

Bacterial Source Tracking Analyses to Support Virginia's TMDLs

Non-Shellfish Stations

Prepared by

MapTech, Inc.

in cooperation with

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1. INTRODUCTION

EPA's document, *Guidance for Water Quality-Based Decisions: The TMDL Process* (USEPA, 1999) states:

According to section 303(d) of the Clean Water Act and EPA water quality planning and management regulations, States are required to identify waters that do not meet or are not expected to meet water quality standards even after technology-based or other required controls are in place. The water bodies are considered water quality-limited and require TMDLs.

. . . A TMDL, or total maximum daily load, is a tool for implementing State water quality standards and is based on the relationship between pollution sources and in-stream water quality conditions. The TMDL establishes the allowable loadings or other quantifiable parameters for a water body and thereby provides the basis for States to establish water quality-based controls. These controls should provide the pollution reduction necessary for a water body to meet water quality standards.

The purpose of this project is to use bacterial source tracking (BST) to identify sources of *E. coli* to support the development of Fecal Bacteria TMDLs for impaired segments in Virginia. In fulfilling the state requirement for the development of a TMDL, a systematic process will be utilized to establish the maximum allowable Bacteria loading for each waterbody to meet the applicable standard, allocate that load among pollutant contributors, and provide a basis for taking actions needed to restore water quality. This report focused on water quality sampling conducted in non-shellfish waters. A companion document will be published later this year to report the results of water quality sampling in shellfish waters. Together, these reports reflect the fourth year of BST sampling conducted by VADEQ (2005-2006).

Bacterial Source Tracking (BST) methods can be subdivided into three basic groups: Molecular, Biochemical, and Chemical. Molecular (genotype) are typically referred to as

"DNA fingerprinting" and are based on the unique genetic makeup of different strains, or subspecies, of fecal bacteria. Biochemical (phenotype) methods are based on an effect of an organism's genes that actively produce a biochemical substance. The type and quantity of these substances produced under various conditions is what is actually measured. Chemical methods are based on finding chemical compounds that are associated with human wastewaters, and generally are restricted to determining if sources of pollution are human or not.

Two techniques were used to determine sources of fecal bacteria for this study. Hagedorn's (Hagedorn et al., 1999) Antibiotic Resistance Analysis (ARA) technique was used because it has been demonstrated to be a reliable procedure for confirming the presence of human, livestock, wildlife and pet sources. Compared to DNA fingerprinting, biochemical profiling is much quicker, typically allows for many more isolates to be analyzed (*e.g.*, hundreds per week vs. a few dozen per week for DNA analysis), is more economical, has survived limited court testing, and has undergone rigorous peer review from the scientific community. Additionally, observation of an increased number of isolates allows for an estimate of the relative proportions of the fecal indicator (*e.g.*, *E. coli*) originating from different sources. Fluorometric analysis was also used to determine the concentration of optical brighteners. Optical brighteners are used in laundry and dishwasher detergent, as well as toilet paper. Their presence in high levels indicates the likely presence of human wastewater

2. OBJECTIVES

As described in Chapter 1, two types of BST were used in this study; a fluorometric technique was used to detect human sewage, and ARA was used to identify sources of *E. coli* as well as the relative percentage contribution from source groups (*e.g.*, livestock, wildlife, human and pets) to support the development of Fecal Bacteria TMDLs for impairments located throughout Virginia. BST results will be used to improve public awareness of the problem, improve model calibration/validation of bacteria densities, and provide a more equitable allocation of loads to source classes. Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported. This change is due to changes in the fecal bacteria standard and economic considerations.

The specific objectives of the project were to:

1. collect fecal samples from known sources in 10 areas, based on Hydrologic Unit Codes (HUCs),
2. use collected samples to develop a known-source library for each impairment area, and
3. perform bacterial enumerations and BST analyses on whole water samples from impaired segments, using the libraries developed for objective 2.

3. METHODS

Hagedorn's ARA method has been extensively and successfully used by MapTech, and separates fecal sources based on patterns of antibiotic resistance in the *enterococci* or *E. coli*. For this study, *E. coli* was the indicator organism analyzed. The premise of ARA is that fecal bacteria from each source (*e.g.*, human, livestock, wildlife, and pets) will have different resistance patterns to the battery of antibiotics and concentrations used in the analysis. Hagedorn's method for *E. coli* tests each isolate on 28 different combinations of antibiotic type and concentration. Confidence in BST techniques is measured by the level of separation of isolates from known sources, represented as the percentage of isolates that are accurately separated into respective source types (*e.g.*, Average Rate of Correct Classification – ARCC). Additional analyses can be applied to test the specificity of the library. These analyses are discussed further in Section 4 of this document. The ARA method, like other methods (*e.g.*, molecular), requires the collection of source samples from feces of known sources to build a source library. In support of this study, known source samples from the four source classes were collected, analyzed, and entered into known-source libraries. Additionally, a fluorescence spectrophotometer was used to quantify the concentration of optical brighteners in each water sample.

