<0.01[a]	2.57	28.44
Nov -2023	P10							
Feb -2024	P10	10.51	7.73	4.16	22.40	<0.01[a]	3.94	29.50

APPENDIX B Table 3: Background and Lagoon raw data

Background (BG) and Lagoon (LG)								
Date	Piezometer	NH_4^+	NO_3^-	DON	TDN	PO4	DOC	CL
Jul -2023	BG	0.15	0.05	0.87	1.07	0.00	3.45	3.93
Aug -2023	BG							
Sep -2023	BG							
Oct -2023	BG							
Nov -2023	BG							
Feb -2024	BG							
Jul -2023	LG	1161.03	0.84	<0.01[a]	1161.88	9.39	1141.00	762.07
Aug -2023	LG	2308.53	0.00	<0.01[a]	2308.49	7.20	1407.00	1273.64
Sep -2023	LG	2801.05	0.00	<0.01[a]	2801.01	7.05	1406.00	1148.90
Oct -2023	LG	2055.65	0.00	<0.01[a]	2055.62	6.12	1260.00	1163.94
Nov -2023	LG	1246.38	0.24	<0.01[a]	1246.63	7.77	1451.00	1003.09
Feb -2024	LG	414.39	0.00	<0.01[a]	414.37	14.03	704.60	461.62

APPENDIX B Table 4: Surface water (Seep and East Creek) raw data

Surface water (Seep and East Creek)									
Date	Site	NH_4^+	NO_3^-	DON	TDN	PO4	DOC	CL	Q (L/min)
Jul -2023	SD	0.27	52.39	<0.01[a]	52.66	<0.01[a]	4.19	71.84	215.75
Aug -2023	SD	8.02	68.60	<0.01[a]	76.62	<0.01[a]	7.90	108.10	62.44
Sep -2023	SD	0.08	66.07	<0.01[a]	66.16	0.01	6.08	98.82	158.72
Oct -2023	SD	0.68	61.34	<0.01[a]	62.02	<0.01[a]	8.94	101.61	31.60
Nov -2023	SD	<0.01[a]	57.72	<0.01[a]	57.72	<0.01[a]	2.87	84.50	128.90
Feb -2024	SD	<0.01[a]	54.36	3.91	58.27	<0.01[a]	2.71	76.91	
Jul -2023	SU	1.12	55.65	<0.01[a]	56.77	<0.01[a]	6.54	72.50	30.29
Aug -2023	SU	3.67	75.80	<0.01[a]	79.47	<0.01[a]	2.91	100.96	222.41
Sep -2023	SU	0.66	76.04	<0.01[a]	76.70	<0.01[a]	2.35	96.22	50.07
Oct -2023	SU	0.72	81.99	<0.01[a]	82.71	<0.01[a]	2.90	98.52	83.69
Nov -2023	SU	0.15	55.75	4.10	60.00	<0.01[a]	2.66	82.73	283.72
Feb -2024	SU								219.80
Jul -2023	EC-D	0.16	16.99	<0.01[a]	17.15	0.15	9.33	35.56	274.59
Aug -2023	EC-D	2.57	1.28	<0.01[a]	3.85	<0.01[a]	72.81	120.11	5.67
Sep -2023	EC-D	0.18	8.27	<0.01[a]	8.45	<0.01[a]	31.55	85.51	52.16
Oct -2023	EC-D	0.09	25.32	<0.01[a]	25.42	0.01	18.50	105.97	370.09
Nov -2023	EC-D	<0.01[a]	16.46	0.76	17.22	<0.01[a]	18.32	75.93	2.62
Feb -2024	EC-D	<0.01[a]	18.57	1.83	20.40	<0.01[a]	3.77	35.92	361.83
Jul -2023	EC-U	0.10	15.72	<0.01[a]	15.82	<0.01[a]	7.35	24.40	308.68
Aug -2023	EC-U	0.83	6.17	<0.01[a]	7.00	<0.01[a]	13.65	73.45	0.00
Sep -2023	EC-U	0.12	10.67	<0.01[a]	10.80	<0.01[a]	9.10	56.25	11.32
Oct -2023	EC-U	0.35	13.63	<0.01[a]	13.99	<0.01[a]	5.72	75.86	11.26
Nov -2023	EC-U	<0.01[a]	10.10	1.42	11.52	0.01	5.05	66.42	2.24
Feb -2024	EC-U	<0.01[a]	18.13	0.35	18.48	<0.01[a]	2.86	28.06	199.83

