Fecal bacteria contamination of floodwaters and a coastal waterway from tidally-driven stormwater network inundation

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Key Points:

- Daily observations of *Enterococcus spp.* concentrations in a coastal waterway were similar during and outside perigean spring tides.
- Tidal inundation of stormwater networks occurred daily, but rainfall runoff produced the greatest bacterial contamination in the waterway.
- High *Enterococcus spp.* concentrations were observed in roadway floodwaters and the receiving waterway during perigean spring tides.

Abstract

Inundation of coastal stormwater networks by tides is widespread due to sea-level rise (SLR). The water quality risks posed by tidal water rising up through stormwater infrastructure (pipes and catch basins), out onto roadways, and back out to receiving water bodies is poorly understood but may be substantial given that stormwater networks are a known source of fecal contamination. In this study, we (1) documented temporal variation in concentrations of *Enterococcus spp*. (ENT), the fecal indicator bacteria standard for marine waters, in a coastal waterway over a two-month period and more intensively during two perigean spring tide periods, (2) measured ENT concentrations in roadway floodwaters during tidal floods, and (3) explained variation in ENT concentrations as a function of tidal inundation, antecedent rainfall, and stormwater infrastructure using a pipe network inundation model and robust linear mixed effect models. We find that ENT concentrations in the receiving water body vary as a function of tidal stage and antecedent rainfall, but also site-specific characteristics of the stormwater network that drains to the waterbody. Tidal variables significantly explain measured ENT variance in the waterway, however, runoff drove higher ENT concentrations in the receiving waterway. Samples of floodwaters on roadways during both perigean spring tide events were limited, but all samples exceed thresholds for safe public use of recreational water. These results indicate that inundation of stormwater networks by tides could pose public health hazards in receiving water bodies and on roadways, which will likely be exacerbated in the future due to continued SLR.

Plain Language Summary

Communities along the US Atlantic Coast have begun to experience the effects of sea-level rise as stormwater drainage pipes, originally designed when sea levels were lower, are now routinely inundated at high tide. We do not know whether frequent inundation of stormwater drainage pipes affects coastal water quality, specifically the fecal bacteria levels of coastal waters. In this study, we collected daily measurements of *Enterococcus spp.* (ENT), the bacteria used by regulators to assess the safety of marine waters for swimmers and recreators, from a tidal creek over a two-month period. We also collected water samples during the highest high tides from roadway floodwaters and the adjacent tidal creek as the stormwater pipes drained during the outgoing tide. After data collection, we developed statistical models to relate the amount of ENT in the tidal creek to tide, rainfall, and stormwater infrastructure. When comparing rainfall and tide, we observed that rainfall caused greater bacterial contamination in the tidal creek than high tide flooding. However, we observed high ENT concentrations in roadway floodwaters and in the creek during the outgoing tide, leading us to conclude that everyday flooding of underground stormwater pipes at high tide could create hazardous water quality.

1 Introduction

Since 1880, global sea level has risen between 21 and 24 cm due to the melting of land-based ice and thermal expansion of ocean water (Sweet et al., 2022). On the United States' South Atlantic and Gulf coasts, sea levels locally have risen at even higher rates (approximately 10 mm $year^{-1}$) due to internal climate variability (e.g., increased wind-driven ocean circulation) and local land elevation changes (i.e., subsidence and isostatic rebound) (Dangendorf et al., 2023).

This accelerated rate of local (relative) sea-level rise (SLR) poses a threat to coastal communities, many which were built on low-lying land. Specifically, for many of these coastal

communities, stormwater infrastructure, like subterranean pipe networks, ditches, and catch basins, was built decades ago when sea levels were lower. Increased water levels have allowed tides to propagate up into stormwater networks and overtop low elevation areas, causing coastal communities to flood in what is referred to as "sunny day flooding" or "high tide flooding". Regular tidal inundation of stormwater networks has become widespread in coastal areas (Gold et al., 2022), and persistent increases in SLR are projected to increase the frequency and magnitude of high tide flooding nationally (Sweet et al., 2022).

As tidal floods increase in frequency and magnitude, aging and faulty stormwater infrastructure may pose a public health risk by creating water quality hazards (Allen et al., 2019). Stormwater networks can act as reservoirs for fecal contaminants, as exfiltrated sewage (Hart et al., 2020; Olds et al., 2018), biofilms (Burkhart, 2013), or septic leachate (Converse et al., 2011) can be present within the network. Frequent inundation by tidal waters could transport fecal contaminants from stormwater networks to roadways during incoming or flood tides, and flush pollutants into coastal waters during outgoing or ebb tides. Other factors, like stormwater runoff (Shen et al., 2019; Gold et al., 2023) and high groundwater tables (Befus et al., 2023; Habel et al., 2020), can compound with tides and reduce the volume capacity of stormwater networks, leading to more extensive or frequent flooding of coastal communities.

In coastal waters, fecal contamination is often measured by *Enterococcus spp.* (ENT) concentrations. ENT are gram-positive cocci bacteria typically found in the intestines of warm-blooded mammals. Because of their origin in intestines, ENT are used as a fecal indicator organism in marine and beach water quality standards from the U.S. Environmental Protection Agency (EPA, Recreational Water Quality Criteria, 2012). ENT concentration is a proxy measure for gastrointestinal illness risk in marine waters (Wade et al., 2021).

Increased attention has been dedicated to tidal inundation as a transport mechanism of fecal contamination through stormwater infrastructure inundation (Hart et al., 2020; Price et al., 2021), overtopping of coastal infrastructure (Macías-Tapia et al., 2021, 2023), and groundwater inundation (McKenzie et al., 2021). Hart et al. (2020) measured human-specific microbial source tracking markers in a municipal separate stormwater system (MS4) in Beaufort, NC, during compound high tide and storm conditions, proposing that diluted sewage could be present in standing water on the roadway when high tide floods occur. This work was continued by Price et al. (2021), who determined that the highest concentrations of fecal indicator bacteria (FIB), including ENT, in stormwater discharge were present in receding tides during nineteen sampling events encompassing both storm and ambient conditions. Macías-Tapia et al. (2021, 2023) reported consistent exceedances of ENT in tidal floodwaters of the Lafayette River in Norfolk, VA, during five annual perigean spring tide flooding events between 2017 and 2021. Perigean spring tides occur during the coincidence of perigee – when the moon is closest to the Earth in its orbit and therefore exerts a large gravitational pull resulting in higher than average tides – and spring tide (during a new or full moon, when the Earth, moon, and sun are in alignment). Because perigean spring tides produce some of the highest water levels of the year, they can result in coastal flooding of low-lying areas. The work of Macías-Tapia et al. (2021, 2023) determined that in Norfolk, loads varied between perigean spring tide floods but no relationship between ENT abundance and floodwater extent could be determined (Macías-Tapia et al., 2023). Lastly, McKenzie et al. (2021) monitored coastal and canal groundwater and surface water and storm drains during high tide events and reported the presence of wastewater discharge in the collected samples through measurement of radon and wastewater tracers. Combined, these

studies provide insight into the potential role of tidal flooding as a mechanism for fecal contamination in coastal waters. However, to understand how stormwater network inundation by tides is a driver of coastal fecal contamination, longitudinal data are needed to understand how contaminant dynamics vary across locations, over time, and with flood events of different drivers (e.g., tides and rainfall) and magnitudes.