3.1 Collection of Known Sources

Known source samples were collected in ten HUCs associated with fecal-bacteria impaired waters throughout Virginia (Figure 3.1). In HUCs where known-source samples had not previously been collected to support VADEQ's BST program (newly sampled HUCs), a total of 60 samples were collected. Watersheds within HUCs with some known source sampling completed within the past two years will require the collection and inclusion of, at minimum, 20 known source samples (160 isolates). Watersheds within HUCs with some source sampling but none within the past two years will require the collection and inclusion of, at minimum, 40 known source samples (320 isolates). Each set of source samples was distributed evenly between human, livestock, wildlife, and pets (Table 3.1). Specific species within each source category (*e.g.*, deer, raccoon, poultry, beef, etc.) that were selected to represent the sources in each region

were identified through field observation, discussion with local stakeholders, and review of available data (e.g., Virginia Agricultural Statistics). From each sample, 8 isolates were analyzed using BST to create a known-source library of 480 isolates for each newly sampled HUC, and to increase known-source libraries by 160 isolates in updated HUCs. To date, approximately 3,451 fecal samples have been collected to support VADEQ’s BST program, resulting in over 25,547 isolates analyzed. In total 486 fecal samples were collected for this study, resulting in 2,915 isolates analyzed.

Table 3.1 Source samples collected for BST library development.

Source	Source Species	Number of Samples Collected in Newly Sampled HUCs	Additional Samples Collected in Updated HUCs
Human	Septic Systems, Portable Toilets, ...	15	5
Livestock	Dairy, Beef, Horse, Sheep, Broilers, Turkeys, Swine, Waste Storage Pits, ...	15	5
Wildlife	Deer, Raccoon, Muskrat, Duck, Goose, ...	15	5
Pets	Dogs & Cats	15	5
Total		60	20

3.2 Development of Known-Source Libraries

An appropriate known-source library was selected for each of the impairments to complete objective 2. A predictive model was developed from each library using logistic regression. A known-source library must be large enough to prevent an over-specified fit to the library. However, known-source responses to ARA analyses have been observed to vary geographically. The characteristics of this variance have not been well defined, so the regional libraries developed for this study were combined in a stepwise procedure and analyzed to measure the resulting specificity and the predictive accuracy of the combined libraries, as detailed in Section 4 of this document.

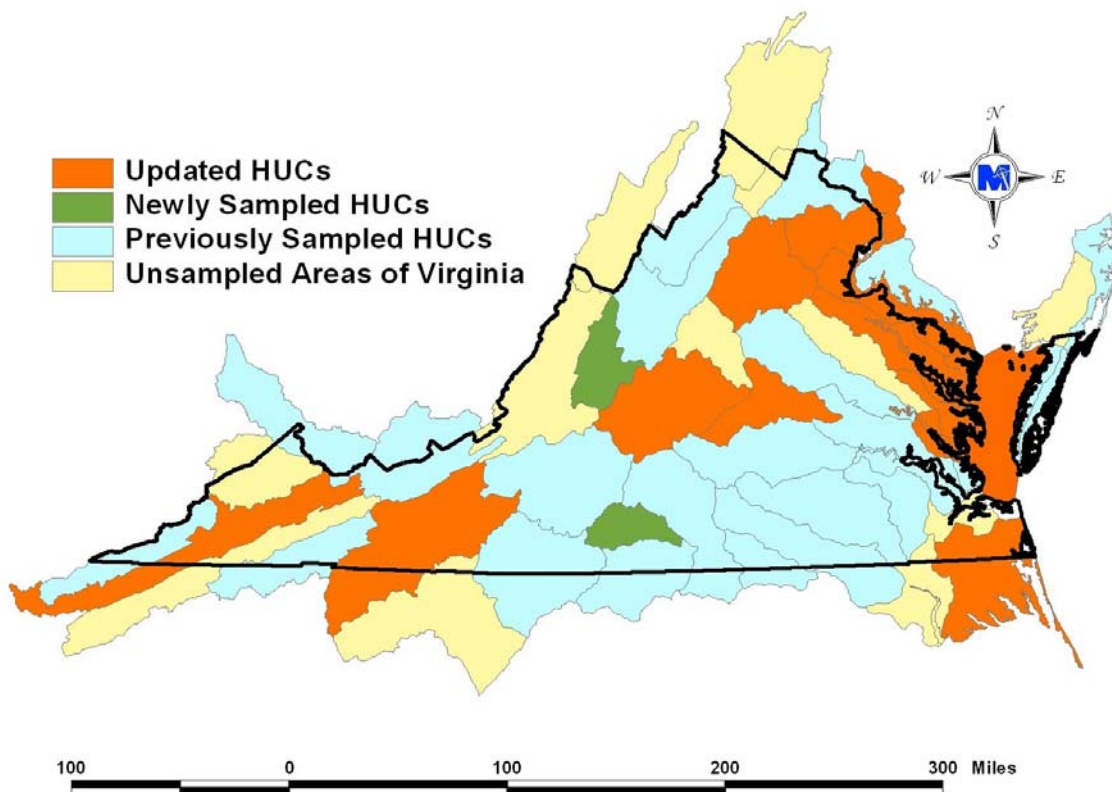


Figure 3.1 Locations of known-source sampling conducted to support this year’s and previous years’ BST analyses.

