Tracking Bacterial Contamination in the Lamprey River Watershed

Final Report
to the Lamprey Rivers Advisory Committee

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Steve Jones and Reka Ivanyi
Dept. of Natural Resources and the Environment
University of New Hampshire

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INTRODUCTION AND BACKGROUND

The main goal of this project is to continue monitoring at key sites in the Lamprey River Watershed (LRW) and expand at sites in areas of concern, especially the Moonlight Brook watershed, to provide essential data for assessing water quality, public health risks and sources of any contamination. Addressing this overarching goal will serve to:

1.) Expand the baseline of information on bacterial pollution to assess water quality status, trends, and contamination sources in the Lamprey River and the Moonlight Brook watersheds.
2.) Continue targeting rainfall events to determine the extent to which these events trigger elevated bacterial concentrations and/or different pollution sources.
3.) Compile data from ongoing and past bacterial monitoring efforts in the Great Bay watershed.
4.) Assess the potential for eliminating or mitigating pollution sources identified by this study.
5.) Extend findings to interested groups through meetings and published reports.

This Final Report is a summary of all project findings, as well as an updated summary of data from other earlier and ongoing projects related to microbial contamination of the watershed. The report relates particularly to a Goal of the 2013 Lamprey River Management Plan (https://www.lampreyriver.org/about-us-2013-management-plan-draft) under "Enough Clean Water": *Ensure that the Lamprey rivers meet or exceed standards for "fishable and swimmable" water for the health and enjoyment of all species.* The specific focus of this study was assessment of water for swimmable and other recreational uses, using study-generated and other data in comparison to State bacterial indicator standards (NHDES 2019a; 2024a) to enable identifying sites and areas that are clean or of public health concern. Note that "NHDES produces an Integrated Report every 2 years, describing the quality of New Hampshire's surface waters and an analysis of the extent to which all such waters provide for the protection and propagation of a balanced population of shellfish, fish and wildlife, and allow recreational activities in and on the water. Some waters are categorized as impaired, and some impaired waters require the creation of a Total Maximum Daily Load (TMDL)" (https://www.des.nh.gov/water/rivers-and-lakes/water-quality-assessment). This report is based on the 2024 evaluation as used for the 2024 report and because the 2026 evaluation is still in preparation. The report also sought to identify data trends to track progress or detect new or emerging problems with water quality.

Providing a baseline of information related to bacterial pollution in the Lamprey River and the Moonlight Brook watersheds is important because there are little to no data related to fecal contamination of recreational surface waters available from the State of New Hampshire in recent years, based on what is presented in their reports related to river water quality (NHDES 2019b; 2022; 2024 b&c). These reports include little discussion of this indicator beyond 'designated' beaches and the NHDES Shellfish Program. There is a searchable category for Beaches with posted fecal bacterial data on the NHDES OneStop database (https://www4.des.state.nh.us/DESOnestop/BasicSearch.aspx), while this report provides a convenient way to access bacterial data for other recreational surface-water uses.

The Intended Audience and beneficiaries of this work include: 1.) The LRAC and local volunteers and citizens by providing information about the water quality and potential public health risks for recreating in the Lamprey River watershed and surrounding estuary; 2.) Local and state resource, public health and public works personnel who can use the data to focus resources and effort on problem areas where water pollution may pose a threat or restricts use. 3.) Monitoring program managers who can augment their programs with similar efforts. We continue to present study findings at relevant regional meetings, and some of the data will be used by students to present research posters at the UNH Undergraduate Research Conference in April 2026. The 2025 data are also part of an ongoing evaluation and summarization of findings by the UNH Jones lab from past years (2021 to present), all supported by LRAC.

The Evaluation Process for this project includes data analysis and interpretation, using comparisons of data to State water quality standards to enable clear explanation of the potential significance of the findings. We will track who gets involved and their interests, and how many State, Federal and local agencies are provided with the Final Report. It will be important to also track what management actions are undertaken because of this work once it is made available. The elimination of identified pollution sources can be a direct benefit that can also be tracked.

METHODS

Water samples were collected from the shorelines of the Lamprey River from Newmarket to Raymond, and the Moonlight Brook watershed in Newmarket for analysis of bacterial pollutants. Sampling in the Lamprey River watershed occurred at 6 sites where surface water recreation occurs (Tab. 1; Fig. 1). Site 1* is near a site listed as NHEPLRDO16 and was sampled in the tidal portion at low tide. Site 2 is in the dam impoundment area (NHRIV600030709-13) of lower Piscassic River. Site 3 corresponds to the NHDES water quality monitoring program site 07T-LMP and is downstream from 08-LMP. Site 4 is located between NHDES sites 11-LMP and 11A-LMP. Site 5 is in section NHRIV600030703-15 behind the Epping Town Hall; Site 6 is in section NHRIV600030703-07-02 at Carroll Beach behind the Raymond Elementary School.

NHDES Water Quality Assessment category information in Table 1 for these sites can be located here:https://www4.des.state.nh.us/onestoppub/SWQA/010600030709_2024.pdf

Table 1. 2025 NHDES Water Quality Assessment categories in the Lower Lamprey River (HUC 12: 010600030709) and the Middle Lamprey River (HUC 12: 010600030703) for the 6 main study sites.

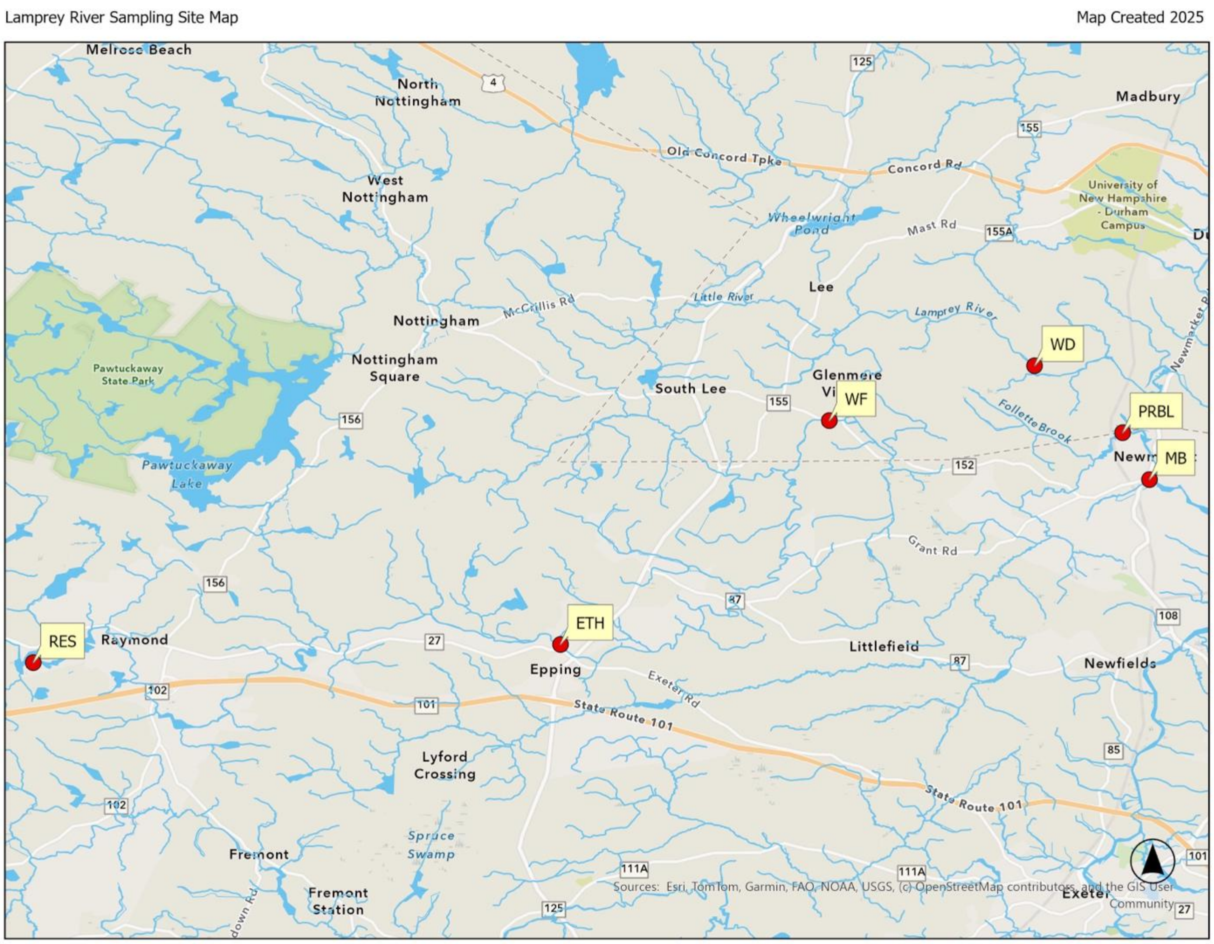
Study Site	Assessment ID number/site ID	Unit Name	Type** of Recreational use	Last sample	Last exceed	Classification Category†
Site 1*	NHRIV600030709-13	Moonlight Brook	Primary Contact	2008	2000	3-ND
MBO	direct drain to E*01-01 area	upstream of Moonlight Bk mouth	Secondary Contact	2008	1996	3-ND/5-P**
Site 2	NHIMP600030708-03	Piscassic Park Boat Launch	Primary Contact	2007	2005	3-ND
PRBL	near 01-PIS	(Lamprey R. impoundment)	Secondary Contact	2007	N/A	3-ND
Site 3	NHIMP600030709-02	Wiswall Dam	Primary Contact	2008	N/A	3-ND
WD	08-LMP	just above the dam	Secondary Contact	2008	N/A	3-ND
Site 4	NHRIV600030709-01	Upstream of Wadleigh Falls	Primary Contact	2007	1999	3-ND
WF	11-LMP	Lee public canoe boat launch	Secondary Contact	2007	N/A	3-ND
Site 5	NHRIV600030703-15	Behind Epping Town Hall	Primary Contact	2018	2018	4A-P†
ETH	13A-LMP	(Middle Lamprey River)	Secondary Contact	2018	2002	3-ND
Site 6	NHRIV600030703-07-02	Carroll Lake Beach	Primary Contact	2006	2006	4A-P†
RES	BCHCLBRAY	Behind Raymond Elem. Sch.	Secondary Contact	2006	N/A	3-ND