In this study, we assessed the potential for fecal contamination in an urban, coastal waterway that experiences tidal floods and daily inundation of its underground stormwater network by tides. Specifically, our objectives were to (1) document temporal variation in fecal bacteria concentrations in a coastal waterway over multiple flood events and under baseline conditions, (2) measure fecal bacteria concentrations in roadway floodwaters during tidal flood events, and (3) explain variation in bacteria concentrations as a function of tidal inundation, antecedent rainfall, and stormwater infrastructure. Since fecal contaminants are highly variable in space and time (Boehm, 2007), we collected observational data at three locations daily over a two month period, and more intensively during nine tidal cycles coinciding with two perigean spring tide events (i.e., some of the highest tide events of the year, when the moon is closest to the earth). These measurements, in conjunction with a simplified pipe network model and linear regression, were used to explain variation in bacteria concentrations in the waterway as a function of tidal inundation, antecedent rainfall, and stormwater infrastructure.

2 Methods

2.1 Site Description

Our study site was Beaufort, North Carolina, USA, a small, coastal town established in the 1700s. Beaufort sits on a peninsula between the Newport River and North River (to the west and east, respectively) and north of Taylor's Creek, a tidal channel that empties through an inlet to the Atlantic Ocean. We focused our sampling efforts on Taylor's Creek, which is the receiving waterbody that is adjacent to the downtown area, and on the roadway closest to the waterway Front Street (**Fig 1a**). The tides in Taylor's Creek are semi-diurnal with a tidal range of 0.95 m (NOAA Tides and Currents Station ID: 8656483, located in Beaufort, NC). In 2016, Taylor's Creek was listed as a 303(d) impaired waterway for ENT by the North Carolina Division of Environmental Quality, but was later delisted in 2022 after stormwater network improvements (NCDEQ, 2022 North Carolina 303(d) List, 2023).

Much of the shoreline in the downtown area is armored with bulkheads, which protects roadways and other low-lying areas from high water levels in Taylor's Creek. Stormwater is conveyed to Taylor's Creek via a MS4, consisting of subterranean pipes and several outfalls within our sampling area (**Fig 1b**). Roadway flooding is monitored on Front Street by water level gauges located within the stormwater network, as well as pole-mounted cameras (Gold et al., 2023). These data have shown that tides inundate the stormwater network on a daily basis; during perigean spring tide events, water can rise all the way up through the network and result in flooding of roadways. During regular tides, flooding can still occur due to the compounding effects of rainfall, as there is limited capacity for pipes to convey even small amounts of rainfall as a result of tidal water volumes occupying some of the capacity (Gold et al., 2023).

Two spring tides (here, full moons) coinciding with a moon orbit in perigee occurred during the study period on June 14 and July 13, 2023 (NOAA, 2022). During the highest-high tides associated with these perigean spring tide events, tidal waters propagated up through the

stormwater network and produced minor roadway flooding nine times. No concomitant rainfall occurred during the highest-high tides of the two perigean spring tide events, so that the floodwaters were produced from tidal influence only. Rainfall occurred during the higher low tide on July 15, 2022, and produced a compound flood event (i.e., rainfall coincided with tidal inundation).

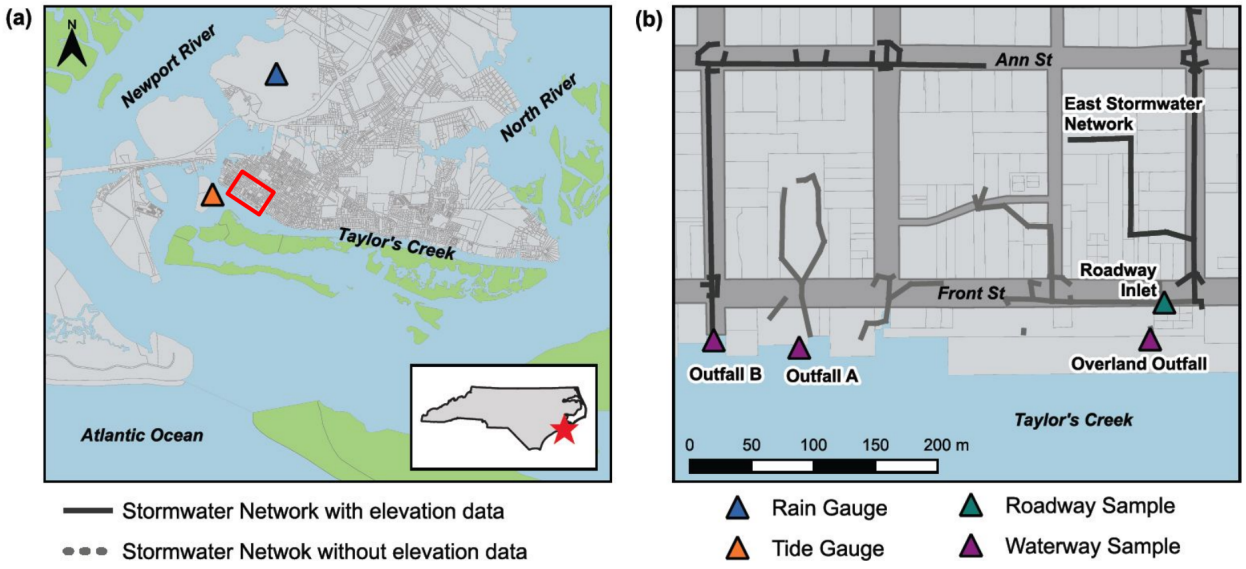


Figure 1. Study map of Beaufort, NC (USA) with an inset map showing the location of Beaufort within North Carolina as a red star (**a**) and the four stormwater networks in the study area (**b**). In (**a**), the red rectangle indicates the study area. The green land area represents natural areas, the gray land area represents developed areas, and the thin gray lines delineate land parcels. The NOAA tide gauge (orange triangle), which informed sampling times and provided data for tidal variables, is located across Taylor's Creek approximately 300 m from the Outfall B sampling location. The rain gauge (blue triangle) is located north of the study site. In (**b**), thick gray lines show the underground stormwater pipe network, with the shade of gray indicating whether there was elevation data available (dark and light for networks with known and unknown elevation, respectively). Waterway samples were collected from Taylor's Creek at three locations (purple triangles) within 2 meters of stormwater network outfalls. Roadway flood samples were collected directly above a single stormwater catchment grate in the East network (teal triangle).

2.2 Data Collection

Water quality samples were collected for enumeration of ENT concentrations, which is a proxy measure for fecal contamination. Collection occurred at three waterway locations in Taylor's Creek (**Fig 1b**). The "Overland Outfall" sampling location is near a bulkhead spillway that conveys stormwater runoff from a parking lot and is not connected to the subterranean pipe network. The overland outfall is relatively high in elevation and was not observed to be inundated during our study period. The "Outfall A" drains a relatively small area (**Fig 1b**), including a parking lot around a museum and catch basins on Front Street. The "Outfall B" sampling location drains a relatively large area, including a residential street and catch basins on Front Street and Ann Street (**Fig 1b**). The Outfall B sampling location was added on June 14, 2022, a week after sampling was initiated at the Overland Outfall and the Outfall A locations.