3.3 Bacterial Enumerations and BST Analyses

For objective 3, water quality monitoring sites were identified and sampled by the granting agency (Figure 3.2 and Table 3.2). For many sites, the contract began in July 2005. At the conclusion of the study, all sites will have been sampled as often as monthly for one year. Samples were received as whole-water samples (*i.e.*, ambient sampling as presented in Table 3.2). All water samples were analyzed for *E. coli*. BST was run on bacteria isolated from the whole-water samples. Bacteria were analyzed using Hagedorn's ARA methodology, yielding the percentage of isolates classified as human, livestock, wildlife, and pets. Up to 24 bacterial isolates were analyzed per sample, limited only by the number of isolates available from the enumeration process.

Additionally, water samples were analyzed using a fluorescence spectrophotometer to determine the concentration of optical brighteners.

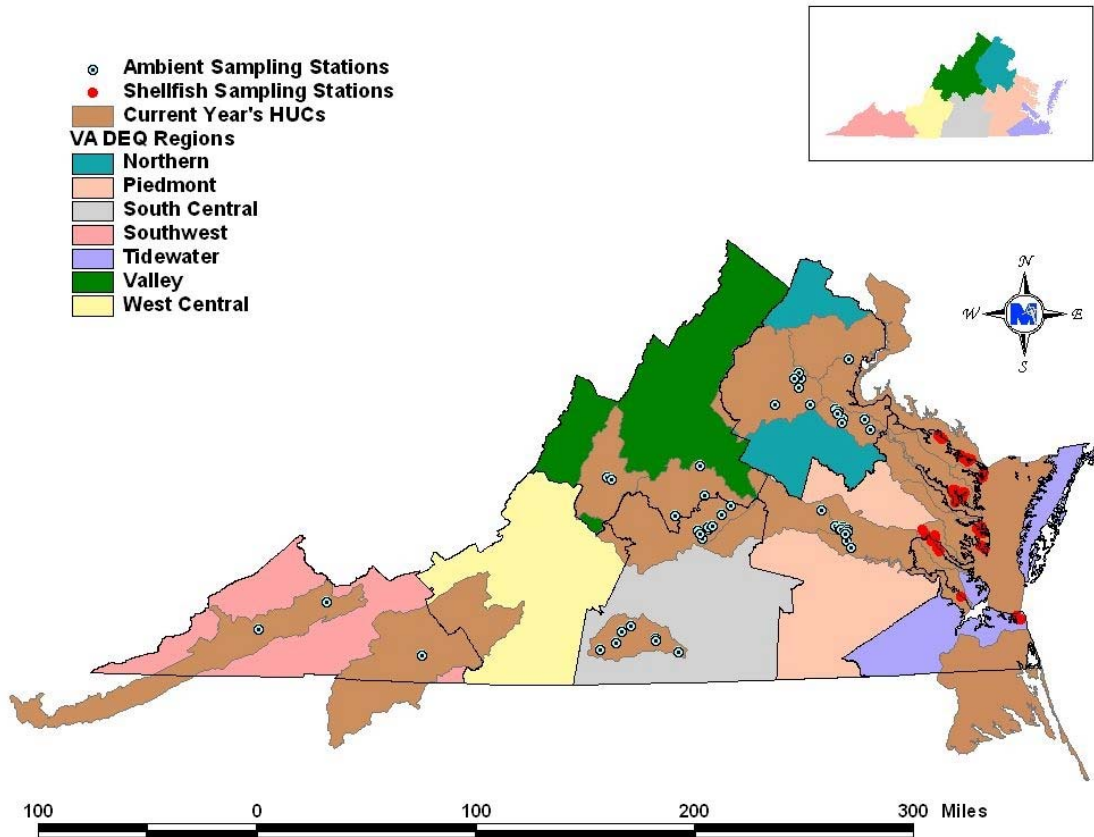


Figure 3.2 Spatial distribution of impaired segments identified by region.

Table 3.2 Distribution of ambient sampling stations addressed in this study.

Waterbody	Hydrologic Unit	BST Stations
Indian Creek	6010205	6BIDI001.49
Lick Creek	6010205	6BLCC000.09
Hays Creek	2080202	2HYS001.41
Ballinger Creek	2080203	2BLR003.00
North Fork Hardware River	2080203	2HNF008.28
Rock Island Creek	2080203	2RKI003.40
Cedar Grove Branch	2080202	2CGB001.80
Slate River	2080203	2-SLT003.68
Slate River	2080203	2-SLT030.19
Slate River	2080203	2-SLT014.52
Slate River	2080203	2-SLT036.92
Austin Creek	2080203	2-AUS001.12
Frisby Branch	2080203	2-FRY000.35
North River	2080203	2-NTH001.65
Sharps Creek	2080203	2-SHR004.96
Troublesome Creek	2080203	2-TBM000.80
Turpin Creek	2080203	2-TPN003.59
Banister River	3010105	4ABAN070.20
Banister River	3010105	4ABAN023.28
Banister River	3010105	4ABAN005.58
Cherrystone Creek	3010105	4ACRR000.80
Whitehorn Creek	3010105	4AWRN005.50
Stinking River	3010105	4ASNE005.30
Sandy Creek	3010105	4ASNA000.20
Neabsco Creek	2070010	1ANEA002.89
Marsh Run	2080103	3-MAH000.19
Browns Run	2080103	3-BOS000.72
Craig Run	2080103	3-CRA000.46
Rappahannock River	2080103	3-RPP147.10
Cedar Run	2080103	3-CED000.59
Rapidan River	2080103	3-RAP006.53
Rappahannock	2080104	3-RPP110.57
Rappahannock	2080104	3-RPP104.47
Rappahannock	2080104	3-RPP080.19
Rappahannock	2080104	3-RPP091.55
Claiborne Run	2080104	3-CLB000.50
Hazel Run	2080104	3-HAL000.57
Massaponax Creek	2080104	3-MAP002.61
James River	2080205	2-JMS115.29
James River	2080205	2-JMS112.79
James River	2080205	2-JMS112.33
James River	2080205	2-JMS111.47
James River	2080205	2-JMS111.17
James River CSO	2080205	2-RDD000.19
Tidal James River	2080206	2-JMS104.16
Tidal James River	2080206	2-JMS099.30
Gillies Creek	2080206	2-GIL001.00
Almond Creek	2080206	2-ALM000.42
Goode Creek	2080206	2-GOD000.77
Chestnut Creek	5050001	9CST002.64
Chestnut Creek	5050001	9CST016.82

