2022 SCHOOL HOUSE BROOK MONITORING | MEMORANDUM



INTRODUCTION AND SUMMARY OF RESULTS

FB Environmental Associates (FBE) has been working with the Town of Bristol and Bristol Shellfish Committee to investigate potential pollution sources in the Pemaquid River, its tributaries, and the Pemaquid River Estuary since 2018. In 2022, The Town of Bristol Shellfish Committee received funds from the Maine Shellfish Restoration and Resiliency Fund (MSRRF) to do further investigation in the School House Brook Tributary which outlets to School House Cove. The cove is located on the east side of the Pemaquid River Estuary and is closed to shellfishing from May 1 through October 31. The Bristol Shellfish Committee has identified School House Cove as valuable shellfishing grounds and an important area for water quality restoration and flat re-opening.

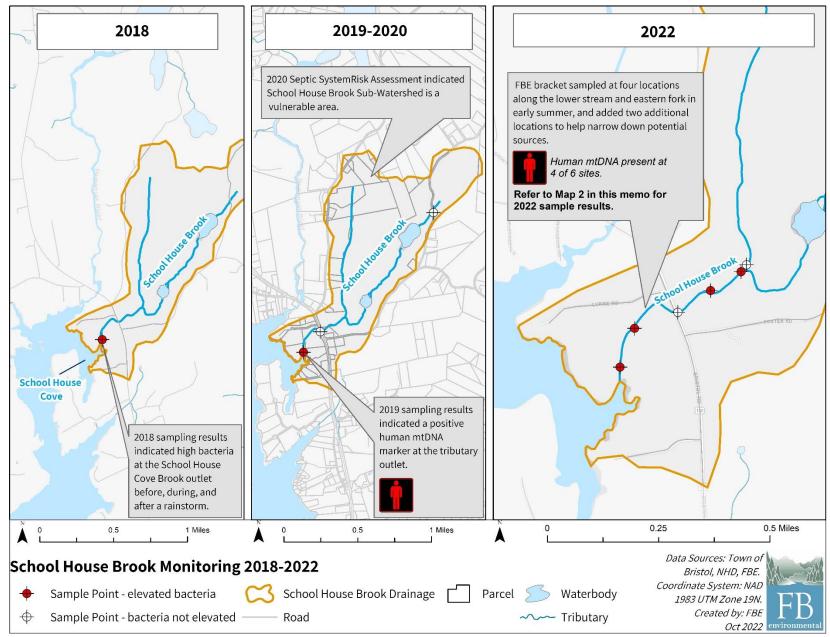
History of Investigative Sampling in the School House Brook Sub-Watershed

In 2017, Maine Department of Environmental Protection (Maine DEP) began sampling the freshwater tributaries of the Pemaquid River to identify possible sources of fecal contamination in the watershed contributing to the impairment of the estuary. As a continuation of this effort, sampling in 2018 by the Town, FBE, and the Maine DEP identified elevated bacteria levels entering the cove from School House Brook during pre-storm, mid-storm, and post-storm sampling across one storm event in June with a cumulative precipitation of 1.86 inches over 24 hours. Further sampling in 2019 on the School House Brook tributary using microbial source tracking at the outlet of School House Brook indicated a positive presence of the human mitochondrial DNA (mtDNA) marker during dry weather conditions. This suggests possible groundwater contamination from septic systems. In addition, the 2021 septic system risk assessment completed by FBE in collaboration with the Town identified areas vulnerable to septic system pollution upstream of the tributary outlet. In 2022, FBE completed bracket sampling at six locations along the lower segment of School House Brook to isolate potential sources of pollution. Refer to Map 1, below for a summary of the history of sampling in School House Brook.

