Season matters when sampling streams for swine CAFO waste pollution impacts

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ABSTRACT

Concentrated (or confined) animal feed operations (CAFOs) are the principal means of livestock production in the United States, and such facilities pollute nearby waterways because of their waste management practices; CAFO waste is pumped from the confinement structure into a cesspit and sprayed on a field. Stocking Head Creek is located in eastern North Carolina, a state with >9,000,000 head of swine confined in CAFOs. This watershed contains 40 swine CAFOs; stream water quality was investigated at seven sites during 2016, with five sampling dates in early spring and five in summer. Geometric mean fecal coliform counts were in the thousands/100 mL at five sites in spring and all seven sites in summer. Excessive nitrate pollution was widespread with concentrations up to >11.0 mg N/L. Seasonality played an important role in pollutant concentrations. In North Carolina, spraying animal waste on adjoining fields is permissible from March 1 through September 30. Seasonal data showed significantly higher ($p < 0.01$) concentrations of conductivity, nitrate, total nitrogen, total organic carbon, and fecal bacteria in summer as opposed to early spring. Thus, sampling performed only in winter–early spring would significantly underestimate impacts from swine CAFO spray fields on nearby waterways.

Key words | CAFO, fecal bacteria, nitrate, pollution, swine

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INTRODUCTION

The vast majority of swine and poultry in the USA is produced on an industrial scale in concentrated, or confined, animal feeding operations (CAFOs). The US Environmental Protection Agency (EPA 2014) has estimated that there are more than 450,000 CAFOs operating in the USA. In North Carolina, nearly all swine (>9,000,000 head) are produced in CAFOs (Mallin & Cahoon 2003). Other states producing millions of swine each year in CAFOs include Iowa, Illinois, Minnesota, and Nebraska (US Department of Agriculture (USDA) 2014a). CAFOs are known to produce large amounts of water and air pollutants that enter the surrounding environment (Ham & DeSutter 2000; Mallin 2000; Burkholder *et al.* 2007). In a large-scale US Geological Survey study within North Carolina, CAFO-impacted streams were found to contain significantly higher concentrations of nitrate, ammonium, various other ions, and total nitrogen (TN) compared with control streams in areas lacking CAFOs (Harden 2015). CAFOs are a direct threat to human health as they produce vast numbers of fecal microbes into the air and neighboring water bodies (Mallin & Cahoon 2003; Mallin *et al.* 2015), a proportion of which are carried downstream into larger water bodies (Arfken *et al.* 2013). Not only is this a human health issue (Burkholder *et al.* 2007) but it is also an environmental justice issue as these facilities are frequently erected in close proximity to low-income minority residents who cannot afford to move away from polluted areas (Wing *et al.* 2000). Thus, it is important for regulatory agencies and health officials to be able to obtain solid data on potential pollutants generated by these industrial-scale animal production facilities in order to protect human and ecosystem health.

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Swine CAFOs manage the wastes generated within the large animal confinement structures by draining or pumping feces and urine into outdoor cesspits which the industry refers to as waste ponds or lagoons, and periodically spraying the liquids out on adjacent fields that are typically planted with Bermuda grass (Cahoon & Ensign 2004; Rajbhandari *et al.* 2015). Importantly, in North Carolina the spraying of animal wastes on Bermuda grass fields is not permitted year-round, but rather constrained to March through September (NCDACS 1996). This coincides with the planting and green-up of Bermuda grass, which is used as a means to take up excess nitrogen. Thus, the magnitude of stream pollution from CAFO-generated swine wastes can potentially vary considerably by season. Such variability should strongly influence when and how often to sample streams containing swine CAFOs in their watersheds to fully understand human health and environmental threat potentials. This point is particularly relevant because sampling regimes (frequency, number of locations, and parameters) for adjacent streams are often negotiated among growers, lobbyists, regulators and citizens' groups, rather than simply dictated by an agency. This research compared stream water quality in a CAFO-rich watershed during March, the onset of the permitted animal waste spraying season, with August/September, several months into the spraying season.

SITE DESCRIPTION

The Northeast Cape Fear River is a fifth-order tributary of the sixth-order Cape Fear River on the Coastal Plain of North Carolina. The watershed of the Cape Fear contains approximately half of the 9,000,000-plus swine produced in North Carolina. Cahoon *et al.* (1999) estimated that the Cape Fear River basin produced 82,700 metric tons of nitrogen and 26,000 metric tons of phosphorus as livestock waste in this watershed. These estimates were from 1995, but the numbers generated by that research are likely very conservative at present. While swine numbers are stabilized at present due to a 1997 waste lagoon construction moratorium imposed on the swine CAFO industry by the state, poultry CAFOs are continuing to increase in that watershed and this state in general (Patt 2017).