4. KNOWN-SOURCE LIBRARY DEVELOPMENT

As discussed in Section 3, a predictive model was developed from each library (HUC) using logistic regression. Where a previously developed library existed (*i.e.*, updated HUCs), this year's data was combined with the existing data and the updated library was used for further assessment. These regional libraries were combined in a stepwise procedure and analyzed to measure the resulting specificity and the predictive accuracy of the combined libraries. The specificity and predictive accuracy were assessed through three analyses. First, the ARCC was calculated for the library. Second, a randomization test was performed by randomly assigning source categories to samples and assessing the ARCC for the randomized library. Twenty-five randomizations were performed and the results averaged. The expected result of randomization of four source categories is an ARCC of 25%, indicating a completely random result. Greater values for the randomized ARCC indicate a more specified model. Third, a jackknifing routine was conducted, where data from each whole fecal sample were individually withheld during development of the statistical model. The model was then tested for predictive accuracy on the withheld sample. In combining regional libraries, a balance was sought between minimizing the randomized ARCC and maximizing the jackknifed ARCC. Table 4.1 shows the resulting analyses on the finalized libraries, and how the libraries were applied to the analysis of whole-water samples by the HUC in which they were sampled.

Table 4.1 Results of known-source library development.

Known-Source Library	ARCC (%)	Randomized ARCC (%)	Jackknifed ARCC (%)
02080103	76.18%	44.15%	61.10%
02070010	72.38%	45.24%	51.89%
02080104	69.57%	46.11%	53.78%
02080203	75.32%	36.91%	71.07%
02080205	73.23%	39.02%	62.62%
02080206	70.37%	39.58%	59.98%
02080202	67.47%	34.80%	61.83%
03010105	65.35%	35.53%	58.31%
06010205	74.01%	42.47%	59.88%
05050001	82.37%	47.45%	70.98%

5. RESULTS

The results of the water quality analyses for VADEQ's 2005-2006 BST sampling in non-shellfish waters are reported in this section. Fecal coliform enumerations, *E. coli* enumerations, and the results of the BST analyses are reported. Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported. The *E. coli* enumerations are reported with the BST results to give an indication of the bacteria concentration at the time of sampling.

The bacteria source proportions reported are formatted to indicate statistical significance (*i.e.*, **BOLD** numbers indicate a statistically significant result). The statistical significance was determined through two tests. The first was based on the sample size. A z-test was used to determine if the proportion was significantly different from zero ($\alpha = 0.10$). During the second test, the rate of false positives was calculated for each source category in each library, and a proportion was not considered significantly different from zero unless it was greater than the false-positive rate plus three standard deviations.

Optical brighteners concentrations (fluorometry results) are reported with the bacteria enumerations. As a rule of thumb, less than 50 ppb signifies little or no indication of human wastewater contamination. Between 50 ppb and 100 ppb signifies a potential for human wastewater contamination, which should be investigated if there is corroborating evidence (*e.g.*, high human proportions, aging infrastructure, or anecdotal evidence of illicit discharges). Over 100 ppb signifies likely contamination from human waste streams.

5.4 Results for Northern Region

The results of the water quality analyses for VADEQ's Northern Region (Figure 5.4) are reported in the following tables. Table 5.56 indicates the number of samples analyzed in the 2005-2006 sampling phase. Bacterial enumerations and optical brightener concentrations are reported in Tables 5.57 through 5.70. The results of the BST analysis are reported in Tables 5.71 through 5.84.