Summary of Results

Between July 2022 and October 2022, FBE performed sampling along the brook four times during wet and dry weather conditions, adjusting sample sites throughout the summer to bracket (or isolate) the hot spot areas. Samples were analyzed for *Escherichia coli* (*E. coli*) bacteria, optical brighteners, nutrients (nitrogen and total phosphorus), and mitochondrial DNA (mtDNA) for the human marker. **Results indicated that there are several locations where bacteria levels were elevated in the majority of samples and where the human mtDNA marker was present. Specifically, two sites in the stream had notably elevated bacteria and had a positive presence of the human mtDNA marker multiple times throughout the summer.** One site is directly adjacent to the Bristol Consolidated School, and the second site is approximately ¼ mile upstream (sites SHB-1b and SHB-3, Map 2). The reoccurring presence of the human mtDNA marker indicates that a likely source of the elevated bacteria levels is from septic system leakage or waste. We recommend the Town follow up with assistance from the Code Enforcement Officer to inspect the vicinity for potential malfunctioning or failing septic systems.

The following memorandum provides the sampling methodology, sampling results, discussion, and recommendations.



Map 1. Historical sampling results on School House Brook and 2022 bracket sampling locations, chosen to isolate sources of pollution, specifically from suspected septic system effluent.

METHODOLOGY

Bracket Sampling

Scientists from FBE completed surface water bracket sampling on 7/21/2022, 8/10/2022, 8/31/2022, and 10/6/2022. Bracket sampling is a technique that assists in pinpointing sources of bacteria entering a tributary. This is completed through sampling upgradient and downgradient of potential sources to isolate pollutant source locations. Based on prior sampling results, FBE initially identified four sampling locations on the School House Brook (SHB) tributary: (SHB-1) just upstream of the outlet to School House Cove where a small wooden bridge crosses the brook, (SHB-2) just upstream of the Route 130 road crossing, (SHB-3) just downstream of the School House Brook tributary fork, and (SHB-4) just above the tributary western fork (Map 2). After the first sampling event in July, site SHB-4 was replaced by site SHB-1b because the location of Site 4 is within a stagnant wetland and the first round of results indicated an additional sampling point between site 1 and site 2 could help narrow down sources. Site SHB-1b is located just upstream of site SHB-1 and closer the Bristol Consolidated School.

After the sampling results for the first three sampling events were received, FBE analyzed the results and used the remaining funds to complete another round of sampling at four sites, including sites SHB-1b, SHB-2, SHB-3, and a new site SHB-2b located between sites 2 and 3.

Samples were collected during two dry weather events (<0.25" of rain within the prior 24 hours) and two wet weather events (targeting >0.25" of rain within the prior 24 hours) (Table 1).

Parameters included 1) *E. coli*, 2) nitrate + nitrite as nitrogen, 3) total phosphorus, 4) optical brighteners, and 5) mitochondrial DNA for human presence. Surface water samples were collected and analyzed as follows:

- Field parameters included dissolved oxygen, temperature, salinity, and specific conductance and were recorded using a YSI ProSolo field meter.
- *E. coli*, total phosphorus, nitrate, and nitrite samples were analyzed at Maine Environmental Laboratory (MEL) in Yarmouth, Maine.
- Optical brighteners were measured using a handheld Aquaflor fluorometer, based on methods described in SOP 3.4.1.4 Measuring Optical Brighteners in Ambient Water Samples Using a Fluorometer, by Erick Burres, dated March 2011.
- Mitochondrial DNA for human presence samples were analyzed at the University of New Hampshire by Dr. John Bucci.

Table 1. Precipitation prior to sampling events. Data from KMEBRIST8 station, sourced via <u>wunderground.com</u>. Weather events are considered 'wet' with >0.25" of rain within prior 24hrs.

| Sample Date | Hours Prior To Sampling | Cumulative Precipitation (in) | Dry or Wet | | |
|----------------|----------------------------|----------------------------------|---------------|--|--|
| 7/21/2022 | 24hrs 0 | | Dry | | |
| 7/21/2022 | 48hrs | | | | |
| 0/10/2022 | 24hrs | 0.03 | Dry | | |
| 8/10/2022 | 48hrs | 0.09 | | | |
| 0/21/2022 | 24hrs | 0.56 | Wet | | |
| 8/31/2022 | 48hrs | 0.56 | | | |
| 10/6/2022 | 24hrs | 0.26 | Wet | | |
| 10/6/2022 | 48hrs | 0.26 | wet | | |



School House Brook outlets to School House Cove, in the eastern side of the Pemaquid River Estuary.