Stocking Head Creek is a second-order stream in the Northeast Cape Fear River basin (Figure 1); its catchment area is 1,980 ha (4,893 acres) and its length to the Northeast Cape Fear River is 22.1 km (13.7 miles). The watershed soils are dominated by Noboco loamy fine sand, Johns fine sandy loam, Autryville loamy fine sand, Pactolus fine sand, Lumbee sandy loam, and Marvyn and Gritney soils (NRCS 2014b). Stocking Head Creek is an example of a swine and poultry CAFO-rich watershed, containing approximately 40 swine CAFOs and 11 poultry CAFOs with its permitted population of swine at approximately 94,000 head, and estimated population of poultry (based on active poultry CAFO structures) at more than 1.3 million broiler chickens or their equivalent in turkeys (Mallin *et al.* 2015). Along one of the roads in this watershed there are some freely grazing cattle, but they are absent in the other areas. During an earlier summer–fall investigation, Stocking Head Creek contained high nitrate and ammonium concentrations, periodic algal blooms, excessive fecal coliform bacteria densities, and elevated biochemical oxygen demand (BOD) during both wet and dry periods (Mallin *et al.* 2015). We note that human fecal microbial sources are sparse in this watershed. There are no point source dischargers in the watershed and septic system density has been estimated as only 0.03/ha (Mallin *et al.* 2015).

METHODOLOGY

A suite of pollutants known to be constituents of swine and poultry wastes was sampled during spring (March) and summer (August–September) of 2016. The following measurements of environmental conditions were made on-site using YSI field meters: water temperature, pH, dissolved oxygen (DO), turbidity, and specific conductance. Also on-site, samples were collected according to standard procedure for nutrients (ammonium-N, nitrate-N, TN, orthophosphate, and total phosphorus (TP), total organic carbon (TOC)) and fecal coliform bacteria. Samples were held in darkness on ice and delivered to a state-certified laboratory for analysis within required holding times. Specific laboratory analyses including ammonia-N (EPA 350.1), nitrate + nitrite-N (hereafter referred to as nitrate;

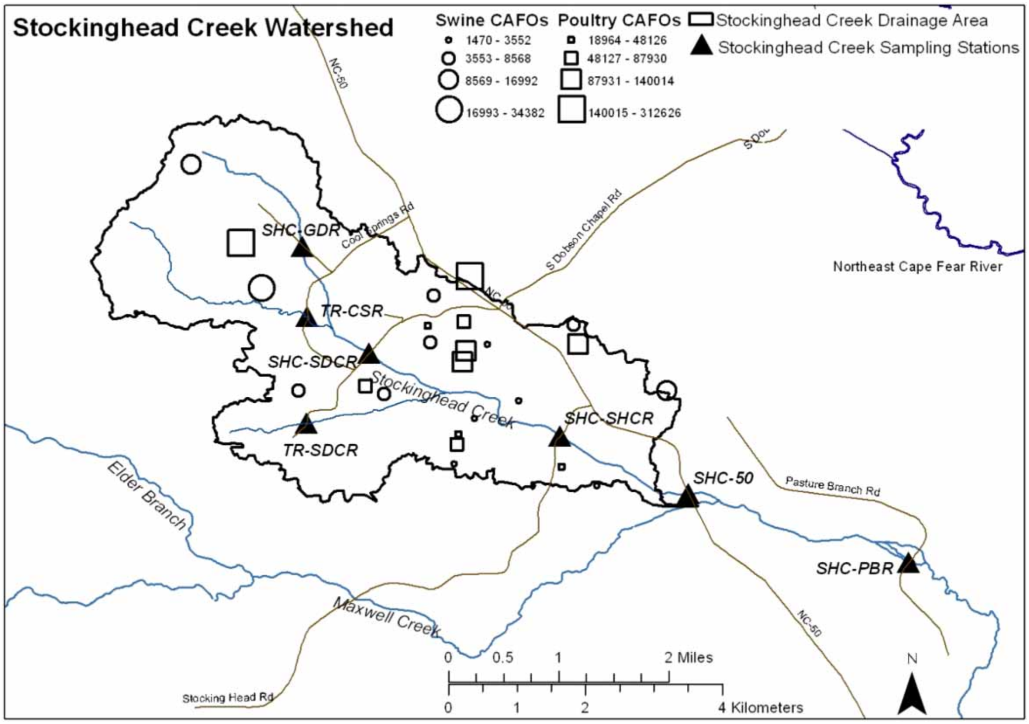

Figure 1 | Stocking Head Creek watershed showing tributaries, selected roads, sampling stations, swine and poultry CAFOs, and estimated animal populations.

EPA 353.2), TKN (EPA 351.2), TN (as the calculation of TKN + nitrate), orthophosphate (SM 4500PE), TP (SM 4500 PE), TOC (SM 5310B), and fecal coliform bacteria (SM 9222D MF). Chain-of-custody records were maintained following State of North Carolina protocols (Department of Environmental Quality (DEQ), Raleigh, NC).