APPENDIX B Table 5: Upstream and Downstream raw data

Surface water (Upstream and Downstream)									
Date	Site	NH_4^+	NO_3^-	DON	TDN	PO4	DOC	CL	Q (L/min)
Jul -2023	UP1	0.44	0.30	<0.01[a]	0.74	<0.01[a]	13.44	6.64	132.69
Aug -2023	UP1								
Sep -2023	UP1								
Oct -2023	UP1								
Nov -2023	UP1								
Feb -2024	UP1	0.02	0.07	1.49	1.56	<0.01[a]	5.98	6.56	229.96
Jul -2023	UP2	0.04	0.06	1.02	1.12	<0.01[a]	10.64	3.94	84.30
Aug -2023	UP2								
Sep -2023	UP2								
Oct -2023	UP2								
Nov -2023	UP2								
Feb -2024	UP2	0.03	0.04	1.32	1.39	<0.01[a]	5.96	5.42	282.39
Jul -2023	DW	0.31	19.34	<0.01[a]	19.65	<0.01[a]	9.83	41.04	164.94
Aug -2023	DW	27.25	0.31	<0.01[a]	27.56	<0.01[a]	37.55	13.82	0.00
Sep -2023	DW	0.21	12.22	0.49	12.92	<0.01[a]	22.67	71.21	108.88
Oct -2023	DW	1.79	0.74	0.30	2.82	<0.01[a]	24.26	59.70	0.00
Nov -2023	DW								0.00
Feb -2024	DW	<0.01[a]	13.09	2.08	15.17	<0.01[a]	5.58	32.85	

APPENDIX B Table 6: Physiochemical parameters raw data for Riparian Buffer

Physiochemical parameters for Riparian Buffer (RB)							
Date	Piezometer	DTW (cm)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
Jul -2023	P3	37.19	23.2	245	6.71	-22	0.95
Aug -2023	P3	87.48	24.1	221	6.9	-32.3	6.7
Sep -2023	P3	74.37	21.1	219	4.67	89	2.1
Oct -2023	P3	75.29	20.04	173	4.6	228	6
Nov -2023	P3	54.25	16.58	278	4.95	183.8	1.92
Feb -2024	P3	24.99	12.7	207	4.6	123.7	1
Jul -2023	P5	41.76	22.3	239	6.79	180	1.69
Aug -2023	P5	130.76	21.9	185	6.88	56	1.6
Sep -2023	P5	122.83	20	141	4.75	57.6	3.3
Oct -2023	P5	145.69	19.4	115	4.85	51	2.55
Nov -2023	P5	127.71	17.02	58	4.75	189.9	1.1
Feb -2024	P5	33.83	12.1	319	5.05	287.1	0.46