Using E. Coli as a Fecal Indicator Bacteria

Fecal indicator bacteria (such as *Escherichia coli* or *E. coli*, Enterococci, and Fecal Coliform) are used to track a wide variety of potentially harmful pathogens such as viruses and bacteria found in fecal waste that would otherwise be too expensive to monitor comprehensively. High in-stream fecal indicator bacteria levels during dry weather events can point to possible groundwater contamination from septic systems. *E. coli* is the most appropriate indicator bacteria for fecal source tracking in freshwaters and is the Maine DEP standard. *E. coli* was used as a sampling parameter for all four sampling events.

"Co-Indicators" to Fecal Indicator Bacteria

In addition to *E. coli*, several other parameters can be used as co-indicators to help confirm humans are the likely the source of contamination, such as nutrients (nitrogen and phosphorus) and optical brighteners. Nutrients (nitrate, nitrite, and phosphorus) can indicate human waste contamination if found in high concentrations. Optical brighteners are commonly used for wastewater detection and their presence can indicate a human contamination source in the sample. Optical brighteners are not naturally occurring and are typically added to laundry soaps, detergent, cleaning agents, and toilet paper to aid in the brightening of fabrics and/or surfaces. Testing positive for optical brighteners in groundwater strongly suggests greywater from leach fields are entering the groundwater without adequate filtration. Co-indicators were used as sampling parameters on the first three sampling events, 7/21/2022, 8/10/2022, and 8/31/2022.

Microbial Source Tracking

Elevated fecal indicator bacteria and nutrients (discussed above) do not alone constitute human waste contamination because fecal indicator bacteria can be sourced from wildlife or proliferate on certain substrates within the environment. Microbial source tracking (MST) is an analytical technique that uses mitochondrial (mtDNA) host-specific markers to determine the source animal of mtDNA found in water samples. The mtDNA analysis was performed at the UNH Microbial Source Identification Laboratory by John Bucci, Ph.D., using molecular biomarkers to assess water quality. If positive, the source of a present sample is detected within the standard microbial source guidelines. If absent, the target biomarker was not detected above the limit of detection and is therefore considered absent. FBE collected samples at all six locations along School House Brook for mtDNA analysis for the human marker. The human marker was used as a sampling parameter for all four sampling events (with the exception of 10/6/2022 at site SHB-3, when the sample was broken during transport and not able to be analyzed).

Fecal contamination is one of the most difficult pollutants to remediate. There are a few reasons for this:

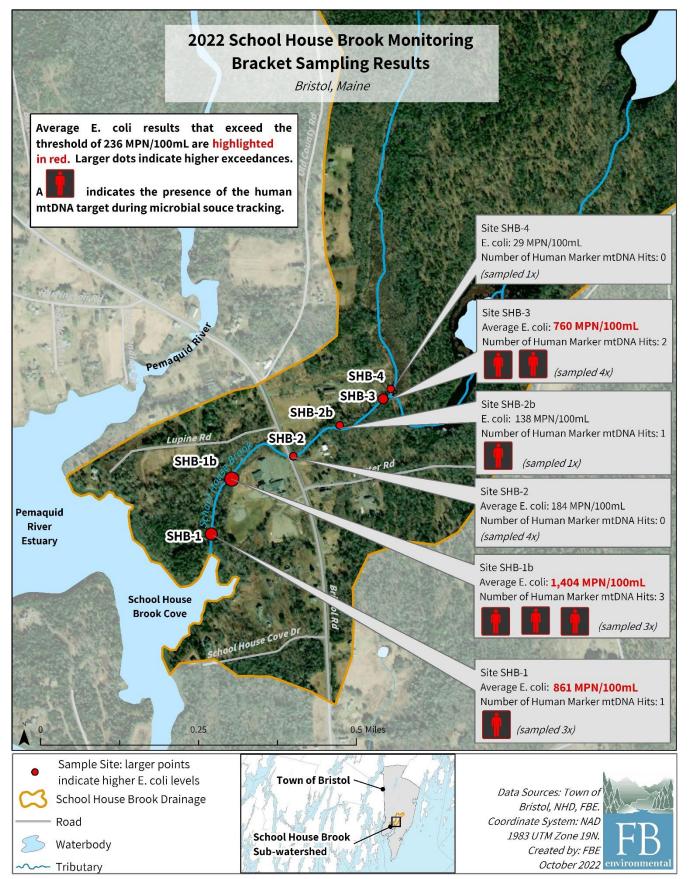
- (1) It is a nonpoint source pollutant, meaning that it can come from many different locations on the landscape.
- (2) Human health concerns are caused by potentially harmful pathogens, that are present within fecal matter. However, it would be too expensive to track and monitor each pathogen individually. Instead, we use indicator organisms (such as *Escherichia coli* or *E. coli*, Enterococci, and Fecal Coliform). These indicator organisms are chosen based on similarities to pathogens in behavior and transport in the environment.
- (3) Synchronicity in behavior between fecal indicator bacteria and the pathogens-of-concern for public health risk (e.g., salmonella, campylobacter, rotavirus, giardia, norovirus, hepatitis, etc.) may break down under certain environmental conditions. Therefore, caution must be used when interpreting fecal indicator bacteria data in the context of risk management decisions.
- (4) Fecal contamination tracking is an evolving science, with new technologies consistently making their way to the market. We do our best to use the tools at our disposal while recognizing their limitations.

RESULTS

Results are displayed in Table 2 and Map 2. Standard water quality metrics – temperature, dissolved oxygen, specific conductance, and flow level were measured at each site to establish baseline conditions. Baseline levels can be used to help discern future effects on water quality and to inform source tracking. Salinity was measured for all samples to ensure samples were taken in freshwater (above head of tide) because research shows *E. coli* can break down in saltwater. All sites met the Maine DEP recommended threshold for temperature, salinity, and specific conductance, however **low dissolved oxygen below the set threshold of 75% was experienced at numerous sites throughout the sampling season**. This is likely due to School House Brook's low flow levels (especially during dry weather conditions) and because the portion of the stream that was sampled drains directly from a large stagnant wetland which could naturally depress dissolved oxygen. All sites were below the dissolved oxygen thresholds of 75% and on 7/21/2022 and on 8/10/2022 (with the exception of site SHB-2 on 8/10/2022). On 8/31/2022 and 10/6/2022, only SHB-3 was below the dissolved oxygen threshold, likely due to higher flow conditions during the wet weather events.

The Maine DEP sets a threshold for *E. coli* bacteria for Class A and B waters at 236 MPN/100mL. The US Environmental Protection Agency establishes guidance for nutrient water quality parameters that assist with evaluating sample results. Results in Table 2 are displayed in context of these thresholds. Nitrate + Nitrite can be highly variable, and the guidance for Nitrate + Nitrite as Nitrogen used for this analysis is that samples should be less than 0.20 mg/L, which is set above the EPA's background reference conditions level of 0.07 mg/L. The guidance for total phosphorus is that samples should be less than 0.02 mg/L. For mtDNA analysis, results are presented as presence/absence. Notable exceedances for each site are summarized by site, downstream to upstream (Map 1).