Sample frequency

The overall approach was to conduct intensive sampling (five sample trips) during two different 30-day periods, one in early spring and the other in late summer. Sampling was planned to abide by the state's protocol for fecal coliform sampling (NC Department of Environment and Natural Resources 1999):

> *'The North Carolina protocol states that fecal coliform counts shall not exceed a geometric mean of 200 colony-forming units (CFU)/100 mL based on at least five consecutive samples during any 30 day period, nor exceed 400 CFU/100 mL in more than 20% of the samples examined during such period.'*

Fecal coliform samples were collected by filling pre-autoclaved containers ca. 10 cm below the surface, facing into the stream; the researchers wore rubber gloves while sampling.

Sample sites

Seven stations were sampled during both 30-day periods, including five on Stocking Head Creek proper and two on first-order tributaries (Table 1; Figure 1). All sites were sampled from bridges on public right-of-ways. Samples were stored on ice until laboratory processing (<6 hr).

Data reduction and statistical analyses

Summary statistics were performed for each period (means, standard deviations, medians, minimum, maximum; and geometric means for fecal coliforms). Special attention was paid to those parameters having NCDEQ standards to determine if they were in exceedance. Other parameters lacking numeric standards, such as nutrients, were assessed to determine if the samples exceeded typical blackwater

Table 1 | Sampling locations on Stocking Head Creek including coordinates and description

Station	Coordinates	Description
SHC-GDR	N34.91197, W77.94507	Stocking Head Creek (SHC) at Graham Dobson Rd (GDR) – the uppermost branch of the creek, with upstream CAFOs and sprayfields present
TR-CSR	N34.90279, W77.94440	Stocking Head Creek tributary at Cool Springs Rd (CSR) – the upper first-order tributary of the creek; no immediately adjoining CAFOs, but CAFOs are near the creek upstream
SHC-SDCR	N34.89796, W77.93628	Stocking Head Creek at South Dobson Chapel Rd (SDCR) – numerous CAFO sprayfields, and grazing cattle near creek
TR-SDCR	N34.88878, W77.94453	First-order tributary entering Stocking Head Creek at South Dobson Chapel Rd (SDCR) – site periodically influenced by lagoon waste spraying
SHC-SHCR	N34.88710, W77.91124	Stocking Head Creek at Stocking Head Creek Rd – CAFO sprayfields immediately adjacent to creek
SHC-50	N34.87950, W77.89441	Stocking Head Creek at SR 50 – site near a wetland area hydrologically connected to the creek; Maxwell Creek joins Stocking Head just downstream as well
SHC-PBR	N34.87043, W77.86539	Stocking Head Creek at Pasture Branch Rd (PBR) – this reach has no CAFOs in the immediate vicinity, is downstream of the Maxwell Creek entry point, and was the farthest downstream sampling location

stream concentrations according to our laboratory's extensive database and the published literature.

The spring sampling period occurred during March 8–30, while the summer sampling period occurred during August 9–September 7. Thus, the stream during the early period would have been affected by little if any recent spraying, whereas the summer sampling period would have followed up to six months of accumulated swine waste application to sprayfields. To determine if season matters when assessing a stream for CAFO impacts, we tested potential response variables between the spring and summer seasons. Nutrient and fecal coliform data were log-transformed and Student's *t*-tests were used to test selected parameter concentrations between spring and summer sampling periods ($p < 0.05$). To investigate the potential for rainfall and runoff influencing parameter concentrations, we accessed the Weather Underground website (www.wunderground.com) and obtained daily meteorological data from Clinton, NC, 36 km distant from the sampling area (the nearest site with complete data) and totaled cumulative rain that fell on the day of sampling plus the rain that fell in the preceding 48 hr (Rain48). Average Rain48 was calculated for spring sampling dates and compared with summer sampling dates using a *t*-test. Further, log-transformed pollutant concentrations for each station and date were correlated against Rain48 to see if any significant ($p < 0.05$) relationship existed.

RESULTS AND DISCUSSION

Spring water temperatures ranged from 12.1 to 18.9°C and summer water temperatures ranged from 24.5 to 27.7°C. There were no unusual variations and no differences in temperature among stations. Most sampling events reflected circumneutral pH conditions ranging from 6.6 to 7.2. Field turbidity was generally low; the tributary site TR-SDCR had the highest mean turbidity of 23 nephelometric turbidity units (NTU) and the highest individual turbidity of 38 NTU. Sandy sediments dominated at several of the sites, so high turbidity would not have been expected to be a widespread issue. Dissolved oxygen concentrations in spring ranged from 7.3 to 10.8 mg/L, and in summer ranged from 4.0 to 7.8 mg/L.