*All sites in the Moonlight Brook watershed fall under this same Assessment Unit; E*01-01.

**5-P: Does not meet water quality standards, TMDL needed; the impairment is more severe and causes poor water quality;

†4A-P: Does not meet water quality standards, TMDL completed; the impairment is more severe and causes poor water quality;

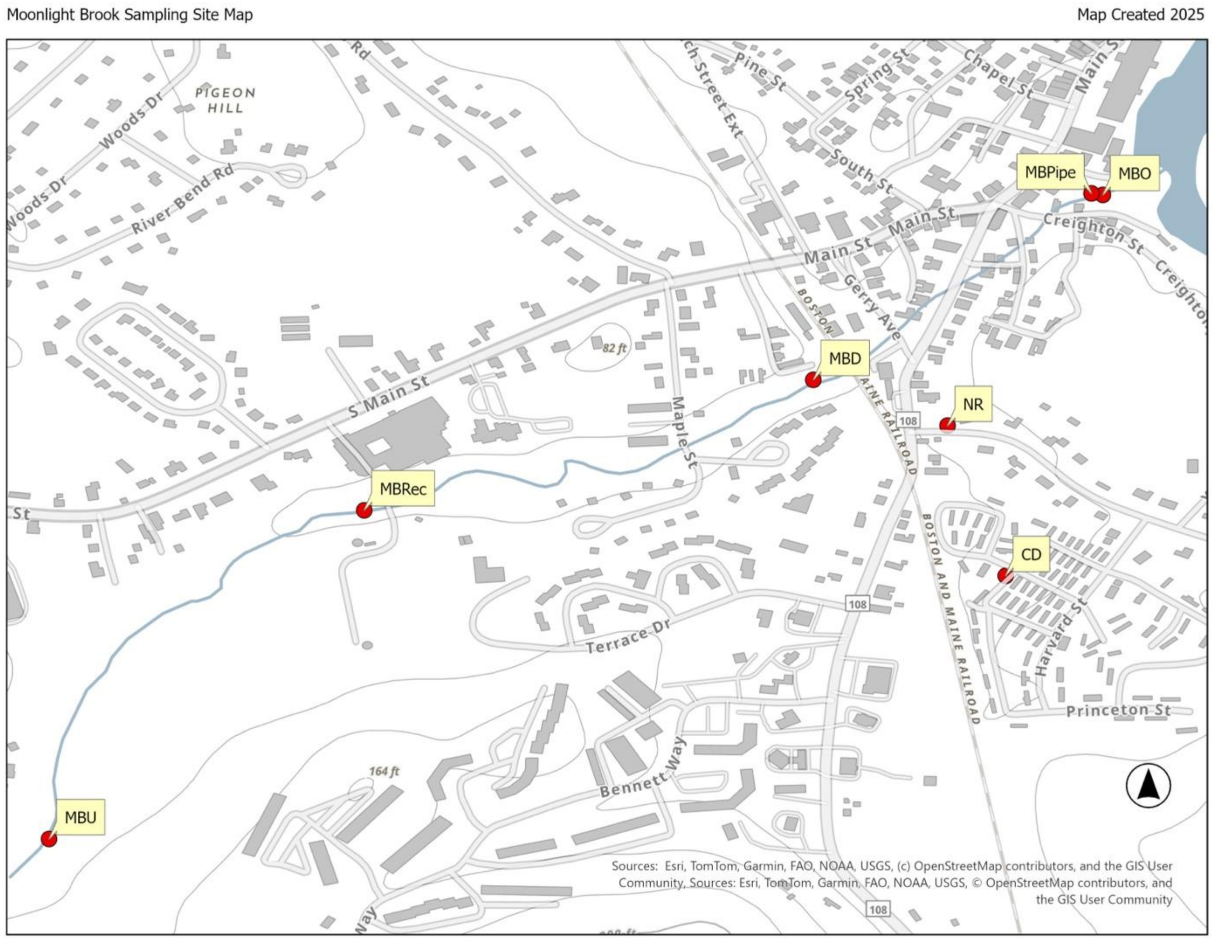
Figure 1. Locations of project study sites during 2025 (NHDES 2024d). This figure and Figure 2 were developed using the NHDES Surface Water Quality Assessment Viewer:
https://experience.arcgis.com/experience/23aca0560af74cfa9f0d39f4125ce479#data_s=id%3A83f83eddd9b8476580896fe20c0e4c5b-4abcbca5f2ae49d784eea43ebcbbb593%3A3379



To enable detailed exploration of sources of fecal contamination in Newmarket, we have established 6 routine sampling sites in the Moonlight Brook watershed including its one other tributary sub-watershed (Fig. 2). Sample sites included Site MBO, the outlet of Moonlight Brook to the tidal portion of the Lamprey River and the same location as Site 1*, then Site MBD upstream next to Moonlight Drive just to the west of the railroad crossing, Site MBRec recreation next to the recreational area behind the high school, and Site MBU near the most upstream section of the brook. Two other sites in a tributary in downtown Newmarket included Site NR next to New Road, and Site CD next to Columbia Drive in the Sleepy Hollow trailer park, where it's probable that little surface water recreation occurs. These sites were chosen instead to help determine the location and types of fecal pollution that contribute to what is discharged into the tidal portion of the Lamprey River, where boating is popular.

*All sites in the Moonlight Brook watershed are classified the same as Site 1 (Table 2) by NHDES.

Figure 2. Locations of project study sites in the Moonlight Brook watershed during 2025.



All samples were collected and stored on ice while being transported to the Jackson Estuarine Laboratory (JEL) for analysis within 4 hours of sampling. The sampling in the Lamprey River watershed occurred once per month during 2025 on May 12, June 27, July 2, August 13, September 23, October 20, and November 17. The sampling in the Moonlight Brook watershed occurred approximately once per month during 2025 on May 28, June 3, July 16, August 6, and October 9. The samples were analyzed to determine concentrations of bacterial indicators of fecal pollution that are used by the State of NH for classifying and managing coastal waters: Enterococci (coastal water recreation), fecal coliforms (shellfish harvesting), and *Escherichia coli* (*E. coli;* freshwater recreation) using standard methods accepted by state agencies for these purposes. Although the fecal coliform test relates to shellfish harvesting, which is not the goal of this study, the laboratory test we use provides data for both fecal coliforms and *E. coli,* so we do report data for both here, as it also is useful for understanding contamination sources for downstream areas where shellfish harvesting is allowed. Analyses included negative and positive controls for each sampling day.

Water samples were filtered to capture bacterial cells and their DNA. Samples deemed polluted (elevated relative to State standards) were further analyzed by established procedures in our lab (Rothenheber and Jones 2018) to identify the presence/absence and, to some extent, relative quantification of sources of fecal contamination in the sample using PCR (polymerase chain reaction- presence/absence) and qPCR (semi-quantitative) methods. This procedure is called microbial source tracking (MST). The potential source species we have targeted include human, dog, bird, gull, Canada goose, cow, horse, ruminants and mammals for the presence/absence PCR assays, and mammal, human and bird for the semi-quantitative qPCR assays.

Water quality measurements were also made using datasondes with sensors for water temperature, salinity, pH, depth, dissolved oxygen, turbidity, and chlorophyll *a*. Data for daily rainfall amounts (inches) are from the UNH Weather daily statistics online database (https://www.weather.unh.edu).