- Site SHB-1 had exceeded the *E. coli* threshold of 236 MPN/100 mL once and had the human mtDNA target present once. Nutrients were also slightly elevated throughout sampling.
- Site SHB-1b exceeded the *E. coli* threshold twice at very elevated bacteria levels, and also had the human mtDNA target present three times. Nutrients were also slightly elevated throughout sampling. **Based on these results, the vicinity of SHB-1 and SHB-1b is considered a hot spot.**
- Site SHB-2 exceeded the *E. coli* threshold one time and had elevated nutrient levels during some sampling events. There were no positive hits for the human mtDNA target at this site.
- Site SHB-2b (only sampled one time on 10/6 during the follow up sampling event) had the human mtDNA target present.
- Site SHB-3 exceeded the *E. coli* threshold twice with high bacteria levels, and also had the human mtDNA target present twice. Nutrients were also slightly elevated throughout sampling. **Based on these results, the vicinity of SHB-2b and SHB-3 is considered a hotspot.**
- SHB-4 was only sampled once on 7/21 and was subsequently replaced with SHB-1b.



Map 2. Summarized water quality results for 2022 School House Brook water quality bracket sampling.

Table 1. Field and laboratory water quality results for 2022 School House Brook water quality bracket sampling. Results are displayed by site. Exceedances above the threshold or guidance criteria are shown highlighted in **red**.

| Site Name | Date | Dissolved Oxygen (mg/L) | Temperature (°C) | Specific Conductance (µS/cm) | Optical Brighteners (positive or negative) | <i>E. coli</i> (MPN/100 mL) | Human mtDNA (present or absent) | Nitrate+ Nitrite (mg/L) | Total Phosphorus (mg/L) | |
|---|--|----------------------------|---------------------|------------------------------------|---|-----------------------------------|---------------------------------------|-------------------------------|-------------------------------|--|
| | Threshold: | < 7 mg/L | >24°C | > 854 µ S/cm | positive | <i>> 236</i> MPN/100 mL | Present | > 0.20 mg/L | > 0.02 mg/L | |
| SHB-1 | 7/21/2022 | 5.78 | 20.3 | 140.5 | negative | 63 | Absent | 0.23 | 0.15 | |
| | 8/10/2022 | 4.37 | 18.5 | 81.5 | negative | 99 | Present | 0.13 | 0.18 | |
| | 8/31/2022 | 7.14 | 20.1 | 147.7 | negative | > 2,420 | Absent | 0.17 | 0.12 | |
| q. | 8/10/2022 | 6.73 | 18.6 | 154.4 | Undetermined* | 1,733 | Present | 0.25 | 1.3 | |
| SHB-1b | 8/31/2022 | 7.37 | 20.2 | 194.5 | negative | > 2,420 | Present | 0.17 | 0.11 | |
| SF | 10/6/2022 | 8.46 | 13.9 | 152.1 | - | 58 | Present | - | - | |
| | 7/21/2022 | 6.52 | 19.6 | 106.7 | negative | 22 | Absent | 0.37 | 0.4 | |
| SHB-2 | 8/10/2022 | 7.5 | 16.7 | 140.2 | negative | 43 | Absent | 0.27 | 0.76 | |
| | 8/31/2022 | 7.02 | 19.2 | 85.7 | negative | 649 | Absent | 0.36 | 0.26 | |
| | 10/6/2022 | 9.41 | 13.7 | 123.9 | - | 21 | Absent | - | - | |
| SHB-2b | 10/6/2022 | 8.1 | 12.8 | 76.3 | - | 138 | Present | - | - | |
| SHB-3 | 7/21/2022 | 2.1 | 23.9 | 57.2 | negative | 488 | Absent | ND | 0.12 | |
| | 8/10/2022 | 1.84 | 20.4 | 57.2 | negative | 117 | Present | ND | 0.67 | |
| SHI | 8/31/2022 | 1.43 | 21.6 | 63.6 | negative | > 2,420 | Present | ND | 0.11 | |
| | 10/6/2022 | 5.16 | 12.8 | 75.2 | - | 15 | n/a | - | - | |
| SHB-4 | 7/21/2022 | 0.56 | 23.1 | 61.1 | negative | 29 | Absent | ND | 0.12 | |
| ** Samples are tested in triplicates; undetermined presence indicates the sample tested positive for optical brighteners in two of the triplicates and negative for optical brighteners in one triplicate. Resampling is recommended. | | | | | | | | | | |
| "-" indicates site was not tested for that parameter. During the last round of testing in October, a reduced parameter list was used. | | | | | | | | | | |
| n/a: T | n/a: The mtDNA sample for SHB-3 on 10/6/2022 was broken during transport and not able to be sampled. | | | | | | | | | |