Conductivity reflects the amount of dissolved material within the water column, and due to the predominant soils, the conductivity of Coastal Plain blackwater streams is normally low (Smock & Gilinsky 1992). Conductivity at the seven sites ranged from 95 to 440 mS/cm, with lowest values seen in the downstream station SHC-PBR, likely reflecting swamp water inputs from adjoining wetlands. The highest levels were found at tributary station TR-CSR. Mean conductivity of all sites in spring was 204 ± 89 mS/cm versus 270 ± 61 mS/cm in summer, a highly significant difference ($p < 0.001$); median values were similar to means (Figure 2(a)). In a large-scale study of streams impacted by CAFOs compared with controls (Harden

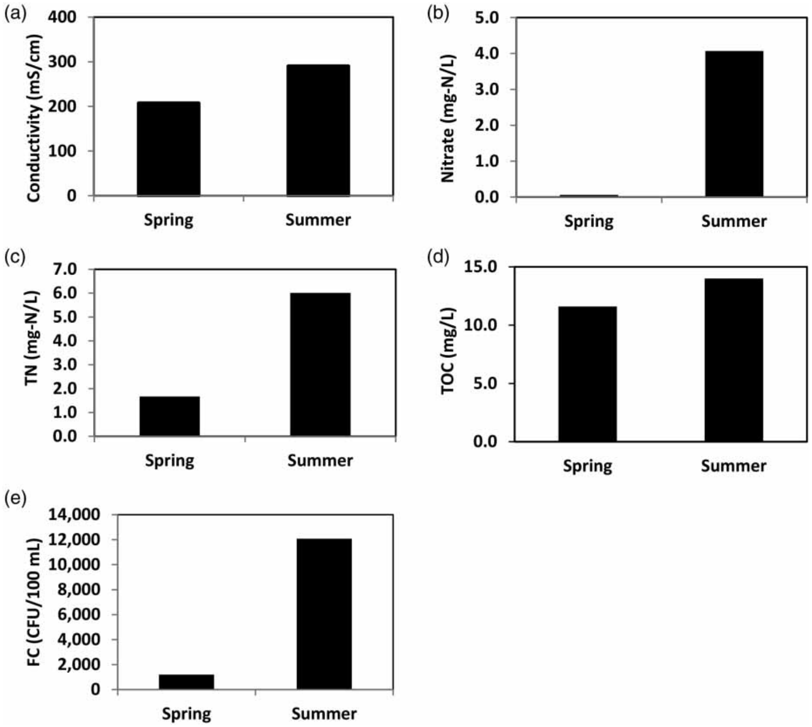

Figure 2 | Statistically significant ($p < 0.01$) seasonal differences in pollutant parameters for Stocking Head Creek spring and summer 2016 samples: (a) median conductivity values; (b) median nitrate concentrations; (c) median TN concentrations; (d) median TOC concentrations; (e) geometric mean fecal coliform bacteria concentrations. $N = 35$ for both spring and summer in all cases.

2015), conductivity was significantly elevated in CAFO-impacted streams compared with non-impacted streams, with the increase attributed to elevated dissolved magnesium, potassium, sodium, and chloride concentrations.

Ammonium in Stocking Head Creek during spring ranged from the detection limit (0.10 mg/L) to 4.17 mg/L (mean = 0.50 mg/L). Highest ammonium concentrations were found at Station SHC-50 and Station SHC-SHCR. Ammonium concentrations in summer ranged from 0.10 to 1.30 mg-N/L (mean = 0.33 mg/L), with highest concentrations at SHC-50 and TR-SDCR. Stations TR-SDCR and SHC-SHCR are the sites nearest to sprayfields. The lowest ammonium concentrations were found farthest downstream at SHC-PBR. Upstream from this site there is a wetland area in association with the stream, and the stream is also joined by another stream which likely creates a dilution effect. As a comparison, ammonium concentrations less than 0.50 mg/L have been demonstrated to stimulate algae blooms in water from blackwater streams (Mallin *et al.* 2004). The average ammonium concentrations found at the sites were well in excess of ammonium concentrations found in many other streams in the Northeast Cape Fear and Black River watersheds (Mallin *et al.* 2004, 2006). During a 2013 summer–fall study of this watershed, there were several sharp peaks in ammonium ranging up to 38 mg ammonium-N/L, particularly at sites adjoining sprayfields (Mallin *et al.* 2015). In the present study there was no significant difference in average ammonium concentrations between spring and summer.