Outfall elevations were only available for the Outfall B, which is positioned at -0.74 m NAVD88 or -0.30 m Mean Higher-High Water (MHHW). The MHHW datum is the mean of the highest diurnal tide, and is useful for identifying extremes in tidal heights. Water levels greater than the 0 m MHHW datum are deviations of tidal heights above the average high tide. At this elevation, the Outfall B is inundated by tides on a daily basis.

Collection of water samples in Taylor's Creek, proximate to the three outfall locations described above, occurred daily for 58 days from June 6 to August 2, 2022. This allowed for characterization of fecal contamination during both baseline conditions and perigean spring tides. Baseline sampling of ENT occurred daily between 8:00-9:00 AM Eastern Time to prevent ENT levels from being affected by UV inactivation from direct sunlight, which peaks around midday. Given the two-month long sampling period, the daily samples also captured every tidal stage (low, rising, high, falling tide). Perigean spring tide sampling of ENT occurred on June 12-16 and July 11-15, 2022, when estimated water levels were above the local risk-based threshold. During these high water level events, we increased the sampling frequency to include measurements at high tide, ebb tide (approximately halfway between high and low tide), and low tide, with measurement times scheduled based on predicted water levels at the nearby NOAA Tides and Currents Station (NOAA, ID 8656483; Fig 1). This sampling frequency was selected to capture the change in fecal contamination conditions from the maximum stormwater inundation (at highest high tide) and during the draining of the stormwater networks (during the ebb tide and at the highest low tide). During these floods, we also sampled roadway floodwaters at a stormwater grate in the easternmost stormwater network of the downtown area, referred to as the "East" network (n = 10). We define a roadway flood as any amount of water above the elevation of the stormwater grate. In total, 166 samples were collected from Taylor's Creek under baseline conditions (58 at Overland Outfall and Outfall A, 50 at Outfall B) and 89 during perigean spring tide events (30 at Overland Outfall and Outfall A and 29 at Outfall B, corresponding to 3 measurements per studied tidal cycle, with five cycles being measured in each of the two perigean spring tide periods). Additionally, 10 samples were collected from roadway floodwaters (9 tidal floods, 1 compound flood).

Sterile, pre-rinsed 100 mL FalconTubes (polypropylene material) attached to a sampling pole were used to collect 100 mL water samples from Taylor's Creek at 0.5-1 m below the water surface across three locations that were within 2 meters of stormwater network outfalls. For flood water samples collected at the roadway inlet, collection occurred by hand at water surface depth. After collection, samples were stored in a cooler at 4°C during transport to the laboratory. Water samples were processed in duplicate for ENT following the IDEXX Enterolert standard protocol with the Quanti-tray 2000 (IDEXX, ASTM Method #D6503-99). The IDEXX assay requires a 100 mL sample volume. Per the IDEXX Enterolert protocol, due to the brackish salinities of the water samples, 10 mL of water sample was mixed with 90 mL of deionized water during processing. Samples were provided the IDEXX media and incubated for 24 hours at 41°C ± 0.5°C. ENT concentrations were estimated using a most-probable number (MPN) technique based on the number of large and small wells of the quanti-trays fluorescing under UV light, with a modified laboratory detection range of 10-24,196 most probable number ENT per 100 mL water (MPN 100 mL^{-1}) due to sample dilution. During sample collection and laboratory analysis, field blanks (n = 27) and laboratory blanks (n = 30) were also collected and processed for quality assurance and quality control purposes. Field blanks consisted of deionized water in sample bottles that were transported to and from the field in a sample cooler, and opened and recapped at

one or more of the sampling sites. Lab blanks consisted of deionized water in sample bottles that remained in the lab and were not transported to the field.

ENT concentrations were classified as above or below the EPA's single sample maximum threshold concentration for safe public use of recreational waters of 104 MPN 100 mL^{-1} (EPA, Recreational Water Quality Criteria, 2012). This EPA standard is for recreational use of marine waters, meaning it directly applies to water samples collected from Taylor's Creek, but does not directly apply to the assessment of roadway floodwaters, which are not recreational waters. Given that a standard does not exist for floodwaters of any kind – whether storm or tidally driven – but that pedestrians can still make contact with floodwaters, we consider the EPA's recreational standard here as a benchmark for comparison of water quality measurements collected from the roadway during the tidal floods.

2.3 Pipe Network Inundation Model

A pipe network inundation model of Front Street was used to characterize the extent of tidal inundation within the stormwater network during the measured flood events. Generally, a pipe network inundation model is a "bathtub" approach that simulates flooding by gradually filling subterranean pipes based on the elevation of the pipe relative to tidal stage (Strauss et al., 2012). The pipe network inundation model used in this study was generated using the *bathtub* R package (Gold et al., 2022) and is the same model used by Gold et al. (2022) for Beaufort, NC. This model was developed using elevations from a 2017 survey, during which some structures were inaccessible. Hence, our pipe network inundation model is incomplete for the Front Street study area, but can be used to estimate stormwater network inundation in two of the studied stormwater pipe systems, specifically the pipe networks of the Outfall B and Roadway Inlet (located in the East network) sampling locations (**Fig 1b**). The percentage of stormwater network inundation was quantified at discrete water levels by averaging the percent fill of surveyed stormwater point structures (e.g., drop inlets and catch basins) on Front Street.

2.4 Explanatory Variables used in Linear Regressions

We sought to explain measured variation in ENT concentrations within the waterway with both tidal and rainfall factors using robust linear mixed effect regressions of the baseline sampling and the perigean spring tide sampling data. Tidal variables used in the explanatory analysis include tidal height at time of sample collection, time since the last high tide before sample collection, and the height of the last high tide before sample collection. Tidal height at the time of sample collection was taken from the nearby NOAA Tides and Currents gauge (Fig 1a, which has been shown to match well with measured water levels in a stormwater catch basin on Front Street when it is not raining (Gold et al., 2023). The height of the last high tide, and the time since the last high tide, were likewise calculated using the NOAA tide gauge data. We chose to include time since the last high tide before sample collection and the height of the last high tide before sample collection to account for a potential lagged response of ENT concentrations in the waterway as tidal waters recede from the stormwater network, relative to the timing of maximum inundation of the stormwater network. While additional meteorological observations are available at the NOAA tide gauge, such as air temperature, we chose to only focus on the effects of tide and rain given our study period spanned two summer months and, thus, was not long enough to account for the effects of seasonal variation.

Because Taylor's Creek is a coastal waterway, it receives ENT loads by both direct runoff from stormwater outfalls and fluvial transport through the local watershed. To account for these transport pathways, we considered rainfall factors in the explanatory analysis. Seven rainfall variables were analyzed, all of which correspond to (antecedent) rainfall totals over finite intervals, including rainfall over 3, 6, 12, 24, 36, 48, and 72 hours prior to sample collection (in mm). These variables were calculated using hourly rainfall depths (millimeters) obtained from the Michael J Smith Airport weather station located approximately a mile north of Front Street in Beaufort, NC (North Carolina State Climate Office, station ID: KMRH; **Fig 1a**). The June rainfall total was 50 mm, and only one storm event produced a rainfall depth over 13 mm (or 0.51 inches). In contrast, the July rainfall total was 400 mm and 8 storm events produced rainfall depths greater than 13 mm.