DISCUSSION & RECOMMENDATIONS

Ultimately, the results from the 2022 water quality sampling on School House Brook found elevated bacteria levels, elevated nutrient levels and positive hits for the presence of the human mtDNA marker at several of the sampling sites during wet and dry weather events. **Based on the results, FBE identifies two "hot spots" along School House Brook that had repeated elevated bacteria** *and* **repeated hits for the human mtDNA marker.** These hotspots are important places for the Town to follow up on to identify the potential source of bacteria, and specifically septic system effluent that is likely causing the repetitive hits for the human marker.

Hotspot #1: The vicinity of SHB-1 and SHB-1b is located directly adjacent to the Bristol Consolidated School. SHB-1b was added approximately 500 feet upstream of SHB-1, to try and narrow down the potential source. SHB-1b specifically tested positive for the presence of the human mtDNA marker on all three sampling events (8/10, 8/31, and 10/6) and SHB-1 received a hit for the human marker on 8/10. The presence of elevated bacteria and presence of human mtDNA marker contamination sources (such as from faulty septic systems). This hotspot area also had elevated bacteria levels during wet weather, which can point to fecal sources on the landscape via stormwater runoff. In addition, nitrate + nitrite was notably elevated at site SHB-1b on 8/10. Nutrients, such as nitrate + nitrite, can indicate human marker and elevated bacteria levels in School House Brook in this area, but the presence of the human marker and elevated bacteria during all weather conditions indicates this area should be followed up on for opportunities to remediate malfunctioning or failing septic systems. Sources of bacteria are possible from upstream (see Hotspot #2, below) but consistently low results at SHB-2 between these two sites suggests that they are isolated sources of contaminants.

Hotspot #2: The vicinity of SHB-3 and SHB-2b, located upstream of where School House Brook crosses Bristol Road/Route 130, also had repeated elevated bacteria and hits for the human mtDNA marker across multiple sampling events. SHB-3 specifically tested positive for the presence of the human mtDNA marker on two sampling events, 8/10 and 8/31. SHB-2b also tested positive for the presence of the human mtDNA marker on 10/6. SHB-2b was added approximately 300 feet downstream between SHB-2 and SHB-3 for the fourth sampling event in an effort to identify the potential source of the human mtDNA marker (SHB-2 did not have any hits for the human mtDNA marker at any point in time). <u>Ultimately, results indicate that there is likely a source of bacterial pollution in the vicinity of SHB-3 and SHB-2b. This area should be followed up on for opportunities to remediate malfunctioning septic systems.</u>

Recommended Action Items

We recommend the Town of Bristol work with the Code Enforcement Officer and/or a licensed septic system evaluator to follow up at the properties in the vicinity of the two identified hotspots. FBE's specific recommendations are to:

- Connect with the Bristol Consolidated School and identify if they have experienced any signs of a failing septic system. However, not all malfunctions or failures will be visible to the property owners and specifically, underground leakages can go undetected. Therefore, we suggest a septic system evaluation by a certified evaluator to determine if it is functioning correctly.
- Connect with landowners adjacent to sites SHB-3. We recommend approaching the landowners to inquire if they are open to discussing their existing septic system or having a septic system inspection completed. Note that the results of this sampling <u>do</u> indicate that human waste is a likely source of pollution in School House Brook but it <u>does not</u> indicate which property may be causing this. The <u>Small Community Grants Program</u>, funded through Maine DEP, does provide funding for replacing septic systems if they are polluting a shellfishing area (some income restraints do apply to this funding source).
- Conduct outreach to landowners within the Town of Bristol about proper system maintenance and care. Education can reduce the risk of malfunctioning or failing systems in the future.