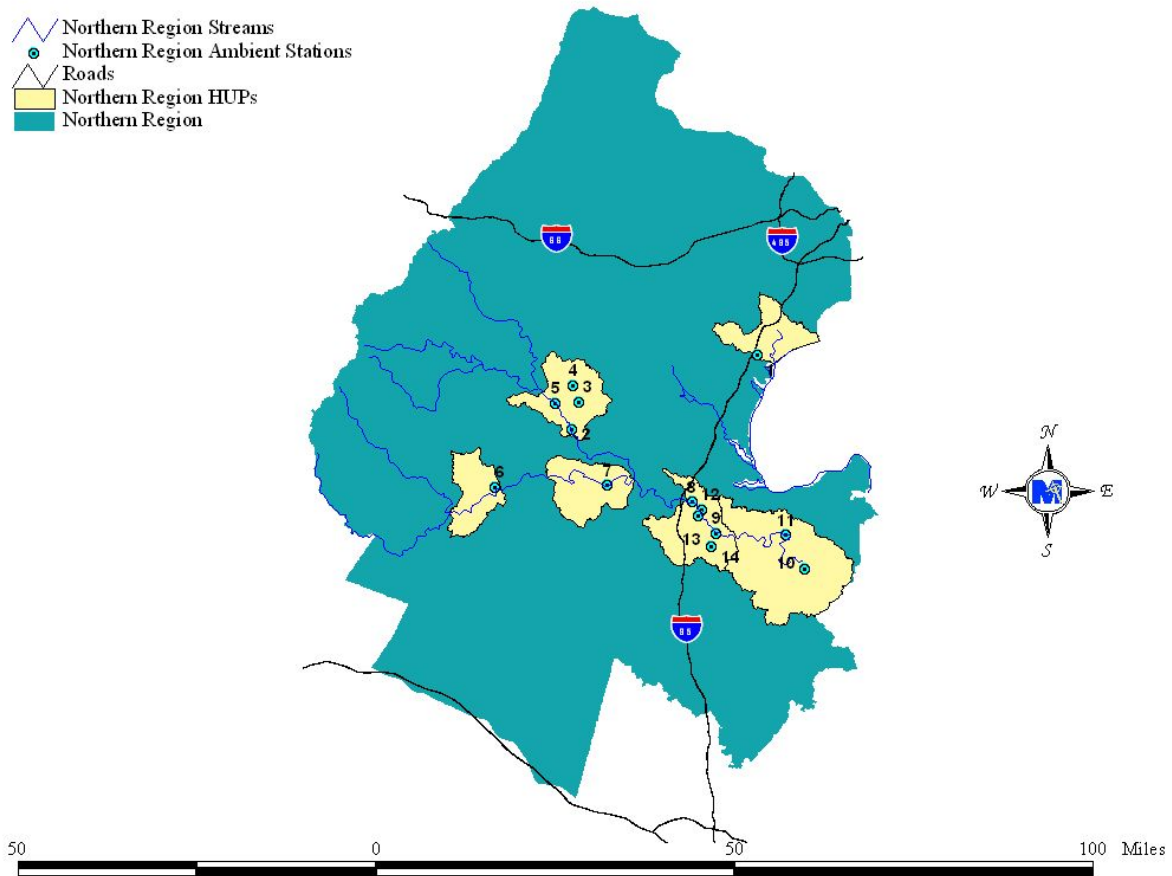


Figure 5.4 Bacterial sampling stations in VADEQ's Northern Region.

Table 5.56 Summary of bacterial sampling in VADEQ's Northern Region.

Station Number	Station ID	HUP	County / City	Stream Name	# of Samples Received	% Violations for <i>E. Coli</i>
1	1ANEA002.89	A25	Prince William	Neabsco Creek	12	17%
2	3-MAH000.19	E08	Fauquier	Marsh Run	12	8%
3	3-BOS000.72	E08	Fauquier	Browns Run	12	33%
4	3-CRA000.46	E08	Fauquier	Craig Run	11	36%
5	3-RPP147.10	E08	Culpeper	Rappahannock River	12	0%
6	3-CED000.59	E16	Culpeper	Cedar Run	12	0%
7	3-RAP006.53	E18	Culpeper	Rapidan River	12	8%
8	3-RPP110.57	E20	Fredericksburg	Rappahannock	12	17%
9	3-RPP104.47	E20	Fredericksburg	Rappahannock	11	18%
10	3-RPP080.19	E21	Caroline	Rappahannock	12	0%
11	3-RPP091.55	E21	Caroline	Rappahannock	12	8%
12	3-CLB000.50	E20	Stafford	Claiborne Run	12	33%
13	3-HAL000.57	E20	Fredericksburg	Hazel Run	12	25%
14	3-MAP002.61	E20	Spotsylvania	Massaponax Creek	12	33%

Table 5.57 Bacterial Enumeration for Neabsco Creek at Station 1ANEA002.89.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
1ANEA002.89	7/20/2005	D4578	7/21/2005	96		330		41.40		7/25/2005	DM
1ANEA002.89	8/24/2005	D4706	8/25/2005	48		200		49.80		8/29/2005	DMT
1ANEA002.89	9/27/2005	D4825	9/28/2005	96		280		156.00		9/30/2005	DMT
1ANEA002.89	10/26/2005	D4921	10/27/2005	254	A			104.00		10/28/2005	DMT
1ANEA002.89	11/29/2005	D5013	11/30/2005	36	B			129.60		12/2/2005	DMT
1ANEA002.89	12/21/2005	D5103	12/22/2005	80				99.90		12/29/2005	DMT
1ANEA002.89	1/24/2006	D5245	1/25/2006	92				98.30		2/14/2006	DMT
1ANEA002.89	2/21/2006	D5381	2/22/2006	4	B			88.20		2/24/2006	DMT
1ANEA002.89	3/28/2006	D5568	3/29/2006	10	B			86.50		3/31/2006	DMT
1ANEA002.89	4/19/2006	D5704	4/20/2006	64				81.50		4/26/2006	DMT
1ANEA002.89	5/9/2006	D5776	5/10/2006	186	A			109.9		5/15/2006	DMT
1ANEA002.89	6/21/2006	D6026	6/22/2006	320				129.6		6/26/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported..