2.5 Regression Analysis

ENT concentrations from the waterway were first logarithmically transformed to account for the exponential range of bacteria counts observed. The relationship between logarithmically transformed ENT concentrations ($\log_{10}$ ENT) and sampling location was then assessed using the Friedman test, which is a hypothesis test for repeated measures. We selected the Friedman test because the daily collection of ENT constitutes a repeated measure and the test does not assume an underlying distribution for the grouped-by-location data. The Friedman test determines whether the median value of a treatment group is significantly different from the median values of the other treatment groups. We assessed significance with an alpha value of 0.05. The Friedman test was calculated using the *stats* R package (R Core Team, 2013).

Next, the relationships between $\log_{10}$ ENT, tidal, and rainfall variables were analyzed using two sets of robust linear mixed effect models with the *robustlmm* R package (Koller, 2016). We selected robust linear mixed effect models because of the repeated-measures structure of our sampling design and the presence of outliers and non-normal residuals in the log10 ENT data set. The first set of models (the "baseline" models) used data from the entire two-month study period, which captured daily stormwater network inundation and multiple rain events. The second set of models (the "perigean spring tide" models) used data only from the perigean spring tide sampling that corresponded with complete inundation of the stormwater network and roadway flooding (i.e., the nine roadway flood events spanning June 12-16 and July 11-15, 2022). Data collected during baseline sampling and perigean spring tide sampling were modeled separately due to differences in sampling design, and to allow for assessment of relationships between tide and rainfall variables under distinct conditions. The model sets were comprised of individual robust linear mixed effect regressions between $\log_{10}$ ENT and a single explanatory variable to evaluate pairwise relationships.

Given that the waterway data varies both in space and time, the baseline mixed effect model has the following equation structure:

$$\log_{10}\text{ENT} = \text{Explanatory Variable} + (1 \mid \text{Site}) \qquad \text{Equation 1}$$

Where "|" denotes that the sampling location, or "Site", is a random effect. The perigean spring tide model has the equation structure:

$$\log_{10}\text{ENT} = \text{Explanatory Variable} + (1 \mid \text{Site}) + (1 \mid \text{Perigean Spring Tide / Categorical Tidal Stage}) \qquad \text{Equation 2}$$

Where "Site" and "Perigean Spring Tide" (June or July) are random effects and "Categorical Tidal Stage" (high, ebb, and low tide) is a nested random effect within the "Perigean Spring Tide" random effect. No interaction exists between the "Site" and "Perigean Spring Tide / Categorical Tidal Stage" terms because the sampling design produced a crossed-data structure between sampling period and site. The tidal and rainfall explanatory variables are fixed effects and were centered and scaled in the baseline models and perigean spring tide models. Conditional R^2 (variance explained by random and fixed effects) and marginal R^2 (variance explained by fixed effects or explanatory variables) were calculated for each regression using the *performance* R package (Lüdecke et al., 2021). Results are discussed in terms of their marginal R^2 value since marginal R^2 values only include the effect of the explanatory variable while conditional R^2 values also include the effects of random variables. Additionally, p-values and slopes were calculated to identify significant positive or negative relationships (alpha = 0.05) using the *parameters* R package (Lüdecke et al., 2020). All analyses were performed using R version 4.2.2 (R Core Team, 2022).

3 Results and Discussion

3.1 Tidal stormwater network inundation occurred daily

Water level data from within a storm drain on Front Street showed that the East network was inundated daily over the two month study period at each higher-high tide (n = 58, Gold et al., 2023). Using the pipe network model, we find that the stormwater network along other areas of Front Street was on average 57% inundated during the higher-high tides (on average 0.16 m MHHW) and 0.6% inundated during the lower-low tides (on average -0.91 m MHHW) during baseline conditions (**Fig 2a-2b**). During the perigean spring tide events, the average lower low tide and higher high tide elevations were -1.1 and 0.40 m MHHW, respectively, which correspond to modeled stormwater network inundation percentages of 0 and 78% along Front Street, respectively (Fig 2c-2d). During these events, inundation of the stormwater network extended inland beyond Front Street, filling pipes nearly 40% with tidal water (**Fig 2c**).

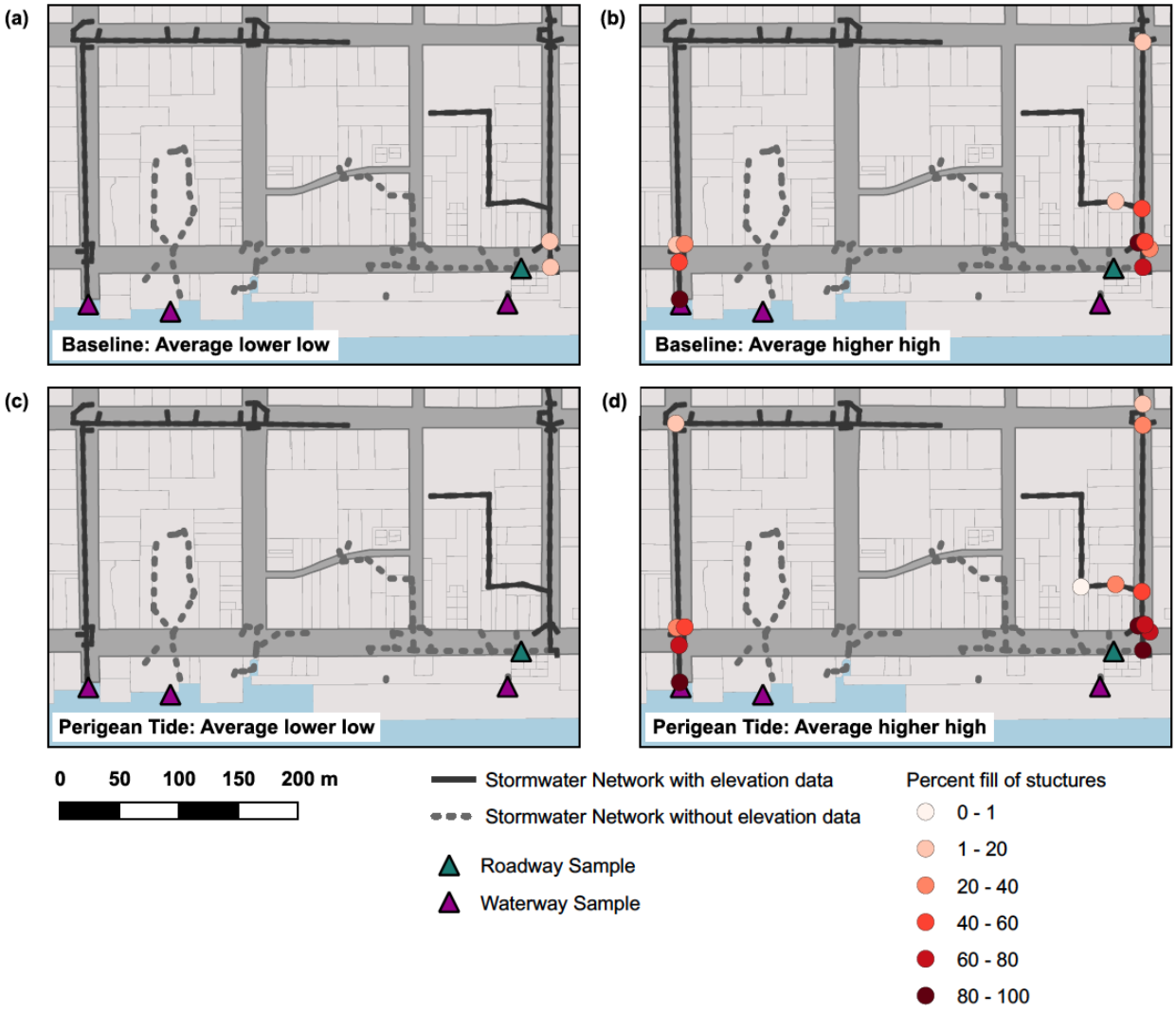


Figure 2. Modeled tidal inundation of two stormwater networks in downtown Beaufort during the study period. The average percent fill of stormwater catchments (attached to subterranean pipes) is shown for lower low tide (**2a**,**c**) and higher high tide (**2b**,**d**) for the baseline data collected from June 6 to August 2, 2022 (**2a,b**), and for the perigean spring tide data collected on June 12-17 and July 11-16, 2022 (**2c,d**).