Data analysis and development of data tables and figures were done on R Studio and Microsoft Excel. Geometric means were calculated for the three types of indicator bacteria for individual sites and for each month for all sites combined. ANOVAs were done between the monthly recorded rainfall values and monthly geometric average concentrations of all three indicator bacteria to test for differences between all 5 years of the study (2021-2025).

Basic comparisons were made of fecal indicator concentrations to those used as State water quality standards (Tab. 2; NHDES 20204a) to determine the frequency and location of areas that exceed the standards. Given the array of different standards for different types of uses and water quality classification, we used the 'Class A' freshwater and tidal water standards for comparisons. This is based on the recognition that recreational activities in the watershed often include both boating and swimming, so though the watershed has no designated beaches for which the standards are most strict, we need to inform potential risks for both activities.

	THRESHOLD RISK LEVEL- Primary Contact Recreation							
	Class A fresh		Class B fresh		Designated beaches		Tidal	
INDICATOR	SSMI*	GM	SSMI	GM	SSMI	GM	SSMI	GM
		# cfu or MPN/100 ml						
E. coli for freshwater recreational uses	**153**	**47**	**406**	**126**	**88**	**47**	N/A	N/A
Enterococci for marine water recreational uses	N/A	N/A	N/A	N/A	**104**	**35**	**104**	**35**
	THRESHOLD RISK LEVEL- Secondary Contact Recreation							
	Class A fresh		Class B fresh		Designated beaches		Tidal	
INDICATOR	SSMI*	GM	SSMI	GM	SSMI	GM	SSMI	GM
		# cfu or MPN/100 ml						
E. coli for freshwater recreational uses	**153**	**235**	**406**	**630**	N/A	N/A	N/A	N/A
Enterococci for marine water recreational uses	N/A	N/A	N/A	N/A	N/A	N/A	**520**	**175**

*SSMI = 'single sample maximum indicator'; GM = geometric mean, or the average of 3 samples within 60 days.

Table 2. State of New Hampshire standard fecal indicator bacteria concentrations for different surface water uses. See citation (State of New Hampshire) in **References** for the source of this information.

The microbial source tracking data were analyzed to determine occurrence and frequency of detection for the different sources at the different sites, noting any temporal trends. The concentrations (copy number per 100 ml) of the human source genetic marker in the qPCR assay are also compared to a threshold (2400 CN/100 ml) above which researchers at EPA and elsewhere have found to exceed acceptable likelihood of human illnesses (Boehm et al. 2015).

The awarded funds were used to support time required by Dr. Jones to oversee the project, analyze data, and write the Final Report. Four undergraduate students from UNH, 2 recently graduated students and Dr. Jones' Lab Supervisor were also partially supported for their involvement in sampling events and lab analyses. They also helped with data compilation and analysis and providing information for the final report. The project also required purchasing supplies for water sampling, bacterial analyses, and the pollution source detection analyses, and transportation to sampling sites.

RESULTS & DISCUSSION

Review and Summary of Existing Data

There are 2024 NHDES Watershed Report Cards for an approximate 34 square mile area representing the Lower and Middle portions of the Lamprey River (NHDES 2024b). These areas are given Hydrologic Unit Codes (HUC12) of HUC 12: 010600030709 (Lower) and 010600030703 (Middle). Within these areas there are 34 and 63 different Assessment Units respectively, each also given unique numerical Assessment IDs. In the Lower Lamprey River there were 2 estuarine, 6 impoundment, 1 lake and 24 river Assessment Units. Most (30 of 34) of these Assessment Units have assessment codes for swimming (Primary contact) or boating (Secondary contact) of "3-ND", which translates to: "No current data, insufficient information to make an assessment decision". The assessment codes for the study sites of assessment units closest to the study sites are all '3-ND' (last sample = 2008; Tab. 1), except for Site 1, which is at the mouth of Moonlight Brook where fresh and tidal water from the Lamprey River mix. The tidal portion of the Lamprey River in that area is classified as 5-P, while the classification of the whole Moonlight Brook is 3-ND. (Tab. 2).

In the Middle Lamprey River portion there were 8 impoundment, 8 lake and 47 river Assessment Units. Most (53 of 63) of these Assessment Units have assessment codes for swimming (Primary contact) or boating (Secondary contact) of "3-ND", which is "No current data, insufficient information to make an assessment decision". The assessment codes for the study sites of assessment units closest to the study sites are all '3-ND' except for Sites 5 and 6 where there are adequate *E. coli* data to classify primary contact (swimming) as poor water quality that does not meet water quality standards (4A-P). The secondary contact (boating) classification is '3-ND' for Sites 5 & 6 (Tab. 1).

Lamprey River Watershed

All intended sample collections occurred on 7 dates from May through November 2025. The bacterial indicator concentrations changed on different sample dates due to numerous causes. 2025 was a dry summer featuring only 4 intensive (>1 in./24 h) rainfall events (2 each in May and September), with numerous periods where no rain fell for more than a week. The wettest conditions for sample dates were August 13 (0.6" rainfall fell during 2 days prior to sampling) and October 20 (0.6" fell in previous day) (Tab. 3). Bacterial indicator concentrations were somewhat higher than on these two days, especially on August 13 when levels of all 3 indicators were at their highest concentrations for 2025 at most sites.

Table 3. Fecal indicator bacteria concentrations in water samples collected in the Lamprey River watershed. Site 1(MBO): Moonlight Brook-mouth at Lamprey River; Site 2 (PRBL): Piscassic River Boat Launch; Site 3 (WD): above Wiswall Dam; Site 4 (WF): Wadleigh Falls canoe access. Site 5 (ETH): Epping Town Hall. Site 6 (RES): Carroll Lake beach behind Raymond Elementary School.

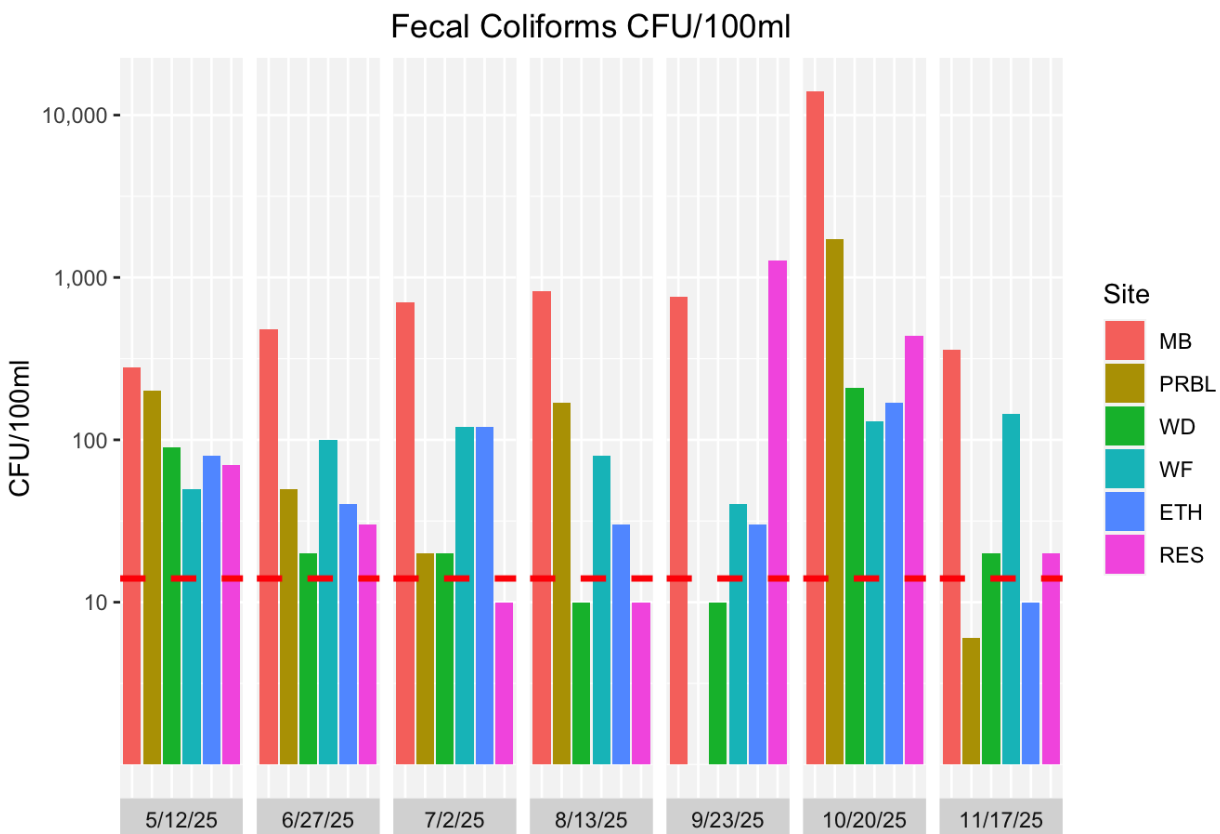
		Bacteria Counts (CFU/100mL)			Rainfall (inches/24hr)		
Date	Site	FC	EC	Ent	Day of Sample	1 Day Before	2 Days Before
5/12/25	MB	280	200	80	0	0	0
5/12/25	PRBL	200	200	100			
5/12/25	WD	90	80	70			
5/12/25	WF	50	40	30			
5/12/25	ETH	80	70	10			
5/12/25	RES	70	70	0			
6/27/25	MB	480	380	40	0	0.2	0
6/27/25	PRBL	50	50	0			
6/27/25	WD	20	20	0			
6/27/25	WF	100	90	0			
6/27/25	ETH	40	40	0			
6/27/25	RES	30	30	0			
7/2/25	MB	700	300	260	0	0.1	0
7/2/25	PRBL	20	10	0			
7/2/25	WD	20	20	0			
7/2/25	WF	120	100	30			
7/2/25	ETH	120	90	50			
7/2/25	RES	10	10	30			
8/13/25	MB	820	80	160	0	0.04	0.85
8/13/25	PRBL	170	10	30			
8/13/25	WD	10	0	90			
8/13/25	WF	80	50	10			
8/13/25	ETH	30	10	20			
8/13/25	RES	10	0	0			
9/23/25	MB	760	680	60	0	0.02	0.07
9/23/25	PRBL	0	0	40			
9/23/25	WD	10	5	0			
9/23/25	WF	40	30	20			
9/23/25	ETH	30	20	10			
9/23/25	RES	1280	960	0			
10/20/25	MB	14000	14000	3320	0	0.6	0
10/20/25	PRBL	1730	1730	890			
10/20/25	WD	210	210	10			
10/20/25	WF	130	130	30			
10/20/25	ETH	170	170	45			
10/20/25	RES	440	440	80			
11/17/25	MB	360	328	82	0.25	0	0
11/17/25	PRBL	6	5	9			
11/17/25	WD	20	20	0			
11/17/25	WF	145	145	0			
11/17/25	ETH	10	10	10			
11/17/25	RES	20	20	20			
		concentration exceeded state standard					