Table 5.58 Bacterial Enumeration for Marsh Run at Station 3MAH000.19.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3MAH000.19	7/6/2005	D4523	7/7/2005	72		50	B			7/22/2005	DM
3MAH000.19	8/17/2005	D4659	8/18/2005	50		310		88.10		8/23/2005	DMT
3MAH000.19	9/14/2005	D4769	9/15/2005	12	B	50	B	116.00		9/17/2005	DMT
3MAH000.19	10/18/2005	D4877	10/19/2005	22	B			121.00		10/24/2005	DMT
3MAH000.19	11/15/2005	D4976	11/17/2005	14	B			118.90		11/18/2005	DMT
3MAH000.19	12/12/2005	D5041	12/13/2005	287	A			138.10		12/16/2005	DMT
3MAH000.19	1/9/2006	D5137	1/10/2006	157	A			95.70		1/17/2006	DMT
3MAH000.19	2/7/2006	D5305	2/8/2006	78				116.10		2/9/2006	DMT
3MAH000.19	3/14/2006	D5496	3/15/2006	56				71.70		3/18/2006	DMT
3MAH000.19	4/11/2006	D5644	4/12/2006	98	L			97.20		4/14/2006	DMT
3MAH000.19	5/2/2006	D5756	5/3/2006	74				110.10		5/9/2006	DMT
3MAH000.19	6/13/2006	D5961	6/14/2006	56	L			98.60		6/16/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.59 Bacterial Enumeration for Browns Run at Station 3BOS000.72.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3BOS000.72	7/6/2005	D4520	7/7/2005	82		100	B			7/22/2005	DM
3BOS000.72	8/17/2005	D4657	8/18/2005	125	A	260		27.20		8/23/2005	DMT
3BOS000.72	10/18/2005	D4874	10/19/2005	308	A			88.40		10/24/2005	DMT
3BOS000.72	11/15/2005	D4977	11/17/2005	44				145.50		11/18/2005	DMT
3BOS000.72	12/12/2005	D5043	12/13/2005	348	A			152.50		12/16/2005	DMT
3BOS000.72	1/9/2006	D5138	1/10/2006	74				107.50		1/17/2006	DMT
3BOS000.72	2/7/2006	D5306	2/8/2006	80				124.80		2/9/2006	DMT
3BOS000.72	3/14/2006	D5497	3/15/2006	132				106.80		3/18/2006	DMT
3BOS000.72	4/11/2006	D5645	4/12/2006	104				127.30		4/14/2006	DMT
3BOS000.72	5/2/2006	D5757	5/3/2006	330				127.3		5/9/2006	DMT
3BOS000.72	6/13/2006	D5962	6/14/2006	700				116.9		6/16/2006	MAF
3BOS000.72	7/25/2006	D6165	7/26/2006	20	B			158.2		7/28/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.60 Bacterial Enumeration for Craig Run at Station 3CRA000.46.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3CRA000.46	7/6/2005	D4521	7/7/2005	243	A	200				7/22/2005	DM
3CRA000.46	10/18/2005	D4875	10/19/2005	54				103.00		10/24/2005	DMT
3CRA000.46	11/15/2005	D4978	11/17/2005	20	B			104.30		11/18/2005	DMT
3CRA000.46	12/12/2005	D5039	12/13/2005	630				108.40		12/16/2005	DMT
3CRA000.46	1/9/2006	D5139	1/10/2006	76				70.60		1/17/2006	DMT
3CRA000.46	2/7/2006	D5307	2/8/2006	48				95.60		2/9/2006	DMT
3CRA000.46	3/14/2006	D5498	3/15/2006	68				83.90		3/18/2006	DMT
3CRA000.46	4/11/2006	D5646	4/12/2006	26	L			114.80		4/14/2006	DMT
3CRA000.46	5/2/2006	D5758	5/3/2006	24	B			113.00		5/9/2006	DMT
3CRA000.46	6/13/2006	D5963	6/14/2006	510	L			125.10		6/16/2006	MAF
3CRA000.46	7/25/2006	D6166	7/26/2006	310	L			180.4		7/28/2006	MAF
3CRA000.46	10/3/2006	D6451	10/4/2006	34	B			122.4		10/6/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.61 Bacterial Enumeration for Rappahannock River at Station 3RPP147.10.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3RPP147.10	7/6/2005	D4522	7/7/2005	220	A	160	B			7/22/2005	DM
3RPP147.10	8/17/2005	D4660	8/18/2005	84		1175	A	73.50		8/23/2005	DMT
3RPP147.10	9/14/2005	D4770	9/15/2005	100		130	B	32.50		9/17/2005	DMT
3RPP147.10	10/18/2005	D4878	10/19/2005	74				37.80		10/24/2005	DMT
3RPP147.10	11/15/2005	D4979	11/17/2005	60				47.00		11/18/2005	DMT
3RPP147.10	12/12/2005	D5040	12/13/2005	163	A			40.50		12/16/2005	DMT
3RPP147.10	1/9/2006	D5140	1/10/2006	60				29.50		1/17/2006	DMT