Our pipe network model only accounts for variation in tidal elevation, as measured by the nearby NOAA tide gauge, and does not consider water level contributions from rainfall runoff, groundwater infiltration into the stormwater network, or pipe hydraulics. Bathtub models, like the pipe network model used here, typically overestimate flooding compared to models that account for pipe hydraulics (e.g., Castrucci & Tahvildari, 2018; Gallien et al., 2014). However, outside of rain events, Gold et al. (2023) showed that water levels in the East network track well with water levels measured at the NOAA tides gauge during baseline conditions, indicating that contributions from groundwater infiltration into the stormwater network are likely small for the pipe network. During rainfall events, the pipe network model is an underestimate of actual inundation.

While the network inundation model does not consider non-tidal forcing and pipe hydraulics, it can be used to estimate where inundation was relatively high and low across the network due to tidal forcing alone. In particular, the network inundation estimates highlight that total network inundation and roadway flooding were most likely to occur in and along the East network. The pipe network model also reveals that the Outfall B network experienced daily and major inundation, but to a lesser extent than the East network. Due to the inlet and outfall elevations not being surveyed, we are unable to assess the frequency at which the Outfall A network was inundated.

3.2 Daily waterway ENT concentrations differed between sampling locations

ENT was detected in 73% of daily, or baseline, samples (n = 241/330) with concentrations ranging from 10 to 5,298 MPN 100 mL^{-1} (**Fig 3**). ENT measurements exceeded the single sample maximum threshold concentration for safe public use of recreational waters (104 MPN 100 mL^{-1}) in 16% of the daily samples, indicating recurring but infrequent levels of unsafe fecal contamination in the waterway during the two month summer sampling period. Similar daily ENT concentrations in the waterway were observed between non-perigean and perigean spring tide periods. Hence, while perigean spring tide events lead to increased stormwater network inundation (**Fig 2d**), they did not necessarily result in increased ENT loading within the waterway.

We also found there were significant differences in ENT concentrations by sampling location (p-value = 0.004, **Table S1**). During baseline conditions, Outfall B had the highest median concentration of ENT in the waterway (31 MPN 100 mL^{-1}) while the sampling locations near Outfall A and Overland Outfall showed lower median ENT concentrations (both 10 MPN 100 mL^{-1}, which corresponds to the minimum detection limit). Inter-site variability between outfall sampling locations was also noted by Price et al. (2021), who posited that site-specific infrastructure can cause localized water quality dynamics. However, while significant differences were observed across sites, the median concentrations demonstrate how ENT levels were frequently low, and well below the EPA recreational water quality standard.

In addition to varying by location, median ENT concentrations across all outfall locations differed between the months of June and July 2022 (Fig 3, **Table S2**). For June, the median ENT concentration was 20 MPN 100 mL^{-1} and had an interquartile range of <10 to 23 MPN 100 mL^{-1} while the July median concentration was 41 MPN 100 mL^{-1} and had an interquartile range of 10 to 86 MPN 100 mL^{-1}. Rainfall occurred more frequently in July than in June. Periods of increased rainfall are known to correspond with increased FIB concentrations in waterways due

to runoff-driven contamination and elevated surficial groundwater elevations, which can also lead to greater sewage exfiltration (Secru et al., 2011).