Nitrate concentrations in Stocking Head Creek were low to moderate in spring (mean 0.09 ± 0.10 mg-N/L, $n = 35$) and very high in summer (mean 6.54 ± 3.71 mg-N/L, $n = 35$), a significant ($p < 0.001$) seasonal difference (Table 2; Figure 2(b)). Nitrate in spring ranged from 0.01 to 0.37 mg-N/L, whereas in summer it ranged from 0.10 to 11.30 mg nitrate-N/L. High nitrate concentrations were measured at various sites (Table 2), some of which were well downstream from sprayfields, including SHC-GDR, TR-CSR, SHC-SDCR, SHC-SHCR, and SHC-50. The 2013 summer–fall study of this stream showed nitrate concentrations very similar to the concentrations measured during summer 2016 (Mallin *et al.* 2015). Additionally, the maximum nitrate concentrations found in summer were

Table 2 | Nitrate concentrations (as mg-N/L) for stocking head creek individual stations, spring vs summer collections, presented as mean ± SD; ranges are also shown

	Sampling period	
Station	**March**	**August**
SHC-GDR	0.15 ± 0.08 0.06–0.25	9.70 ± 1.06 8.67–11.30
TR-CSR	0.21 ± 0.14 0.06–0.37	7.55 ± 3.49 1.85–11.10
SHC-SDCR	0.04 ± 0.01 0.01–0.10	4.85 ± 2.31 1.87–8.32
TR-SDCR	0.02 ± 0.03 0.01–0.07	0.81 ± 0.82 0.10–2.12
SHC-SHCR	0.10 ± 0.09 0.01–0.21	4.48 ± 2.95 1.02–8.97
SHC-50	0.11 ± 0.10 0.01–0.24	4.33 ± 2.60 0.91–8.12
SHC-PBR	0.01 ± 0.01 0.01–0.03	0.55 ± 0.21 0.23–0.80

Table 3 | TN concentrations (as mg-N/L) for stocking head creek individual stations, spring vs summer collections, presented as mean ± SD; ranges are also shown

	Sampling period	
Station	**March**	**August**
SHC-GDR	1.81 ± 0.24 1.50–2.10	10.80 ± 1.13 9.80–12.50
TR-CSR	1.59 ± 0.34 1.30–2.17	8.50 ± 3.48 2.80–12.20
SHC-SDCR	1.09 ± 0.52 0.26–1.68	5.98 ± 2.41 2.80–9.30
TR-SDCR	1.24 ± 1.10 0.80–3.80	2.02 ± 2.92 1.30–3.60
SHC-SHCR	2.34 ± 0.62 1.50–3.20	5.62 ± 2.99 2.10–10.10
SHC-50	3.01 ± 1.97 1.30–6.24	6.08 ± 2.45 2.70–9.50
SHC-PBR	1.17 ± 0.19 1.01–1.43	1.40 ± 0.28 1.00–1.80

similar to the maximum concentrations found in CAFO-impacted streams during a large-scale study in North Carolina (Harden 2015). Nitrate concentrations of 0.50 mg/L (ppm) and greater have been demonstrated to stimulate algal blooms in water from blackwater streams and rivers and cause significant increases in BOD (Mallin *et al.* 2004). There is a federal well water standard of 10 mg/L to prevent blue-baby syndrome (methemoglobinema), which was exceeded twice in this study.

Nitrogen concentrations are generally low in pristine blackwater streams, due to both low concentrations in soils and retention of nutrients in the floodplain (Smock & Gilinsky 1992). In Stocking Head Creek, by contrast, TN concentrations were very high, especially in summer (Figure 2(c)). In spring 2016, TN ranged from 0.26 to 6.24 mg-N/L, whereas in summer it ranged from 1.0 to 12.50 mg-N/L (Table 3). The stations with highest overall TN concentrations were the same as those with highest nitrate concentrations. For perspective, using a large dataset of 1,070 streams, Dodds *et al.* (1998) determined that TN concentrations >1.5 mg/L were characteristic of eutrophic (nutrient-enriched) conditions. During both spring and fall, mean nitrate concentrations exceeded that eutrophication indicator at most sampling locations (Table 3).

In spring, the TN values on average were dominated by organic nitrogen, which comprised about 67% of the TN. By contrast, in summer, TN was dominated by dissolved inorganic nitrogen (DIN, i.e., nitrate plus ammonium) representing 86% of the TN on average. In the spring, nitrate comprised only 5% of average TN, but in summer, nitrate comprised 80% of the average TN overall. A large-scale study of Wisconsin streams found that those with TN primarily in the form of nitrate were strongly human-impacted watersheds, especially agriculturally (Stanley & Maxted 2008). In unimpacted blackwater streams of North Carolina, inorganic N usually comprises only a small percentage of TN (Mallin *et al.* 2004, 2006), thus, N sourced from animal manure is clearly dominant in summer in Stocking Head Creek. As with nitrate, there was a large difference in TN concentrations in Stocking Head Creek between spring and summer samples (Figure 2(c)). Mean summer concentrations (5.77 ± 3.72 mg-N/L, $n = 35$ samples) were significantly ($p < 0.001$) higher than mean spring concentrations (1.58 ± 1.07 mg-N/L, $n = 35$ samples).