3RPP147.10	2/7/2006	D5308	2/8/2006	30	B			37.50		2/9/2006	DMT
3RPP147.10	3/14/2006	D5499	3/15/2006	56				33.30		3/18/2006	DMT
3RPP147.10	4/11/2006	D5647	4/12/2006	42				35.90		4/14/2006	DMT
3RPP147.10	5/2/2006	D5759	5/3/2006	94				92.10		5/9/2006	DMT
3RPP147.10	6/13/2006	D5964	6/14/2006	42				41.90		6/16/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.62 Bacterial Enumeration for Cedar Run at Station 3CED000.59.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100 ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3CED000.59	7/12/2005	D4533	7/13/2005	60		110	B		Sample was depleted at time of filtering	7/22/2005	DM
3CED000.59	8/3/2005	D4641	8/4/2005	36	B	60	B	33.60		8/5/2005	DMT
3CED000.59	9/7/2005	D4744	9/8/2005	18	B	520		81.20		9/9/2005	DMT
3CED000.59	10/19/2005	D4900	10/20/2005	34	B			68.70		10/26/2005	DMT
3CED000.59	11/16/2005	D4995	11/17/2005	48				80.90		11/18/2005	DMT
3CED000.59	12/19/2005	D5100	12/20/2005	210	A			90.70		1/18/2006	DMT
3CED000.59	1/10/2006	D5147	1/11/2006	32	B			62.00		1/27/2006	DMT
3CED000.59	2/14/2006	D5354	2/15/2006	28	B			92.80		2/20/2006	DMT
3CED000.59	3/22/2006	D5536	3/23/2006	8	B			53.70		3/27/2006	DMT
3CED000.59	4/26/2006	D5733	4/27/2006	134				122.3		5/9/2006	DMT
3CED000.59	5/31/2006	D5894	6/1/2006	22	B			94.8		6/5/2006	MAF
3CED000.59	7/11/2006	D6081	7/12/2006	88				124.7		7/19/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.63 Bacterial Enumeration for Rapidan River at Station 3RAP006.53.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3RAP006.53	7/12/2005	D4532	7/13/2005	96		70	B	48.00		7/22/2005	DM
3RAP006.53	8/3/2005	D4640	8/4/2005	30	B	20	B	122.00		8/5/2005	DMT
3RAP006.53	9/7/2005	D4743	9/8/2005	28	B	50	B	34.10		9/9/2005	DMT
3RAP006.53	10/19/2005	D4899	10/20/2005	56				31.30		10/26/2005	DMT
3RAP006.53	11/16/2005	D4994	11/17/2005	60				45.70		11/18/2005	DMT
3RAP006.53	12/19/2005	D5101	12/20/2005	223	A			57.40		1/18/2006	DMT
3RAP006.53	1/10/2006	D5146	1/11/2006	140	A			33.20		1/17/2006	DMT
3RAP006.53	2/14/2006	D5353	2/15/2006	60				76.00		2/20/2006	DMT
3RAP006.53	3/22/2006	D5535	3/23/2006	6	B			31.00		3/27/2006	DMT
3RAP006.53	4/26/2006	D5732	4/27/2006	310				52.30		5/9/2006	DMT
3RAP006.53	5/31/2006	D5893	6/1/2006	24	B			50.60		6/5/2006	MAF
3RAP006.53	7/11/2006	D6080	7/12/2006	58				50		7/19/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.64 Bacterial Enumeration for Rappahannock River at Station 3RPP110.57.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3RPP110.57	7/20/2005	D4574	7/21/2005	92		110	B		Sample was depleted at time of filtering	7/25/2005	DM
3RPP110.57	8/24/2005	D4702	8/25/2005	56		340		103.00		8/29/2005	DMT
3RPP110.57	9/27/2005	D4821	9/28/2005	68		290		34.50		9/30/2005	DMT
3RPP110.57	10/26/2005	D4920	10/27/2005	328	A			113.00		10/28/2005	DMT
3RPP110.57	11/29/2005	D5012	11/30/2005	62				46.30		12/2/2005	DMT
3RPP110.57	12/21/2005	D5104	12/22/2005	60				49.80		12/29/2005	DMT
3RPP110.57	1/24/2006	D5244	1/25/2006	356	A			86.00		1/27/2006	DMT
3RPP110.57	2/21/2006	D5380	2/22/2006	4	B			35.70		2/24/2006	DMT
3RPP110.57	3/28/2006	D5567	3/29/2006	8	B			27.90		3/31/2006	DMT
3RPP110.57	4/19/2006	D5703	4/20/2006	116				56.70		4/26/2006	DMT
3RPP110.57	5/9/2006	D5775	5/10/2006	22	B			43.70		5/15/2006	DMT
3RPP110.57	6/21/2006	D6022	6/22/2006	82				41.90		6/26/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.65 Bacterial Enumeration for Rappahannock River at Station 3RPP104.47.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3RPP104.47	7/7/2005	D4526	7/8/2005	42		60	B	71.00		7/22/2005	DM