APPENDIX B Table 7: Physiochemical parameters raw data for Spray Field

Physiochemical parameters for Spray Field (SF)							
Date	Piezometer	DTW (cm)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
Jul -2023	P1						
Aug -2023	P1	132.59	25.7	1025	6.03	-107.8	2.77
Sep -2023	P1						
Oct -2023	P1	184.40	21.7	360	5.84	43	1.8
Nov -2023	P1	195.99	18.5	692	5.47	101	0.01
Feb -2024	P1	121.92	13.2	645	5.2	153.6	1
Jul -2023	P2	66.14	26	150	6.74	86	1.4
Aug -2023	P2	121.01	27.3	107.6	5.8	50.7	2.13
Sep -2023	P2	99.06	23.9	87	5.08	36	1.38
Oct -2023	P2	110.34	22.6	64	5.4	151	5.2
Nov -2023	P2	113.69	18.6	48	5.97	112.4	2.42
Feb -2024	P2	65.53	13.3	83	5.5	183.4	1
Jul -2023	P8	28.35	25.7	439	6.73	82	1.8
Aug -2023	P8	88.39	25.5	407.9	5.16	78.4	2.83
Sep -2023	P8	56.39	22.4	382	5.54	15.7	0.33
Oct -2023	P8	64.01	21.4	325	5.39	172	5.97
Nov -2023	P8	76.20	17.6	318	5.63	132.4	3.12
Feb -2024	P8	27.43	12.9	307	4.78	194.7	0.58
Jul -2023	P9	53.34	26.1	516	6.77	1.7	1.9
Aug -2023	P9	167.94	25.9	370	6.9	-91	1.96
Sep -2023	P9	62.48	22.33	546	5.61	-24.8	1.51
Oct -2023	P9	93.88	20.2	546	4.57	226	2.88
Nov -2023	P9	118.57	17.7	500	4.68	187.5	0.32
Feb -2024	P9	36.27	13	535	4.65	219	0.03
Jul -2023	P11	57.30	27.7	338	6.73	-153	0.5
Aug -2023	P11	131.67	27.4	418	6.1	-53.4	0.44
Sep -2023	P11	121.62	23	487	6.5	-99.9	1.06
Oct -2023	P11	128.93	21.8	471	6.38	-92	2.3
Nov -2023	P11	143.87	18	464			
Feb -2024	P11	96.01	13.3	192	5.6	56.6	1

APPENDIX B Table 7: Physiochemical parameters raw data for Spray Field

Physiochemical parameters for Spray Field (SF)							
Date	Piezometer	DTW (cm)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
Jul -2023	P6	82.91	26	322	6.81	-136	0.9
Aug -2023	P6	111.25[a]					
Sep -2023	P6	102.41	21.7	400			
Oct -2023	P6	233.17	19.6	269			
Nov -2023	P6	103.63	17.6	1568			
Feb -2024	P6	70.71	13.4	736	6.5	-83.6	1
Jul -2023	P7	77.11	25.8	727	6.74	-69.3	1.13
Aug -2023	P7	91.44	25.5	804	6.93	-45.6	0.96
Sep -2023	P7	80.16	22.6	690	6.23	-101	0.33
Oct -2023	P7	83.82	20.2	691	6.47	-149	0.01
Nov -2023	P7	86.26	16.9	651	5.97	-72.8	0.01
Feb -2024	P7	74.98	13.5	669	5.3	69.3	1
Jul -2023	P10	145.69[a]					
Aug -2023	P10	68.88	26.8	334.7	4.56	170.3	2.44
Sep -2023	P10	65.84	22.47	307	4.62	211.6	1.79
Oct -2023	P10	3.35	21.4	609	5.98	82	1.05
Nov -2023	P10	145.69[a]					
Feb -2024	P10	64.01	12.7	432	4.84	180.2	0.8
Jul -2023	BG	137.16	21.1	31	6.84	219	2.95
Aug -2023	BG	232.26[a]					
Sep -2023	BG	232.26[a]					
Oct -2023	BG	232.26[a]					
Nov -2023	BG	232.26[a]					
Feb -2024	BG	232.26[a]					
Jul -2023	LG		30.7	10450	6.75	-237	0.3
Aug -2023	LG		29.9	14250	6.97	-293	0.6
Sep -2023	LG		22.25	14970	8.65	-315.9	0.32
Oct -2023	LG		21.4	12500	8.6	-190	0.01
Nov -2023	LG		13.87	13710	8.74	-177.6	0.01
Feb -2024	LG		11.52	5735	8.4	5.4	1

a= DTW measured in July 2023, all other sampling events were dry thus DTW was > 232 cm (BG), > 111.25 (P6), and >145.87 (P10) (total piezometer depth).

APPENDIX B Table 8: Physiochemical parameters raw data for Lagoon (LG) and Background (BG)

Physiochemical parameters for Lagoon (LG) and Background (BG)							
Date	Piezometer	DTW (cm)	Temp (°C)	SC (uS/cm)	pH	ORP (mV)	DO (mg/L)
Aug -2023	BG	232.26[a]					
Sep -2023	BG	232.26[a]					
Oct -2023	BG	232.26[a]					
Nov -2023	BG	232.26[a]					
Feb -2024	BG	232.26[a]					
Jul -2023	LG		30.7	10450	6.75	-237	0.3
Aug -2023	LG		29.9	14250	6.97	-293	0.6
Sep -2023	LG		22.25	14970	8.65	-315.9	0.32
Oct -2023	LG		21.4	12500	8.6	-190	0.01
Nov -2023	LG		13.87	13710	8.74	-177.6	0.01
Feb -2024	LG		11.52	5735	8.4	5.4	1