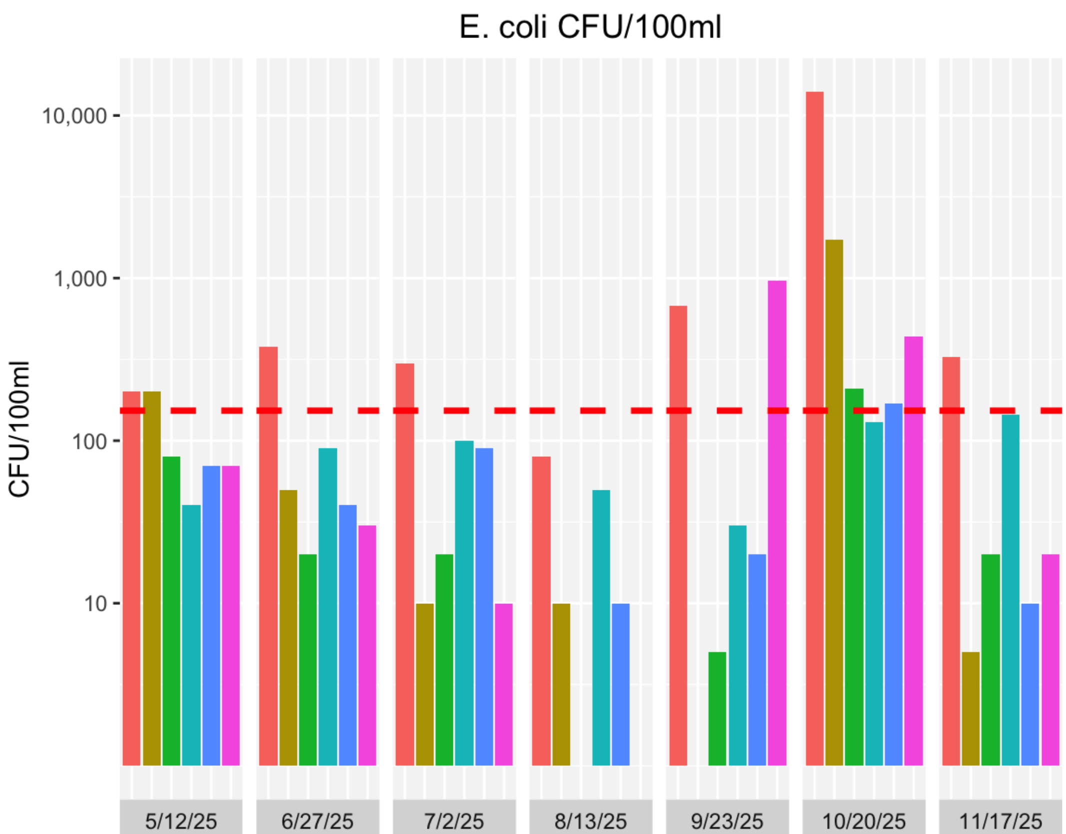
The three bacterial fecal indicators exceeded State water quality standards at varying frequencies (Tables 3 & 4). Fecal coliform levels exceeded the 14 FC/100ml standard in 35 of 42 samples. *E. coli* levels, which are most pertinent to this study as they relate to freshwater recreation, exceeded the State standard (153 *E. coli*/100 ml) on up to 6 of the 7 sample events at 5 sites for a total of 12 events (29%). Enterococci levels exceeded the State standard (104 enterococci/100 ml) on only 3 of the 7 sample events at up to 2 sites for a total of 4 of 42 events. Fecal coliforms and *E. coli* were detected at high frequencies, and non-detection occurred in 5-7% of samples. Enterococci were found less frequently and were not detected in 31% of samples. Non-detection occurred in over half of the sample months (Tab. 3 & 4). Fecal coliforms and *E. coli* were detected in a higher percentage of the samples in 2025 than in 2024.

Table 4. Frequency of exceedance of State water quality standards and non-detection of bacterial indicators at the 6 study sites.

2025	State Standard Exceedance			Non-Detection		
Site	**Fecal Coliforms**	***E. coli***	**Enterococci**	**Fecal Coliforms**	***E. coli***	**Enterococci**
	>14/100 ml	**>158/100ml**	**>104/100ml**	**<5 CFU/100ml**	**<5 CFU/100ml**	**<5 CFU/100ml**
MB	7	6	3	0	0	0
PRBL	5	2	1	1	1	2
WD	5	1	0	0	1	4
WF	7	0	0	0	0	2
ETH	6	1	0	0	0	1
RES	5	2	0	1	1	4
Totals	**35**	**12**	**4**	**2**	**3**	**13**
% of Samples	**83%**	**29%**	**10%**	**5%**	**7%**	**31%**

In all the studies conducted during 2021 through 2025, indicator bacteria were detected at much higher levels at Site 1 (MB) compared to all other sites (Jones 2024). In 2025, Site 1 continued to be contaminated at a relatively high rate and had the highest fecal coliform and *E. coli* single sample concentration (14,000/100 ml) as well as the highest enterococci concentration (3,320/100ml) (Fig. 3; Tables 3 & 4). In addition to the previously described impact of rainfall on indicator bacterial concentrations, there were also some general temporal trends. All three types of indicator bacteria were higher during June through October, and lower in May and November. Fecal coliforms increased gradually from May to October, while the changes for the other two types of indicator bacteria were more variable. Enterococci levels were generally lower than the other two indicator bacteria at all the sample sites and dates.