Orthophosphate concentrations in spring ranged from 0.05 to 1.08 mg-P/L, with station means at 0.07–0.38 mg-P/L. In summer orthophosphate concentrations ranged from 0.04 to 1.19 mg-P/L, with station means at 0.11–0.33 mg-P/L. There was no significant ($p > 0.05$) difference between spring and summer mean concentrations. In spring, TP ranged from 0.100 to 1.67 mg-P/L (mean = 0.20 mg/L), with station means at 0.14 (station TR-CSR) to 0.48 mg-P/L (station TR-SDCR). In summer, TP ranged from 0.11 to 1.19 mg-P/L (mean = 0.27 mg/L), with station means at 0.11 to 0.40 mg-P/L. There was no significant ($p < 0.05$) difference between spring and summer mean concentrations. Stations TR-SDCR, SHC-SHCR, and SHC-50 generally maintained the highest concentrations. Using data from 1,366 streams, Dodds *et al.* (1998) concluded that TP concentrations >0.075 mg/L were characteristic of eutrophic streams, thus, Stocking Head Creek and its tributaries would be considered eutrophic by that criterion.

Spring TOC concentrations ranged from 8.9 to 19.5 mg/L (mean = 12.6 mg/L), while summer concentrations ranged from 11.3 to 34.2 mg/L (mean = 14.2 mg/L); means were significantly different at $p = 0.008$. Spring station means ranged from 10.1 to 14.1 mg/L, whereas summer station means ranged from 12.8 to 18.2 mg/L. Median TOC concentrations in spring and summer were 11.6 mg/L and 14.0 mg/L, respectively (Figure 2(d)).

The state of North Carolina uses fecal coliform bacterial densities as a proxy for potentially pathogenic bacteria in freshwaters. Potential sources include human sewage, wildlife, and livestock including cattle, swine, and poultry. As noted, in this watershed, cattle and human fecal sources are at low densities, but confined swine and poultry densities are very high. The NC protocol for sampling and means for determining fecal impairment of a water body is explained above.

Fecal coliform counts for Stocking Head Creek in spring and summer 2016 were, in general, very high and place this creek clearly as one impaired per the State of North Carolina definition. In spring, geometric mean fecal coliform counts were in the thousands at five of seven sites. Spring geometric mean fecal coliform counts exceeded 200 CFU/100 mL at six of seven sites; 86% of the samples collected exceeded 200 CFU/100 mL, and 80% of the samples exceeded 400 CFU/100 mL. In summer, geometric mean fecal coliform counts exceeded 200 CFU/100 mL at seven of seven sites; 100% of the samples collected (35/35) exceeded 200 CFU/100 mL and 97% of the samples (34/35) exceeded 400 CFU/100 mL within a 30-day period.

While the stream was polluted by fecal bacteria in both seasons, there was a large disparity in fecal coliform counts between seasons (Table 4; Figure 2(e)). Overall summer counts (geometric mean 12,080 CFU/100 mL; $n = 35$ samples) were significantly ($p < 0.001$) higher than spring counts (geometric mean 1,195 CFU/100 mL, $n = 35$ samples).

Rainfall considerations

Based on three statistical techniques, rainfall did not drive the seasonal pollutant patterns. First, average Rain48 (rain on day of sample plus previous 48 hr) for the spring sampling dates was 7.31 ± 10.01 mm vs average Rain48 for the summer dates of 7.72 ± 12.49 mm (no significant difference). Second, there were no significant ($p > 0.05$) positive or negative correlations between concentrations of nitrate-N, ammonium-N, and fecal coliform bacteria compared to rainfall concentrations. Finally, in a 2013 study of this creek (Mallin *et al.* 2015), we analyzed whether or not rainfall produced higher pollutant parameter concentrations than occurred on non-rain periods. During that study measurable rainfall occurred either on the day of sampling or within the 48 hours preceding the sample day on five of ten sampling occasions. *T*-tests were used to test ammonium, nitrate, and fecal coliform concentrations between wet and dry periods. There were no significant ($p > 0.05$, df = 68) differences in means between wet and dry sample dates for any of the three parameters tested. Thus, we conclude that the large seasonal disparity in pollutant concentrations in this CAFO-rich stream was not due to rainfall.