APPENDIX B Table 9: Physiochemical parameters raw data for Surface water (Seep and East Creek)

Physiochemical parameters for Surface water (Seep and East Creek)							
Date	Location	Temp (°C)	SC (uS/cm)	Turb (NTU)	pH	ORP (mV)	DO (mg/L)
Jul -2023	SD	26.02	701	95	5.76	209.7	3.2
Aug -2023	SD	24.99	908	112	6.2	198	3.28
Sep -2023	SD	20.2	828	1.4	6.38	259	6.1
Oct -2023	SD	17.65	901	7.5	6.59	309.9	1.44
Nov -2023	SD	13.12	930	3.1	5.99	304.5	4.51
Feb -2024	SD	13.2	839	33.2	4.64	326.9	12.5
Jul -2023	SU	28.4	846	0.1	4.96	281.4	3.8
Aug -2023	SU	26.94	905	5.3	5.1	278	4.2
Sep -2023	SU	20.8	858	0	5.13	283	6.1
Oct -2023	SU	16.8	937	0	4.85	421	5.34
Nov -2023	SU	10.93	969	5	4.82	329.9	7.32
Feb -2024	SU	11.93	933	0	4.86	306	8.5
Jul -2023	EC-D	26.7	348	65	6.11	178	3.8
Aug -2023	EC-D	24.1	762	689	7.01	13.2	0.01
Sep -2023	EC-D	21	552	253	6.93	-8	2.9
Oct -2023	EC-D	19.09	670	10.1	6.6	126.4	5.51
Nov -2023	EC-D	10.38	463	40.2	6.84	188.4	3.21
Feb -2024	EC-D	11.77	356	5.1	6	168	10.14
Jul -2023	EC-U	26.2	297	55	5.89	191.3	4.7
Aug -2023	EC-U	25.3	228	40	7.01	218	4.91
Sep -2023	EC-U	20.4	232	1000	6.2	202	5.8
Oct -2023	EC-U	18.73	379	18	5.94	146.6	7.27
Nov -2023	EC-U	10.86	200	0.7	6.36	196.1	9.13
Feb -2024	EC-U	10.42	309	3.4	5.41	203	7.43

APPENDIX B Table 10: Physiochemical parameters raw data for Surface water (Upstream and Downstream)

Physiochemical parameters for Surface water (Upstream and Downstream)							
Date	Location	Temp (°C)	SC (uS/cm)	Turb (NTU)	pH	ORP (mV)	DO (mg/L)
Jul -2023	UP1	24.45	89	6	5.82	164	5
Aug -2023	UP1						
Sep -2023	UP1						
Oct -2023	UP1						
Nov -2023	UP1						
Feb -2024	UP1	10.6	40.8	3.33	3.6	384.2	7.8
Jul -2023	UP2	24.3	53	80	4.6	288	5.1
Aug -2023	UP2						
Sep -2023	UP2						
Oct -2023	UP2						
Nov -2023	UP2						
Feb -2024	UP2	10.3	43.3	4.23	3.77	372.04	9.1
Jul -2023	DW	25.15	365	174	5.94	205	4.7
Aug -2023	DW	26	446	26.1	7.1	158	1.45
Sep -2023	DW	20.27	481	14	6.57	73.9	3.67
Oct -2023	DW	14.39	418	850	6.89	130.7	1.6
Nov -2023	DW						
Feb -2024	DW	11.02	255	9.5	5.92	210.5	9.7

APPENDIX C: RAW SPRAY FIELD IRRIGATION DATA

The following tables contain date, flow rate, time (min), and total Volume applied (gallons) for surface water and groundwater locations organized by date and measured in the field, then lab-analyzed.