Fecal Coliforms CFU/100ml
CFU/100ml
10,000
1,000
100
10
5/12/25
6/27/25
7/2/25
8/13/25
9/23/25
10/20/25
11/17/25
Site
MB
PRBL
WD
WF
ETH
RES


E. coli CFU/100ml
CFU/100ml
10,000
1,000
100
10
5/12/25
6/27/25
7/2/25
8/13/25
9/23/25
10/20/25
11/17/25

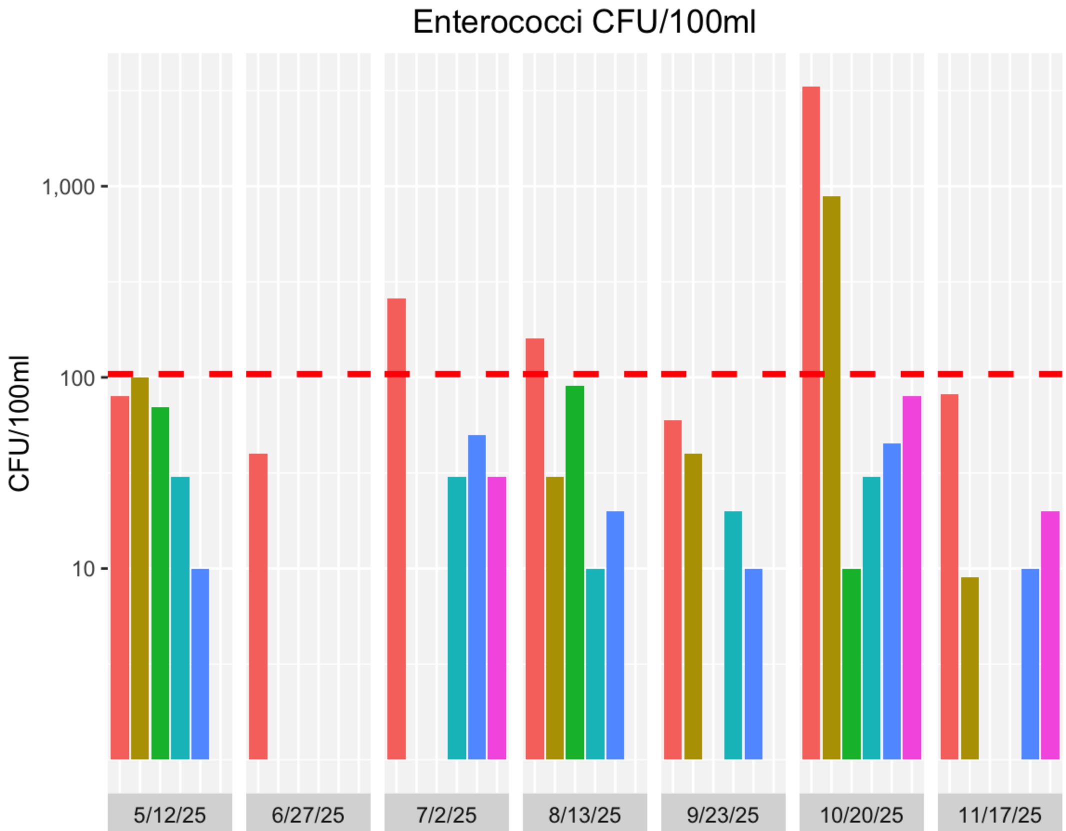


Figure 3. Concentrations of fecal coliforms, *E. coli*, and enterococci for all 7 sample dates at each of the 6 sampling sites. Horizontal lines indicate State standard limits.

As in past years (Jones 2022; 2023, 2024), the geometric mean concentrations for the fecal indicator bacteria for 2025 show differences between sites (Fig. 4). Fecal coliforms and *E. coli* average concentrations were once again highest at Site 1 and slightly elevated at Sites 4 & 6, while the relatively lower enterococci concentrations were highest at Site 1, and uniformly lower at the other sites. As in 2022, 2023, and 2024, the impact of rainfall and associated runoff was a focus of this 2025 study, but drought conditions dominated the weather which did not allow for capturing wet weather events to determine potential impacts.

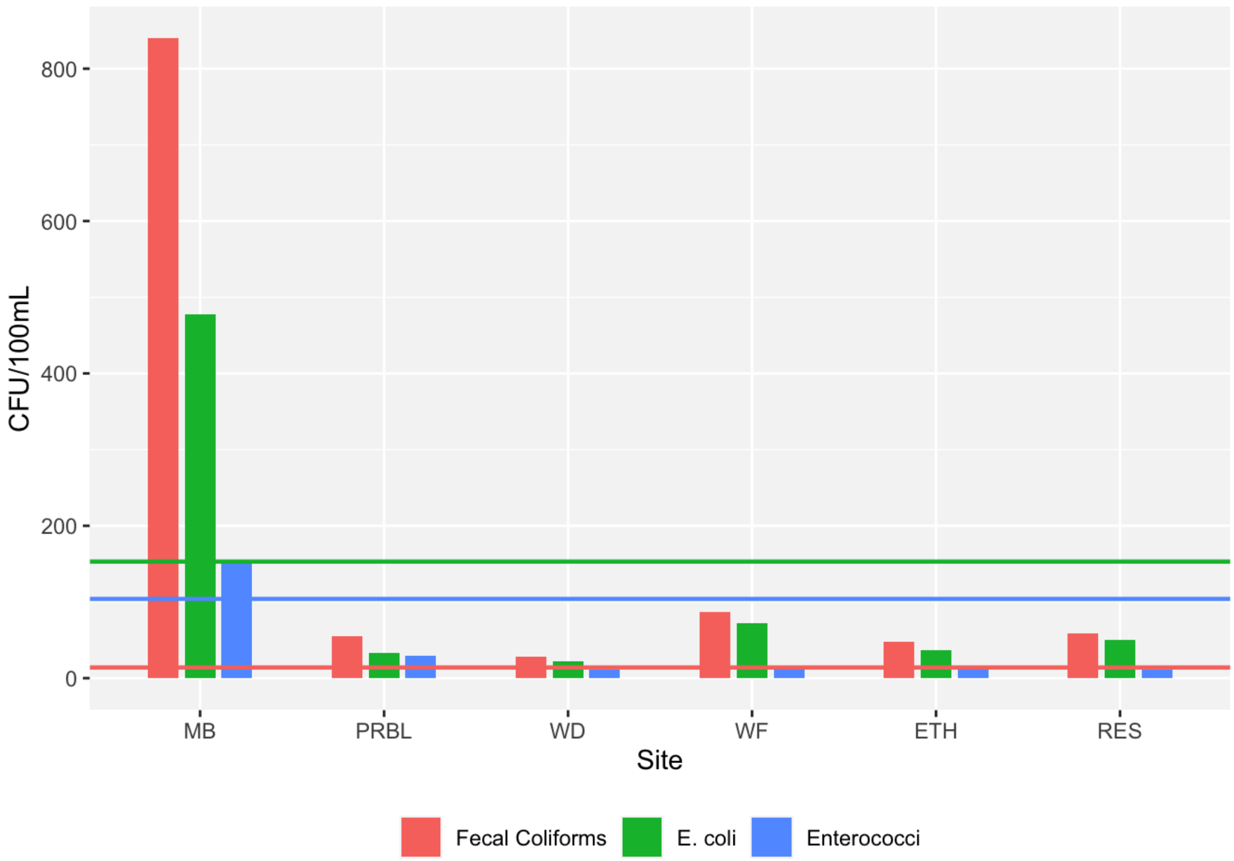


Figure 4. Geometric average concentrations (CFU/100 ml) of fecal indicator bacteria at the 6 sample sites for May to November 2025. Horizontal lines indicate State standard limits.

The bacterial indicator levels at a tidal site at the Newmarket waterfront just upstream from Site 1 (MB) measured by UNH-JEL for the GBNERR/Piscataqua Regional Estuaries Partnership (PREP) monitoring program for the past four years (2022, 2023, 2024, and 2025) were useful for comparisons to the 2025 results for the upstream watershed. In 2025, the levels of indicator bacteria exceeded State standards far less frequently than in previous years, with only the fecal coliform levels exceeding the standards and only on four sample dates. The 2025 concentrations are generally lower than in 2024 for all three fecal indicators, and especially lower than in 2023. In 2023, rainfall/runoff occurred more frequently and had the greatest total rainfall during sample events. There was also a sewage leak from a broken pipe under the upper tidal river in 2023.

Collection Date	Fecal Coliforms (CFU/100ml)	*E. coli* (CFU/100ml)	Enterococci (CFU/100ml)
5/17/22	<4	<4	12
6/21/22	29	25	8
7/18/22	40	36	16
8/15/22	20	12	24
9/19/22	NA	NA	NA
10/17/22	84	80	20
11/15/22	3240	3100	150
12/2/22	1200	1100	960
4/10/23	8440	8200	1440
5/8/23	120	106	6
6/6/23	3280	3200	440
7/19/23	373	310	30
8/7/23	220	220	50
9/18/23	208	204	12
10/16/23	30	30	<3
11/6/23	50	50	30
12/6/23	40	40	100
4/15/24	<10	<10	<10
5/13/24	40	40	8
6/11/24	60	60	100
7/9/24	40	40	40
8/5/24	230	230	120
9/9/24	17	13	43
11/5/24	28	28	2
12/9/24	8	8	<4
4/21/25	<4	<4	<4
5/20/25	NA	NA	NA
6/16/25	56	56	36
7/14/25	72	41	52
8/11/25	4	<4	52
9/15/25	4	4	4
10/27/25	8	8	4
11/12/25	18	8	10
12/1/25	16	16	4

Table 5. Fecal indicator bacteria concentrations in water samples collected at Site GBNERR-LR. Highlighted data are levels that exceed water quality standards.