Table 4 | Fecal coliform bacteria concentrations (as CFU/100 mL) at individual stations in spring vs summer 2016

	Sampling period	
Station	**March**	**August**
SHC-GDR	1,635 1,090–2,300	11,887 1,550–60,000
TR-CSR	3,948 1,730–13,000	14,413 8,000–60,000
SHC-SDCR	1,347 637–8,000	16,436 2,800–60,000
TR-SDCR	982 145–31,000	22,028 4,900–60,000
HC–SHCR	1,711 650–10,000	16,450 1,360–60,000
SHC-50	1,412 330–7,000	14,178 3,900–60,000
SHC-PBR	169 73–1,910	2,595 390–34,000

Data are given as geometric means; ranges are also shown. For samples too numerous to count, the maximum is conservatively set at 60,000 CFU/100 mL (per laboratory protocol).

Spray season influence

As noted earlier, legal animal waste spraying on Bermuda grass fields extends from March 1 through September 30 (North Carolina Department of Agriculture and Consumer Services 1996). The March samples would therefore have been expected to reflect minimal spraying influence, while the summer samples would have been expected to reflect several months' worth of sprayed waste applications. As such, some pollutant or response variables indeed were at significantly higher levels in summer than in spring. Higher conductivity during summer indicates more solutes added to the stream in general (Figure 2(a)). Ammonium was mostly elevated at the stations adjoining sprayfields but not at sites away from such fields, whereas nitrate was highly elevated at sites well away from sprayfields (Table 2). Nitrate in this watershed is, in part, a product of the nitrification of ammonium originating from the spraying of swine waste that has been stored in lagoons. Spraying ammonia-rich liquid waste into the air and onto the nearby soils oxidizes the waste and exposes it to nitrifying soil bacteria. From there the nitrate can migrate readily downward through the porous sandy soils of the Coastal Plain (Keeney 1986; National Resources Conservation Service 2014).

Nitrate in particular, as well as TN, showed much higher concentrations in summer than in spring (Figure 2(b) and 2(c)) and would have entered the stream and its tributaries as surface runoff and as subsurface groundwater (Mallin *et al.* 2015). In other areas, it has similarly been observed that both spreading of waste on the landscape and spraying of waste will lead to excessive nitrate in groundwaters (Liebhardt *et al.* 1979). Organic carbon makes up some portion of swine lagoon waste (as well as poultry waste) and, in a previous study, was strongly positively correlated with biochemical oxygen demand (Mallin *et al.* 2015). TOC also was significantly higher in summer than in spring (Figure 2(d)), presumably as a result of swine waste spraying in late spring and summer. Summer is a critical period for Coastal Plain streams due to elevated water temperatures and naturally lowered dissolved oxygen (Smock & Gilinsky 1992). Thus, further additions of BOD pollutants to streams in summer lead to downstream increases in BOD and lowering of DO in downstream water bodies, such as the Northeast Cape Fear River. Both TOC and TN were positively correlated with BOD in the earlier study of Stocking Head Creek (Mallin *et al.* 2015).

The inter-seasonal differences in fecal coliform densities were very large (Figure 2(e)). Whereas the spring geometric mean (1,195 CFU/100 mL) and median (1,369 CFU/100 mL) densities were high (six-fold higher than the 200 CFU/100 mL standard), the summer geometric mean (12,080 CFU/100 mL) and median (11,000 CFU/100 mL) far exceeded the spring densities. This disparity raises a serious sampling concern. If regulatory agencies are constrained to sample on only one or two occasions per year, sampling in the winter or early spring is likely to significantly underestimate the actual pollution levels (and human health and ecological threats) caused by industrial animal production facilities. The data demonstrate that late summer sampling is the optimal window for detection of CAFO waste pollution impacts on stream waters.

CONCLUSIONS

The 2016 sample sets (five dates, seven sites within 30 days, performed in both spring and summer) indicates that Stocking Head Creek is highly polluted by fecal bacteria during both spring and summer. Summer concentrations were significantly higher than spring concentrations. Additionally, there were significant seasonal differences in other pollutant concentrations, with much higher summer conductivity, nitrate, TN, and TOC reflecting the seasonal waste application practices. Thus, sampling schemes for streams in swine CAFO impacted watersheds should consider seasonal animal waste spraying regulations to obtain representative data on waste disposal impacts to streams.

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REFERENCES

Arfken, A. M., Mallin, M. A., Cahoon, L. B. & Song, B. 2013 *Monitoring Swine Fecal Contamination in the Cape Fear River Watershed Based on the Detection and Quantification of Hog-Specific Bacteroides-Prevotella 16S rRNA Genes*. Report No. 436. Water Resources Research Institute of the University of North Carolina.

Burkholder, J. M., Libra, B., Weyer, P., Heathcote, S., Kolpin, D., Thorne, P. S. & Wichman, M. 2007 Impacts of waste from concentrated animal feeding operations on water quality. *Environmental Health Perspectives* **115** (2), 308–312.

Cahoon, L. B. & Ensign, S. H. 2004 Spatial and temporal variability in excessive soil phosphorus levels in eastern North Carolina. *Nutrient Cycling in Agroecosystems* **69** (2), 111–125.