Field	Size (Acres)	Date	Flow rate (gal/min)	Time (min)	Total Volume (gallons)
1A	2.13	5/29/2024	182	90	16380
		6/3/2024	182	90	16380
		6/10/2024	182	90	16380
		6/15/2024	182	90	16380
		6/26/2024	182	60	10920
		7/2/2024	182	60	10920
		8/14/2024	182	90	16380
		10/2/2024	182	90	16380
		10/7/2024	182	90	16380
1B	3.9	5/29/2024	182	120	21840
		6/3/2024	182	120	21840
		6/9/2024	182	120	21840
		6/15/2024	182	120	21840
		6/25/2024	182	120	21840
		7/2/2024	182	120	21840
		8/5/2024	182	120	21840
		10/2/2024	182	120	21840
		10/7/2024	182	180	32760
2A	1.9	5/30/2024	182	90	16380
		6/4/2024	182	90	16380
		6/10/2024	182	90	16380
		6/17/2024	182	90	16380
		6/26/2024	182	90	16380
		7/3/2024	182	90	16380
		8/14/2024	182	90	16380
		10/2/2024	182	60	10920

Field	Size (Acres)	Date	Flow rate (gal/min)	Time (min)	Total Volume (gallons)
3F	1.5	5/31/2024	182	60	10920
		6/6/2024	182	60	10920
		6/11/2024	182	60	10920
		6/20/2024	182	60	10920
		8/20/2024	182	60	10920
		10/3/2024	182	60	10920
		10/10/2024	182	60	10920
3E	3.85	5/31/2024	182	120	21840
		6/6/2024	182	120	21840
		6/11/2024	182	120	21840
		6/20/2024	182	120	21840
		8/20/2024	182	120	21840
		10/3/2024	182	120	21840
		10/10/2024	182	120	21840
3D	1.49	5/30/2024	182	75	13650
		6/5/2024	182	90	16380
		6/11/2024	182	90	16380
		6/17/2024	182	90	16380
		6/28/2024	182	90	16380
		8/15/2024	182	90	16380
3A	2.47	5/30/2024	182	120	21840
		6/5/2024	182	120	21840
		6/11/2024	182	120	21840
		6/17/2024	182	120	21840
		6/27/2024	182	120	21840
		8/15/2024	182	120	21840
		10/3/2024	182	120	21840
2B	2.47	5/30/2024	182	120	21840
		6/4/2024	182	120	21840
		6/10/2024	182	120	21840
		6/17/2024	182	120	21840
		6/26/2024	182	120	21840
		7/3/2024	182	120	21840
		8/14/2024	182	120	21840
		10/2/2024	182	60	10920

Field	Size (Acres)	Date	Flow rate (gal/min)	Time (min)	Total Volume (gallons)
4E	1.57	6/1/2024	182	90	16380
		6/7/2024	182	90	16380
		6/12/2024	182	90	16380
		8/21/2024	182	90	16380
		10/5/2024	182	90	16380
4B	2.69	5/31/2024	182	90	16380
		6/6/2024	182	90	16380
		6/12/2024	182	90	16380
		6/20/2024	182	90	16380
		8/21/2024	182	90	16380
		10/5/2024	182	90	16380
		10/11/2024	182	90	16380
4E	2.19	5/31/2024	182	90	16380
		6/6/2024	182	90	16380
		6/12/2024	182	90	16380
		6/20/2024	182	90	16380
		8/20/2024	182	90	16380
		10/3/2024	182	90	16380
		10/11/2024	182	90	16380

Field	Size (Acres)	Date	Flow rate (gal/min)	Time (min)	Total Volume (gallons)
5C	2.52	6/2/2024	182	120	21840
		6/8/2024	182	120	21840
		6/13/2024	182	120	21840
		8/27/2024	182	120	21840
		10/1/2024	182	120	21840
		10/6/2024	182	120	21840
5D	1.65	6/2/2024	182	75	13650
		6/8/2024	182	75	13650
		6/13/2024	182	75	13650
		10/1/2024	182	75	13650
		10/6/2024	182	90	16380
5B	3.51	6/1/2024	182	180	37620
		6/7/2024	182	180	37620
		6/13/2024	182	180	37620
		8/22/2024	182	180	37620
		10/6/2024	182	180	37620
5A	5.78	6/1/2024	182	240	43680
		6/7/2024	182	240	43680
		6/12/2024	182	240	43680
		8/22/2024	182	240	43680
		10/5/2024	182	240	43680