There was evidence of animal (mammal) contamination at all 6 sites for all 33 analyzed samples (Tab. 6). This finding is common to all our related studies, and the mammal assay is useful as a positive control. Human contamination was detected only in 7 samples, 5 of them from Site 1 (MB) (Tab. 6), unlike in 2024 when it was detected only in 4 samples from 2 sites. Bird contamination was the most prevalent specific contamination source as it was detected in all but one sample. Ruminant and cow contamination were also present fairly frequently, showing up in 27 and 28 samples, respectively. Contamination from dogs was detected in 21 samples, and human and gull contamination was much less common. There was no detection of fecal contamination from horses in any of the samples. The positive controls in the lab used to test for Canada goose contamination were found to be faulty, so the data for Canada goose contamination was incomplete and this analytical issue continued to be under investigation during this study.

			PCR (Presence/Absence)									qPCR Markers (Copy #/100ml)		
Date	Site	MST vol	Mammal	Human	Dog	Ruminant	Cow	Bird	Gull	Horse	CG	Mammal	Human	Bird
5/12/25	MB	300 ml	+	+	-	+	-	+	-	-	NA		2.95E+03	1.98E+03
5/12/25	PRBL	300 ml	+	-	+	+	+	+	-	-	-		<267	1.77E+03
5/12/25	WD	300 ml	+	+	+	+	-	+	-	-	NA		-	1.87E+03
5/12/25	WF	300 ml	+	-	+	+	+	+	-	-	NA		-	1.83E+03
5/12/25	ETH	300 ml	+	+	+	+	+	+	-	-	NA		<267	2.00E+03
5/12/25	RES	300 ml	+	-	+	+	+	+	-	-	NA		-	1.93E+03
6/27/25	MB	300 ml	+	-	-	+	+	+	-	-	-		-	
6/27/25	PRBL	300 ml	+	-	+	-	+	-	-	-	-		-	
6/27/25	WD	300 ml	+	-	-	-	+	+	-	-	+		-	
6/27/25	WF	300 ml	+	-	-	+	+	+	-	-	+		-	
6/27/25	ETH	300 ml	+	-	+	+	+	+	-	-	-		-	
6/27/25	RES	300 ml	+	-	-	-	-	+	-	-	-		-	
7/2/25	MB	300 ml	+	-	+	+	+	+	-	-	+		-	
7/2/25	WD	300 ml	+	-	-	+	+	+	-	-	+		-	
7/2/25	WF	300 ml	+	-	+	+	-	+	-	-	-		-	
7/2/25	ETH	300 ml	+	-	+	+	+	+	-	-	-		-	
8/13/25	MB	300 ml	+	+	+	-	+	+	-	-	NA			
8/13/25	PRBL	300 ml	+	-	+	+	+	+	-	-	NA		-	
8/13/25	WF	300 ml	+	-	-	-	+	+	-	-	NA		-	
8/13/25	ETH	300 ml	+	-	+	-	-	+	-	-	NA		-	
9/23/25	MB	300 ml	+	+	-	+	+	+	+	-	NA			
9/23/25	WF	300 ml	+	-	+	+	+	+	-	-	NA			
9/23/25	ETH	300 ml	+	-	+	+	+	+	+	-	NA			
9/23/25	RES	300 ml	+	-	+	+	+	+	-	-	NA			
10/20/25	MB	300 ml	+	+	+	+	+	+	+	-	NA			
10/20/25	PRBL	300 ml	+	-	+	+	+	+	-	-	NA			
10/20/25	WD	300 ml	+	-	-	+	+	+	-	-	NA			
10/20/25	WF	300 ml	+	-	-	+	+	+	-	-	NA			
10/20/25	ETH	300 ml	+	-	-	+	+	+	+	-	NA			
10/20/25	RES	300 ml	+	-	+	+	+	+	-	-	NA			
11/17/25	MB	300 ml	+	+	+	+	+	+	+	-	NA			
11/17/25	WD	300 ml	+	-	-	+	+	+	+	-	NA			
11/17/25	WF	300 ml	+	-	+	+	+	+	-	-	NA			
		# of positive	33	7	21	27	28	32	6	0	NA			

Table 6. Detection of the presence of different pollution sources by PCR analyses for all samples from May through November 2025. Blue highlight denotes detection.

qPCR assays were run on some of the early samples where human and bird contamination were detected (Tab 6). Human contamination appeared to be below detectable levels by qPCR while bird contamination was consistently low at 1,770 to 2,000 copy number/100 ml. The presence of gull contamination did not correspond to more elevated levels of bird contamination. The remaining samples positive for human and bird contamination were saved for analysis in the fall of 2025, but the qPCR analysis equipment became inconsistent and finally unusable, so later sample analyses are on hold until replacement equipment arrives after a long-delayed delivery.

There was some seasonality for a few source types, while detection of bird, ruminant, dog, and cow contamination was spread across the full study period (Tab. 6). Gull contamination only occurred in the fall. Human contamination was slightly more prevalent in May, and consistent contamination at Site MB appeared in September, October, and November.

In addition to showing the highest concentrations of all three indicator bacteria, Site 1 (MB) also had a slightly higher diversity for identified types of contamination, with an average of 4.4 types (out of 8 possible, excluding mammal) per sample (Tab. 7). In 2024, this analysis showed a lower diversity of contamination types at all sites, more frequent detection of human contamination by PCR and qPCR, and one instance of human contamination that was above the safety threshold (Jones 2024).

Site	# of Samples	Avg # of Source Types Detected	Human Source Detection	Human Source >Threshold
MB	7	4.4	5	0
PRBL	4	3.5	0	0
WD	5	3.2	1	0
WF	7	3.3	0	0
ETH	6	4.0	1	0
RES	4	3.3	0	0

Table 7. The frequency of site-specific fecal-borne bacterial contamination sources.

<u>Moonlight Brook watershed</u>

A significant focus of the 2025 study was to continue exploration of how contaminated Moonlight Brook is in relation to the historically elevated levels of bacterial contamination at Site 1-MB/MBO located at the mouth of Moonlight Brook next to the Newmarket boat launch (Figs. 1 & 2). The Moonlight Brook watershed sites were sampled during 5 of the 7 months of this study (excluding September and November) on different days than the sites on the full Lamprey River watershed. The Moonlight Brook watershed sites included 3 upstream of the downtown railroad crossing, and 2 more sites in a small tributary waterway to the south of the brook outlet that crosses New Road and that extends into the Sleepy Hollow trailer park. A drainage pipe that discharges into Moonlight Brook at the Site 1 sampling area was also sampled once, when enough drainage was available.

All intended sample collections occurred on 5 dates in May through October 2025 (Tab. 8). Like the Lamprey River watershed, 2025 conditions were also dry in the Moonlight Brook watershed. There were no instances where sampling occurred on a date following significant (>1") rainfall within 2 days prior to the sample dates, and only two of the sample dates had rainfall (0.36" or less) in the previous two days. Statistical analysis showed that there was no significant difference between the fecal coliform (p=0.2095), *E. coli* (p=0.1640), and enterococci (p=0.6680) concentrations from the sample dates that had rainfall in the prior days vs those that did not. Concentrations of all three bacterial indicators were higher and more often exceeded State risk thresholds during July and were at the lowest concentrations in May (Tab. 8). Comparing 2025 to previous years, there were no significant differences in concentrations of the indicator bacteria between the years since the study of the Moonlight Brook Watershed began. Though the most rain fell during 2023, the lack of significant differences in yearly bacteria concentrations prevent us from forming strong conclusions about the effect of rainfall on the bacterial contamination of the MB Watershed.

		Bacteria Counts (CFU/100mL)			Rainfall (inches/24hr)		
Date	**Site**	**FC**	**EC**	**Ent**	**Day of Sample**	**1 Day Before**	**2 Days Before**
5/28/25	MBO	140	140	40	0	0	0
5/28/25	MBpipe	40	30	50			
5/28/25	CD	50	50	40			
5/28/25	NR	90	90	60			
5/28/25	MBD	140	140	80			
5/28/25	MBRec	70	70	30			
5/28/25	MBU	30	30	30			
6/3/25	MBO	280	200	40	0	0	0
6/3/25	CD	160	160	20			
6/3/25	NR	80	80	55			
6/3/25	MBD	280	235	110			
6/3/25	MBRec	46800	45600	8000			
6/3/25	MBU	80	80	30			
7/16/25	MBO	680	510	120	0	0	0.36
7/16/25	CD	4800	4400	200			
7/16/25	NR	2800	2320	100			
7/16/25	MBD	500	440	30			
7/16/25	MBRec	200	200	100			
7/16/25	MBU	445	405	95			
8/6/25	MBO	370	130	110	0	0	0
8/6/25	CD	240	80	140			
8/6/25	NR	1600	800	200			
8/6/25	MBD	240	80	95			
8/6/25	MBRec	140	100	40			
10/9/25	MBO	340	340	260	0.26	0	0
10/9/25	CD	2360	2080	240			
10/9/25	NR	480	480	340			
10/9/25	MBD	250	250	40			
10/9/25	MBRec	160	140	10			
10/9/25	MBU	200	120	0			

Table 8. Fecal indicator bacteria concentrations in water samples collected in the Moonlight Brook watershed. Site MBO: Moonlight Brook Outlet-mouth at Lamprey River; Site CD: Columbia Drive,

upstream of New Road; Site NR: New Road 3; Site MBD: Moonlight Drive upstream of the railroad crossing; Site MBRec: Moonlight Brook behind the Newmarket High School near the recreational facilities; and Site MBU: Moonlight Brook upstream.