Cahoon, L. B., Mickucki, J. A. & Mallin, M. A. 1999 Nutrient imports to the Cape Fear and Neuse River basins to support animal production. *Environmental Science and Technology* **33** (3), 410–415.

Dodds, W. K., Jones, J. R. & Welch, E. B. 1998 Suggested classification of stream trophic state: distributions of temperate stream types by chlorophyll, total nitrogen, and phosphorus. *Water Research* **32** (5), 1455–1462.

Ham, J. M. & DeSutter, T. M. 2000 Toward site-specific design standards for animal-waste lagoons: protecting groundwater quality. *Journal of Environmental Quality* **29** (6), 1721–1732.

Harden, S. L. 2015 *Surface-water Quality in Agricultural Watersheds of the North Carolina Coastal Plain Associated with Concentrated Animal Feeding Operations*. US Geological Survey, Scientific Investigations Report 2015-5080. http://dx.doi.org/10.3133/sir20155080.

Keeney, D. 1986 Sources of nitrate to ground water. *CRC Critical Reviews in Environmental Control* **16** (3), 257–304.

Liebhardt, W. C., Golt, C. & Tupin, J. 1979 Nitrate and ammonium concentrations of ground water resulting from poultry manure applications. *Journal of Environmental Quality* **8** (2), 211–215.

Mallin, M. A. 2000 Impacts of industrial-scale swine and poultry production on rivers and estuaries. *American Scientist* **88**, 26–37.

Mallin, M. A. & Cahoon, L. B. 2003 Industrialized animal production – a major source of nutrient and microbial pollution to aquatic ecosystems. *Population and Environment* **24** (5), 369–385.

Mallin, M. A., McIver, M. R., Ensign, S. H. & Cahoon, L. B. 2004 Photosynthetic and heterotrophic impacts of nutrient loading to blackwater streams. *Ecological Applications* **14** (3), 823–838.

Mallin, M. A., Johnson, V. L., Ensign, S. H. & MacPherson, T. A. 2006 Factors contributing to hypoxia in rivers, lakes and streams. *Limnology and Oceanography* **51**, 690–701.

Mallin, M. A., McIver, M. R., Robuck, A. R. & Dickens, A. K. 2015 Industrial swine and poultry production causes chronic nutrient and fecal microbial stream pollution. *Water, Air and Soil Pollution* **226** (12), 407. DOI: 10.1007/s11270-015-2669-y.

National Resources Conservation Service (NRCS) 2014 *Web Soil Survey*. NRCS, US Department of Agriculture, Washington, DC. On-line tool available at: http://websoilsurvey.nrcs.usda.gov/app/.

North Carolina Department of Agriculture and Consumer Services (NCDACS) 1996 *Senate Bill 1217 Interagency Guidance Documents, Appendix 1.1A*. Division of Soil and Water Conservation, NCDACS, Raleigh, NC.

North Carolina Department of Environment and Natural Resources (NC DENR) 1999 *Administrative Code Section: 15A NCAC 2B .0200 Classifications and Water Quality Standards Applicable to Surface Waters and Wetlands of North Carolina*. Division of Water Quality, NC DENR, Raleigh, NC.

Patt, H. 2017 *A Comparison of PAN and P2O5 Produced From Poultry, Swine and Cattle Operations in North Carolina*. Division of Water Resources, North Carolina Department of Environmental Quality.

Rajbhandari, N. B., Pradham, S., DiLuzio, M., Kebede, A. & Baker, V. 2015 Identifying nutrient contributors in North Carolina's Coastal Plain rivers. *American Journal of Environmental Sciences* **11** (4), 313–324.

Smock, L. A. & Gilinsky, E. 1992 Coastal plain blackwater streams. In: *Biodiversity of the Southeastern United States* (C. T. Hackney, S. M. Adams & W. H. Martin, eds). John Wiley and Sons, Inc. New York, NY.

Stanley, E. H. & Maxted, J. T. 2008 Changes in the dissolved nitrogen pool across land cover gradients in Wisconsin streams. *Ecological Applications* **18** (7), 1579–1590.

United States Department of Agriculture (USDA) 2014a *2012 Census of Agriculture*. Natural Resources Conservation Service, USDA. Available at: http://www.agcensus.usda.gov.

USDA 2014b *Official Soils Descriptions*. United States Department of Agriculture, Natural Resources Conservation Service, Soil Survey Division. Available at: http//soilseries.sc.egov.usda.gov/osdname.asp.

US EPA 2014 United States Environmental Protection Agency, National Pollutant Discharge Elimination system (NPDES). Available at: http://cfpub1.epa.gov/npdes/search.cfm.

Wing, S., Cole, D. & Grant, G. 2000 Environmental injustice in North Carolina's hog industry. *Environmental Health Perspectives* **108** (3), 225–231.

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