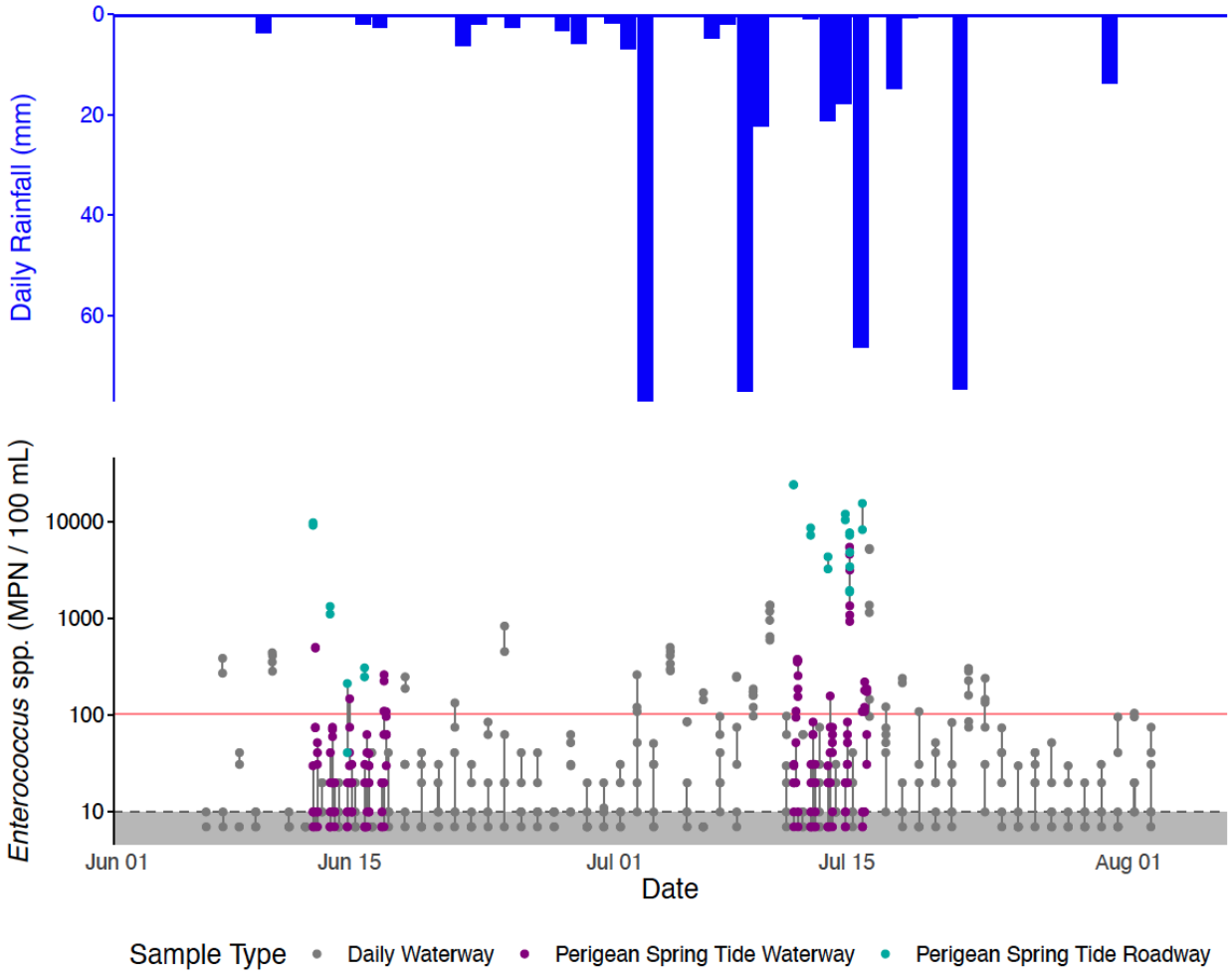


Figure 3. Daily rainfall (top) and ENT concentrations (bottom) during flood (perigean spring tide) and baseline conditions throughout June and July 2022. ENT concentrations are plotted on a $\log_{10}$ scale. The red line represents the EPA's single sample maximum threshold concentration for safe public use (104 MPN 100 mL^{-1}), and the grayed area represents the minimum detection limit (below 10 MPN 100 mL^{-1} for samples diluted 10:1). Gray and purple points correspond to measurements taken in Taylor's Creek during baseline and flood conditions at the three sampling locations, respectively, while teal points correspond to measurements taken from roadway floodwaters above a single stormwater catchment grate in the East network. Samples were processed in duplicate, and duplicate measurements are connected by vertical lines to visualize uncertainty.

3.3 Roadway floodwaters had consistently high ENT concentrations

We visually observed nine minor floods localized to the East stormwater network, and no roadway flooding via the Outfall A and Outfall B stormwater networks during perigean spring tides. The presence of flooding was corroborated by the storm drain water level measurements and the pipe network model. The floodwater samples exceeded the EPA's single sample

maximum threshold concentration for safe public use (104 MPN 100 mL^{-1}) for 8 of the 9 high tide flood events, in many cases by an order of magnitude (**Fig 3**). These ENT concentrations are comparable to the concentrations reported by Macías-Tapia et al. (2021, 2023) in Norfolk, VA in 2017, when 95% of the ENT samples exceeded the single sample maximum threshold concentration and ENT concentrations ranged from 30 to >24,000 MPN 100 mL^{-1}.

In June, the median floodwater concentration was 710 MPN 100 mL^{-1} and concentrations ranged from 41 to 9,804 MPN 100 mL^{-1} (**Fig 3**). By comparison, in July, the median floodwater concentration was 9,563 MPN 100 mL^{-1} and concentrations ranged from 1,882 to >24,196 MPN 100 mL^{-1}. The perigean high tide elevations were similar between June and July (0.35-0.51 and 0.34-0.45 m MHHW, respectively), as was the modeled inundation percentages at higher high tide (75-87 and 73-81%), so we hypothesize that differences in concentrations stem from antecedent rainfall factors. These findings suggest that antecedent rainfall can enhance fecal contamination in roadway floodwaters during high tide flood events. But, importantly, given that the June floodwaters were still more than 6 times higher than the EPA standard in the absence of rainfall, complete inundation of stormwater networks by tides in drier conditions can create concerning water quality conditions in roadway floodwaters.

The sources of fecal bacteria in the roadway floodwater samples are unknown. We believe the primary fecal bacteria sources during the high tide floods sampled were within the stormwater network. We observed fecal contamination in roadway floodwaters in the absence of rain, due solely to stormwater network inundation by tides, indicating a source from either Taylor's Creek or the stormwater network. However, water quality samples collected near stormwater outfalls in Taylor's Creek did not have similarly high concentrations (**Fig 3**), so Taylor's Creek is likely not a primary source of fecal bacteria to the floodwaters. Lastly, the majority of the roadway floods were small and confined to puddles around the storm drains (**Fig 4**), indicating that the floodwaters likely did not flush fecal matter from the land surface.

Because tidal inundation of stormwater networks can impede drainage of stormwater runoff, it may be that stormwater runoff with high ENT concentrations remain in the network as stagnant water for extended periods of time, potentially spilling into the roadway during the next high tide. Additional sources of fecal bacteria in the stormwater network could include exfiltrated sewage (Hart et al., 2020), and local environmental reservoirs of bacteria in biofilms and sediment (Burkhart, 2013). Importantly, since the majority of our measurements were collected from roadway floods with small overland extents, we are unable to assess whether high ENT concentrations would persist during more extensive floods, or if more extensive floods may dilute ENT concentrations instead, as proposed by Lewis et al. (2013).

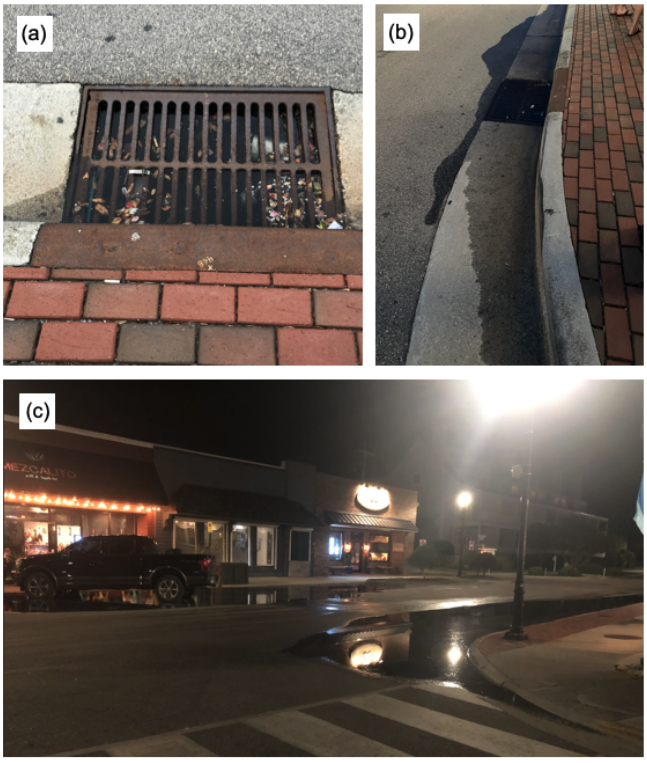
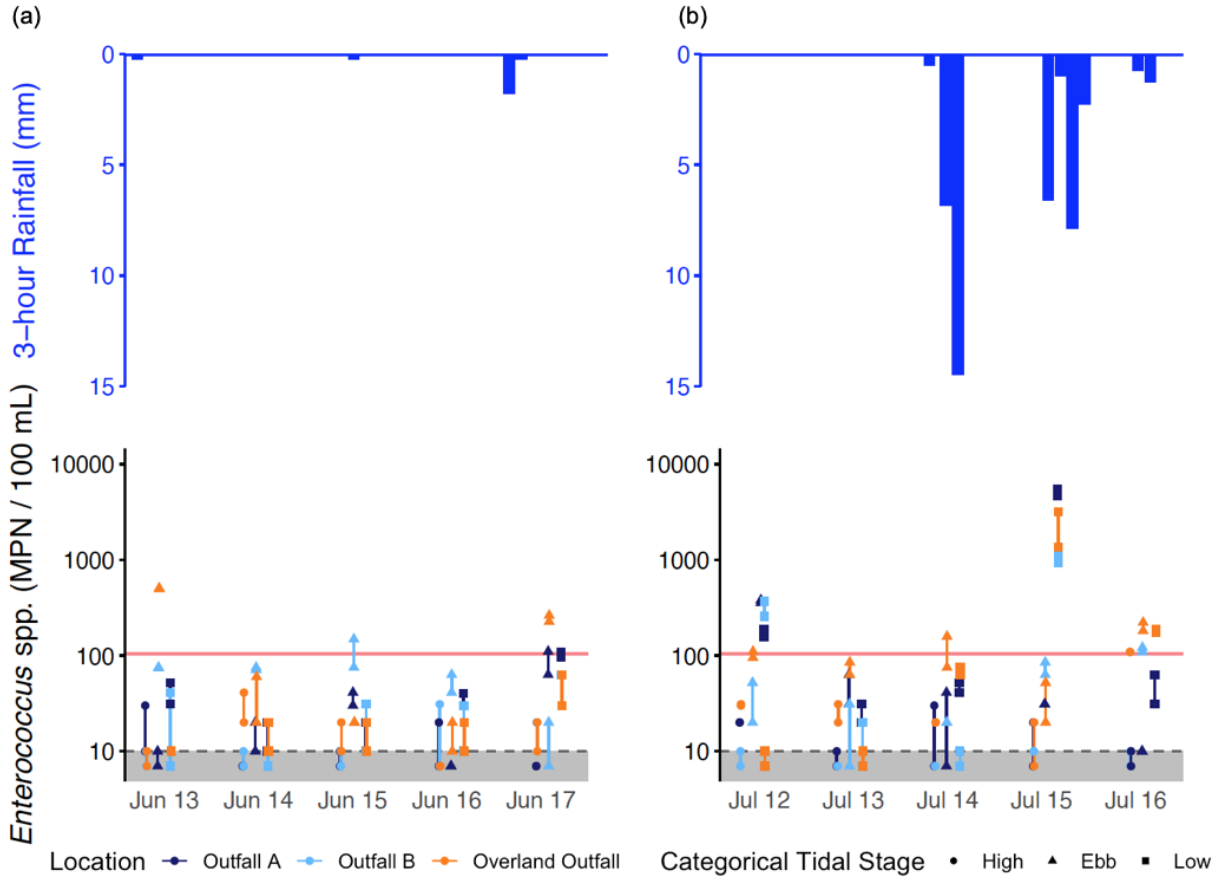