The three bacterial fecal indicators exceeded State water quality standards to varying degrees (Tables 8 & 9). Fecal coliform concentrations exceeded the standard (14 FC/100 ml) in all 30 samples. *E. coli* levels exceeded the single sample standard (153 *E. coli*/100 ml) in 47% of the samples including all 6 sites in July. Enterococci levels exceeded the State standard (104 enterococci/100 ml) in 33% of samples. The bacterial indicators were detected at a high frequency, with fecal coliforms and *E. coli* always detected, and non-detection occurring only 1 time for enterococci on the October sample date (Tab. 9).

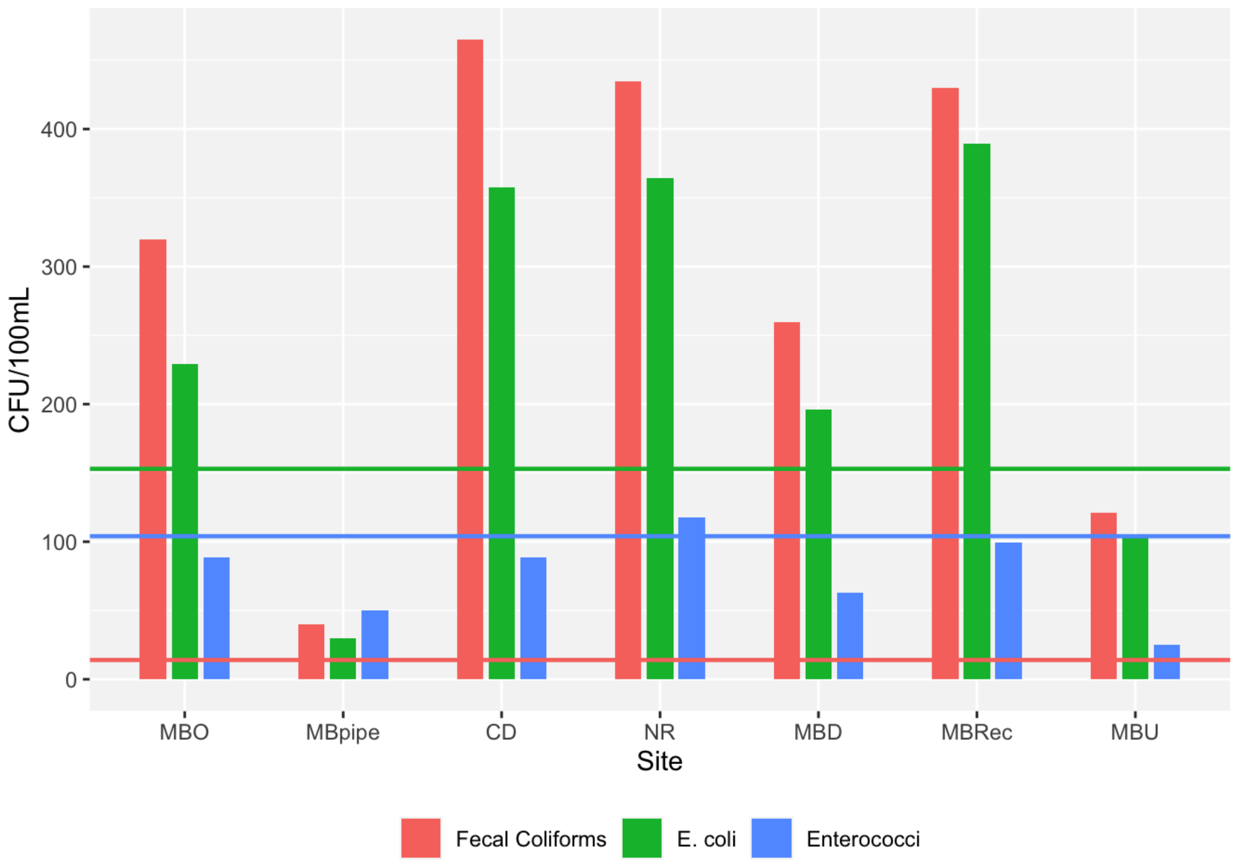
2025	State Standard Exceedance			Non-Detection		
Site	**Fecal Coliforms**	***E. coli***	**Enterococci**	**Fecal Coliforms**	***E. coli***	**Enterococci**
	>14/100 ml	**>158/100ml**	**>104/100ml**	**<5 CFU/100ml**	**<5 CFU/100ml**	**<5 CFU/100ml**
MBO	5	3	3	0	0	0
MBpipe	1	0	0	0	0	0
CD	5	2	3	0	0	0
NR	5	3	2	0	0	0
MBD	5	3	1	0	0	0
MBRec	5	2	1	0	0	0
MBU	4	1	0	0	0	1
Totals	**30**	**14**	**10**	**0**	**0**	**1**
% of Samples	**100%**	**47%**	**33%**	**0%**	**0%**	**3%**

Table 9. Frequency of exceedance of State water quality standards and non-detection of bacterial indicators at the 7 study sites in the Moonlight Brook watershed.

The geometric mean concentrations for the fecal indicator bacteria show trends across the 7 sites in Figure 6. Fecal coliform and *E. coli* concentrations were higher at the MBO, CD, NR, and MBRec sites. Enterococci concentrations were highest at Site NR, slightly lower at MBRec, CD, and MBO, and even lower at the last three sites. These results contrast with the 2024 study results where most of the sites had similar levels of fecal coliforms and *E. coli*, and MBU had clearly the highest levels of enterococci. Overall, indicator levels in 2025 were similar to those in 2024 but showed more variability.

The geometric mean concentrations for the fecal indicator bacteria show trends across the 6 sites in Figure 6. Fecal coliform and *E. coli* concentrations were relatively equal across all sites except Site MBU, which had the highest geometric mean in 2024, and MBpipe, where both were substantially lower in 2025. Enterococci concentrations were highest at Site NR but overall, there were relatively equal levels at all sites. These results are relatively similar to results reported for the 2024 study results, except that the relatively low levels of all 3 indicators at MBU were back to low levels is a new development. Site MBO is at the mouth of both watersheds.

Figure 6. Geometric average concentrations (CFU/100 ml) of fecal indicator bacteria at the 7 sample sites in the Moonlight Brook watersheds: May-October 2025. Horizontal lines are State standard limits.



There was evidence of animal (mammal) contamination at all 7 sites based on all the 29 samples analyzed (Tab. 10). Human contamination was present in only 8 of the analyzed samples and was not found at all in May and July and was found most frequently at the MBO and MBD sites. Human contamination was detected at least once for three other sites in the Moonlight Brook watershed, but not at all at the MBRec site or the single sample collected in a pipe discharge (MBpipe) at Site MBO. Bird contamination was present in all of the samples analyzed. Cow contamination was found in 22 out of the 29 samples, and dog and ruminant contamination were both found in 18. Gull contamination was only found during the fall in the full LR watershed, and here in the MB watershed in 4 samples during October. Horse contamination was only found once in June at the MBRec site.

			PCR (Presence/Absence)									qPCR Markers (Copy #/100ml)		
Date	Site	MST vol	Mammal	Human	Dog	Ruminant	Cow	Bird	Gull	Horse	CG	Mammal	Human	Bird
5/28/25	MBO	300 ml	+	-	+	-	-	+	-	-	-		-	2.22E+03
5/28/25	MBpipe	300 ml	+	-	-	-	+	+	+/-	-	-		-	1.75E+03
5/28/25	CD	300 ml	+	-	+	-	+	+	-	-	-		-	2.89E+03
5/28/25	NR	300 ml	+	-	-	+	+	+	-	-	-		-	
5/28/25	MBD	300 ml	+	-	-	+	-	+	-	+/-	-		-	3.49E+03
5/28/25	MBU	300 ml	+	-	-	+	+	+	-	-	-		-	2.10E+03
6/3/25	MBO	300 ml	+	-	+	+	+	+	-	-	-		-	2.46E+03
6/3/25	CD	300 ml	+	+	+	+	+	+	-	-	-		<267	
6/3/25	NR	300 ml	+	+	+	+	-	+	-	-	-		<267	
6/3/25	MBD	300 ml	+	+	+	+	+	+	-	-	+		<267	
6/3/25	MBRec	300 ml	+	-	-	+	+	+	-	+	-		-	
6/3/25	MBU	300 ml	+	+	+	-	-	+	-	-	-		4.83E+03	
7/16/25	MBO	300 ml	+	-	+	+	+	+	-	-	+/-		-	
7/16/25	CD	300 ml	+	-	-	+	-	+	-	-	+/-		-	
7/16/25	NR	300 ml	+	-	+	+	+	+	-	-	+		-	
7/16/25	MBD	300 ml	+	-	-	+	+	+	-	-	+		-	
7/16/25	MBRec	300 ml	+	-	+	-	-	+	-	-	+		-	
7/16/25	MBU	300 ml	+	-	-	-	+	+	-	-	+		-	
8/6/25	MBO	300 ml	+	+	+	-	+	+	-	-	NA			
8/6/25	CD	300 ml	+	-	+	-	+	+	-	-	NA		-	
8/6/25	NR	300 ml	+	-	-	-	+	+	-	-	NA			
8/6/25	MBD	300 ml	+	+	-	-	+	+	-	-	NA		-	
8/6/25	MBRec	300 ml	+	-	-	+	+	+	-	-	NA		-	
10/9/25	MBO	300 ml	+	+	+	+	+	+	+	-	NA			
10/9/25	CD	300 ml	+	-	+	-	+	+	-	-	NA			
10/9/25	NR	300 ml	+	-	+	+	-	+	+	-	NA			
10/9/25	MBD	300 ml	+	+	+	+	+	+	+	-	NA			
10/9/25	MBRec	300 ml	+	-	+	+	+	+	-	-	NA			
10/9/25	MBU	300 ml	+	-	+	+	+	+	+	-	NA			
		# of positive	29	8	18	18	22	29	4	1	NA			

Table 10. Detection of the presence of different pollution sources by PCR and qPCR analyses for all samples from May through October 2025. Blue highlight denotes detection; red highlight denotes level above or nearly equal to a human contamination risk threshold level.

The follow-up semi-quantitative assay (qPCR), which has a higher detection limit than the presence/absence PCR assay, indicated that the early samples positive for human contamination were at low levels (below detection limit) except at Site MBU where the level of human contamination was 4,830 copy number/100 ml), above a public health safety threshold (4,200 copy number/100 ml; Boehm et al. 2015) in 1 sample collected on June 3rd (Tab. 10). The relatively common detection of human contamination at MBO, and the frequent human contamination at MBD remain to be concerns.

The quantified level of bird contamination for some of the samples where bird contamination was also detected by the non-quantitative PCR assay reflected relatively low levels of contamination, as none of the samples detectable levels exceeded 10,000 CN/100 ml, with the highest level at 3,490 CN/100 (Tab. 10). The presence of gulls did not correspond to elevated levels of bird contamination.

There was some seasonality for a few source types (Tab. 10). Gulls were detected only in October, and human contamination was detected most widely in June. The other sources of contamination were found pretty evenly across the study months, except for horse contamination which was only found once.

Sites MBO and MBD had slightly more diverse identified types of contamination, with an average of 4.0 and 3.8 types (out of 8 possible), respectively, per sample (Tab. 11). The MBPipe site, that was only sampled once, had the lowest diversity with 2 sources of contamination. All other sites had similar average numbers of types (3.2 to 3.3) that were between the low and higher averages. Again, human contamination was detected at 5 of 7 sites at levels below the safety threshold except for the 1 sample at Site MBU.

Site	# of Samples	Avg # of Source Types Detected	Human Source Detection	Human Source >Threshold
MBO	5	4.0	2	0
MBpipe	1	2.0	0	0
CD	5	3.2	1	0
NR	5	3.2	1	0
MBD	5	3.8	3	0
MBRec	4	3.3	0	0
MBU	4	3.25	1	1

Table 11. The frequency of site-specific fecal-borne bacterial contamination sources within the Moonlight Brook watershed.

Significant Findings, Accomplishments and Next Steps

This study represents an up-to-date and comprehensive summary of the sanitary water quality conditions in the Lower and Middle Lamprey River watershed. This is important as New Hampshire rivers, streams and impoundments are increasingly used by boaters and swimmers, who may be at risk for water-borne illnesses under contaminated conditions.

The detailed review of existing data on microbial pollution in the watershed showed that very few of the assessment units had any available or recent data to provide water quality assessments for swimming and boating uses. The findings from this study are useful as a starting point for all watershed users and groups like LRAC to communicate with NHDES and other agencies about where to focus potential monitoring that could provide data to inform protecting people involved in recreational uses from water-borne illnesses. The new database generated by this study represents a continuation of a 4-year synoptic dataset for 6 key sites in the watershed related to recreational uses and thus serves as a start for continued monitoring and water quality assessments.

The continued exploration of water quality assessment to the Moonlight Brook sub-watershed provides context for previous detection of consistent and elevated levels of bacterial contamination at Site MBO. In 2025, levels of FC and *E. coli* were high again, similar to 2021 levels, despite infrequent rainfall events. This may, in part, be a result of upstream sources of pollution. There was some evidence of potential pollution sources upstream, like at Sites NR, CD and MBRec where bacterial indicators were detected at levels higher than at Site MBO. There may also be some sources of contamination to MBO from downtown portions of the brook. There has been an effort to upgrade sewage system infrastructure, including a section of sewer pipe on New Road suspected of having leaks. The elevated levels at CD suggest upstream contamination, including humans in June, is occurring.

The concerns about elevated bacterial contamination at Site 6/RES in Raymond during 2023 and 2024 were less evident in 2025. Levels of fecal coliforms and *E. coli* were relatively low although Site 6 had the highest concentration of these 2 indicators during September and *E. coli* levels exceeded the State standard twice.

The abundant rainfall in 2023 allowed for assessment of the impacts of rainfall and associated runoff on bacterial contamination in the Lamprey River and Moonlight Brook watersheds. In 2024 and 2025, no samples were collected after substantial rainfall events, yet contamination was still present at somewhat lower overall levels. Thus, water quality concerns are not only tied to rainfall/runoff conditions as indicated by previous comparisons of the 2023 samples with previous year samples.

Microbial source tracking is an invaluable tool for assessing watershed water quality, as it shows what sources are contributing to contamination and where resources for eliminating pollution sources should be used. Human sources are the highest priority/of most concern. Site 1/MBO continued to be a consistent concern due to elevated bacterial indicator concentrations accompanied by consistent detection of elevated levels of human contamination. The lack of detection of human contamination at all other Lamprey River sites except for Sites 3/WD and 4/ETH during May was encouraging. In the Moonlight Brook watershed, the frequent (3 samples) detection of human contamination at Site MBD is a concern as it had human contamination in 2 samples during 2024. The sources of human contamination are not yet apparent, so the towns along the Lamprey River and Moonlight Brook will need to conduct further investigations to pinpoint the sources. More in-depth sampling at sites upstream and following rainfall events could help with that process.

The next most manageable source is probably dogs. Dog contamination was consistently present at all sites in both 2022 and 2023, though not prevalent in 2024. In 2025, detection was more sporadic on different sample dates and at all sites. These multi-year results suggest runoff from rainfall events may exacerbate water quality contamination from dogs. Several management approaches are typical for reducing the significance of this source including signage that is located at water access points (all sites in this study) that alerts dog owners to pick up and dispose of dog feces, plus the provision of dog feces collection bags at the signage locations. The

NHDES has a Scoop the Poop Campaign webpage that can help: https://www.des.nh.gov/home-and-recreation/your-health-and-environment/pet-health-and-environment.

The LRAC will be able to use the findings to help communicate to recreational users about potential water quality issues and precautions to be taken. These were delineated in a separate 2-page document provided in Jones (2022) that is based on NH Dept. of Health and Human Services/Division of Public Health Services and US CDC fact sheets and information (NHDES 2019a).

Future work could take several directions, the most obvious being a continuation of routine monitoring for bacterial pollution indicators at key sites. This is especially important as the Town of Newmarket conducted 2 new sewage infrastructure projects in 2025. One dimension that remains uncaptured is the duration of impacts of rainfall and associated runoff, a condition that is now known to be widely responsible for elevated levels of bacterial pollution in the coastal watersheds of NH. Typically, watersheds impacted by runoff-borne contamination require one to several days before elevated levels of contamination are transported out of the system. Five years of data reflecting both dry and wet conditions provides a solid baseline to compare with future findings that hopefully can include more rainy-condition results. As our regional climate continues to change, rainfall patterns are expected to become more extreme and may change the dynamics of bacterial contamination levels and types of contamination sources, including birds and animal migration patterns that are influenced by climate change. The differences in rainfall frequency and amounts over the past 3 years exemplify these points.

This Final Report will be made available to key people involved in the PREP and GBNERR monitoring programs, the Town of Newmarket, as well as water quality managers and the Shellfish Program Manager in NHDES.

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