Figure 4. Tidal flooding at high tide on June 12, 2022 at 7:18PM (**a**), June 14, 2022 at 8:52PM (**b**), and June 15, 2022 at 9:51PM (**c**). We define a roadway flood as any amount of water above the elevation of the stormwater grate.

3.4 Highest median ENT concentrations occurred during ebb tide

To capture the water quality of receding tidal waters from the stormwater network during the perigean spring tide events, ENT measurements were collected at multiple tidal stages in the waterway: at high tide, ebb tide, and low tide (**Fig 5**). We observed a general trend that ENT concentrations increased from high tide to ebb tide and decreased from ebb tide to low tide. This trend has been previously reported for Taylor's Creek in Price et al. (2021), who observed that ENT increased during ebb tide as tidal height decreased and the stormwater network drained. In this present study, median concentrations at high, ebb, and low tide across both the June and July perigean spring tide events were 10, 52, and 30.5 MPN 100 mL^{-1}, respectively (**Fig 5**). Sixty percent (n = 36/60) of high tide samples had ENT concentrations below the minimum detection limit of 10 MPN 100 mL^{-1}. Then, as the tidal waters receded from the stormwater network during ebb tide, ENT concentrations increased, with exceedances of the EPA recreational water quality standard primarily occurring on ebb tide. From the linear regression analysis, we found that this trend was reflected in a significant negative relationship (coefficient = -0.28, p-value < 0.001) between tidal stage at sample collection and ENT concentrations in the perigean spring tide models (Table 1). This increase in median ENT concentrations from high to ebb tide suggests that receding tides transport ENT from the stormwater network to the waterway. However, we also observe a decrease in median ENT concentrations from ebb to low tide, which suggests that the effect of receded tidal waters on ENT concentrations in the waterway was short lived. This could potentially be explained by flushing or dilution. UV inactivation should not have affected

459 the measured ENT concentrations given that the perigean spring tide sampling occurred at night
460 during both the June and July events. While most of the EPA recreational water quality standard
461 exceedances occurred from samples collected during ebb tide, a few exceedances were observed
462 outside of ebb tide during the last monitored tidal cycle, which occurred following rainfall (**Fig**
463 **5b**).



464

465 **Figure 5.** 3-hour rainfall (top) and ENT concentrations (bottom) from Taylor's Creek during
466 perigean spring tide events from June 12-17, 2022 (**a**) and July 11-16, 2022 (**b**). The red line
467 represents the EPA's single sample maximum threshold concentration for safe public use (104
468 MPN 100 mL^{-1}), and the grayed area represents the minimum detection limit (below 10 MPN
469 100 mL^{-1} for samples diluted 10:1). Point shapes correspond to the tidal stages of high, ebb, and
470 low, while the colors correspond to sampling locations.

471 Although the highest ENT concentrations in the waterway during the perigean spring tide
472 events were observed during ebb and low tide, the majority of concentrations were below the
473 EPA's threshold for safe public use, demonstrating that the observed conditions did not pose a
474 public health hazard. Yet, the observed increase in ENT concentrations during ebb tide in a large
475 and dynamic waterway like Taylor's Creek demonstrates that tidal floods and stormwater
476 network inundation can serve as a source of fecal contamination to surface waters and are worthy

of further investigation as an emerging public health concern, particularly in coastal communities that drain to relatively small tidal creeks with minimal potential to flush or dilute pollutants.

Table 1. Linear regression models of baseline conditions (top) and perigean spring tide conditions (bottom) between log_{10}-transformed ENT concentrations (log_{10} MPN) and tidal or rainfall factors.

Baseline Conditions (n =166 observations); random effect = sampling location					
Explanatory Variables (fixed effect)	Conditional R^2 (full model)	Marginal R^2 (fixed effects)	p-value (alpha)	Coefficient	Standard Error
Tidal Factors					
Tidal Height	0.174	0.056	< 0.001	-0.15	0.03
Time to Last High Tide	0.159	0.037	< 0.001	0.12	0.03
Height of Last High Tide	0.124	0.009	0.073	-0.06	0.03
Rainfall Factors					
Prior 3-hour rainfall	0.231	0.105	< 0.001	0.21	0.03
Prior 6-hour rainfall	0.273	0.144	< 0.001	0.24	0.03
Prior 12-hour rainfall	0.385	0.268	< 0.001	0.34	0.03
Prior 24-hour rainfall	0.458	0.346	< 0.001	0.38	0.03
Prior 48-hour rainfall	0.395	0.313	< 0.001	0.36	0.03
Prior 72-hour rainfall	0.301	0.212	< 0.001	0.29	0.03
Perigean Spring Tide Conditions (n = 89); random effects = sampling location, categorical tidal stage, flood					
Explanatory Variables (fixed effect)	Conditional R^2 (full model)	Marginal R^2 (fixed effects)	p-value (alpha)	Coefficient	Standard Error
Tidal Factors					
Tidal Height	0.34	0.206	< 0.001	-0.28	0.07
Time to Last High Tide	0.348	0.134	0.041	0.22	0.11
Height of Last High Tide	0.394	0.006	0.212	-0.05	0.04
Rainfall Factors					
Prior 3-hour rainfall	0.415	0	0.775	-0.01	0.05

Prior 6-hour rainfall	0.373	0.003	0.516	0.03	0.05
Prior 12-hour rainfall	0.376	0.002	0.558	0.03	0.05

From the results of our robust linear mixed effect models for the perigean spring tides (**Table 1**), tidal height at sample collection and time to last high tide were associated with the greatest and second greatest marginal R^2 values (0.21 and 0.13, respectively). Though they cannot explain the majority of ENT variance (i.e., they do not have marginal $R^2 > 0.5$), the marginal R^2 values of tidal height at sample collection and time to last high tide support prior recommendations by Price et al. (2021) that tidal variables be included and documented as part of coastal ENT monitoring efforts, as tidal effects can play a role in transporting ENT within coastal systems.

Height of the last high tide and rainfall variables had low marginal R^2 values for the perigean spring tide conditions and coefficients that were not significantly different from zero (**Table 1**). Because height of last high tide was not a significant descriptor of ENT concentration variance, we conclude that maximum inundation prior to sampling was not the most important factor explaining the elevated ENT concentrations during ebb tide for our study site. Regardless, height of the last high tide should not be discounted as a potential variable to monitor, as our observational period may not have captured high enough water level events to exhibit a significant relationship with ENT concentration. Future SLR projections indicate more frequent and greater inundation of stormwater networks, such that height of the last high tide may become an important factor to explain elevated ENT concentrations as tidal water could reach sources of fecal contamination further into the stormwater network. Additionally, since data collection occurred in one municipality, other coastal stormwater network configurations may have increases in ENT concentrations at tidal heights comparable to the tidal heights measured in this study.

Likewise, our results should not indicate that rainfall is not an important descriptor to ENT variability during the ebb tide and drainage of the stormwater network, as other studies have demonstrated a direct link between rainfall and ENT concentration in coastal waterways (Mallin et al., 2000). The low marginal R^2 values of our tested rainfall variables likely stem from our sampling design, which targeted the tidal dynamics of the 6-7 hour period between the higher high tide and the higher low tide, and did not include many rainfall events.

3.5 Stormwater runoff drives higher ENT concentrations than tidal inundation

During baseline conditions, stormwater runoff drove higher ENT concentrations in the waterway than tidal inundation. Prior rainfall totals were significantly related to daily ENT variance in the waterway during baseline conditions (Table 1). Specifically, antecedent rainfall over 3, 6, 12, 24, 48, and 72 hours prior to sample collection had larger marginal R^2 values (0.10-0.35) in the baseline model than the significant tidal variables (0.04-0.06). The marginal R^2 values were greatest for the 24-hour ($R^2 = 0.346$) and 48-hour rainfall models ($R^2 = 0.313$), likely demonstrating lagged effects between rainfall and the accumulation of runoff over the broader drainage area. Many studies have reported increased FIB concentrations in waterways following rainfall events (Converse et al., 2011; Gonzalez et al., 2012; Parker et al., 2010; Stumpf et al., 2010). However, while antecedent rainfall predominantly explained ENT

concentrations during baseline conditions (i.e., when there is no roadway flooding from complete stormwater inundation by tides), rain variables did not significantly explain ENT variance during perigean spring tides, as discussed above.

Though rain variables were not significant descriptors of ENT variance during perigen tides, we observed that recent rainfall events coincided with the maximum ENT concentrations observed in Taylor's Creek during the perigean spring tide periods (Fig 5). On July 15, 2022, sample collection of the low tide samples coincided with a compound flood that was caused by rainfall plus reduced capacity in the stormwater network due to tidal inundation. These samples were collected 20 hours after a 22-mm, 8-hour rainfall event that occurred the previous day, and during the first hour of a 17-mm rainstorm that lasted over 9 hours. Approximately 3.6 mm of rainfall occurred during the first hour of the event. This compound flood event had the greatest ENT concentrations observed in the waterway during both perigean spring tides, indicating that compound flood events have the potential to bring greater magnitudes of fecal contamination to waterways compared to tidal inundation alone. Further research is needed to quantify the relationship between rainfall depth, extent of tidal inundation, and the magnitude of fecal contamination in the waterway, however our sampling of this one compound flood event demonstrates the importance of how different flood types can cause varying amounts of fecal contamination in coastal waterways.

4 Conclusions

Our findings indicate that chronic stormwater network inundation and tidal flooding caused by SLR present pathways for fecal contamination of coastal surface waters, but research is needed to fully understand whether network inundation and floods create public health hazards. In this study, we consistently observed high ENT concentrations above the US EPA's single sample maximum threshold for safe public use of recreational waters in tidal and compound floodwaters on a roadway in Beaufort, NC, but did not observe similarly concerning ENT concentrations in Taylor's Creek, the adjacent coastal waterway, as a function of tide-driven stormwater network inundation. We observed minor floods with minimal overland footprints, leading us to believe that the sources of the elevated ENT in the floodwaters were within the stormwater network, and could include undrained stormwater runoff, pipe biofilms and sediment, and exfiltrated sewage. Rainfall-driven runoff produced higher ENT concentrations in Taylor's Creek than stormwater network inundation and tidal flooding, reinforcing that stormwater runoff is the dominant driver of increased ENT concentrations in the coastal waterway. However, we observed short-lived increases in ENT concentrations in Taylor's Creek as perigean spring tides receded from the stormwater network, and we hypothesize the temporary increase in ENT concentrations is due to dilution or flushing given that Taylor's Creek is a large coastal waterway. In relatively small tidal creeks draining urban centers, the effects of dilution and flushing may not be as apparent, and the role of stormwater network inundation and tidal flooding on ENT concentrations may be more pronounced.

This study provided needed longitudinal water quality observations across a two-month period that included nine tidal floods from two perigean spring tide events, which allowed us to assess potential public health hazards from tidal flooding and make inferences on the relative roles of stormwater network inundation, tidal flooding, and rain on fecal contamination both in a coastal waterway and roadway floodwaters. However, the small size of the tidal floods we

observed prevents us from concluding whether more extensive floods are associated with higher or lower ENT concentrations. Additionally, while our results demonstrate that tidal floods are associated with problematic water quality impacts as was shown in previous studies (Macías-Tapia et al., 2021, 2023; Hart et al., 2020; Price et al., 2021; McKenzie et al., 2021), similar studies as the one presented here should be conducted across more locations to better understand site-specific effects related to stormwater infrastructure and land use. We suggest that future work incorporate sampling across multiple seasons, communities, and flood extents to better understand the prevalence of poor water quality from chronic stormwater network inundation and tidal flooding. Seasonal differences may contribute to the persistence of ENT within the stormwater network, while community differences in infrastructure, topography, land use, and waterway size could contribute to the type and number of ENT sources as well as the effect of tidal flushing on ENT concentrations in the waterway.

Ultimately, the water quality effects of chronic stormwater network inundation and tidal flooding are evolving given the non-stationarity of global climate change and SLR. SLR brings uncertainty to whether tidally-driven inundation and associated pollutant loading will remain at their current levels, as the magnitude of the loading will likely change with increased flooding frequency and damage to infrastructure. For Beaufort and other cities on the Atlantic coast of the US, rates of SLR are 30% higher than the global average of 3 mm/year (Ezer & Atkinson, 2014), meaning that future repetition of this study could produce differing results as tidally-driven inundation becomes more intense and widespread.

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Open Research

All data and code are provided as supporting materials with the manuscript for peer review purposes. Following acceptance, the data and code will be archived with Dryad.

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