3RPP104.47	9/6/2005	D4740	9/7/2005	22	B	70	B	43.20		9/9/2005	DMT
3RPP104.47	10/18/2005	D4879	10/19/2005	256	A			40.70		10/24/2005	DMT
3RPP104.47	11/1/2005	D4937	11/2/2005	28	B			75.70		11/7/2005	DMT
3RPP104.47	12/12/2005	D5045	12/13/2005	388	A			63.10		12/16/2005	DMT
3RPP104.47	2/8/2006	D5314	2/9/2006	46				51.00		2/14/2006	DMT
3RPP104.47	3/8/2006	D5452	3/9/2006	6	B			50.60		3/13/2006	DMT
3RPP104.47	4/5/2006	D5595	4/6/2006	6	B			41.80		4/7/2006	DMT
3RPP104.47	5/3/2006	D5763	5/4/2006	70				40.10		5/9/2006	DMT
3RPP104.47	6/20/2006	D5978	6/21/2006	2	B			48.90		6/26/2006	MAF
3RPP104.47	8/9/06	D6230	8/10/06	2	B			66.3		8/11/06	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.66 Bacterial Enumeration for Rappahannock River at Station 3RPP080.19.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3RPP080.19	7/7/2005	D4524	7/8/2005	6	B	190	B	57.50		7/22/2005	DM
3RPP080.19	8/9/2005	D4642	8/10/2005	22	B	40	B	79.30		8/13/2005	DMT
3RPP080.19	9/6/2005	D4741	9/7/2005	16	B	30	B	61.90		9/9/2005	DMT
3RPP080.19	10/18/2005	D4873	10/19/2005	180	A			90.40		10/24/2005	DMT
3RPP080.19	11/1/2005	D4935	11/2/2005	66				90.30		11/7/2005	DMT
3RPP080.19	12/12/2005	D5042	12/13/2005	108				47.70		12/16/2005	DMT
3RPP080.19	1/4/2006	D5122	1/5/2006	76				64.10		1/11/2006	DMT
3RPP080.19	2/8/2006	D5312	2/9/2006	136	A			61.30		2/14/2006	DMT
3RPP080.19	3/8/2006	D5450	3/9/2006	2	B			48.30		3/13/2006	DMT
3RPP080.19	4/5/2006	D5593	4/6/2006	8	B			41.30		4/7/2006	DMT
3RPP080.19	5/3/2006	D5761	5/4/2006	24	B			81.30		5/9/2006	DMT
3RPP080.19	6/20/2006	D5976	6/21/2006	6	B			51.60		6/26/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.67 Bacterial Enumeration for Rappahannock River at Station 3RPP091.55.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3RPP091.55	7/7/2005	D4525	7/8/2005	10	B	120	B	66.70		7/22/2005	DM
3RPP091.55	8/9/2005	D4643	8/10/2005	12	B	80	B	14.90		8/13/2005	DMT
3RPP091.55	9/6/2005	D4742	9/7/2005	28	B	60	B	68.40		9/9/2005	DMT
3RPP091.55	10/18/2005	D4876	10/19/2005	74				60.50		10/24/2005	DMT
3RPP091.55	11/1/2005	D4936	11/2/2005	114				99.10		11/7/2005	DMT
3RPP091.55	12/12/2005	D5044	12/13/2005	246	A			48.50		12/16/2005	DMT
3RPP091.55	1/4/2006	D5123	1/5/2006	141	A			85.80		1/11/2006	DMT
3RPP091.55	2/8/2006	D5313	2/9/2006	210	A			83.80		2/14/2006	DMT
3RPP091.55	3/8/2006	D5451	3/9/2006	1	U			33.80		3/13/2006	DMT
3RPP091.55	4/5/2006	D5594	4/6/2006	12	B			36.80		4/7/2006	DMT
3RPP091.55	5/3/2006	D5762	5/4/2006	20	B			53.00		5/9/2006	DMT
3RPP091.55	6/20/2006	D5977	6/21/2006	20	B			52.30		6/26/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.68 Bacterial Enumeration for Claiborne Run at Station 3CLB000.50.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3CLB000.50	7/20/2005	D4575	7/21/2005	279	A	1120	A	50.10		7/25/2005	DM
3CLB000.50	8/24/2005	D4703	8/25/2005	114		350		52.80		8/29/2005	DMT
3CLB000.50	9/27/2005	D4822	9/28/2005	142	A	390		47.80		9/30/2005	DMT
3CLB000.50	10/26/2005	D4919	10/27/2005	300	A			80.50		10/28/2005	DMT
3CLB000.50	11/29/2005	D5011	11/30/2005	90				57.70		12/2/2005	DMT
3CLB000.50	1/24/2006	D5243	1/25/2006	66				71.50		1/27/2006	DMT
3CLB000.50	2/21/2006	D5379	2/22/2006	12	B			41.10		2/24/2006	DMT
3CLB000.50	3/28/2006	D5566	3/29/2006	34	B			38.60		3/31/2006	DMT
3CLB000.50	4/19/2006	D5702	4/20/2006	94				59.30		4/26/2006	DMT
3CLB000.50	5/9/2006	D5774	5/10/2006	128				64.6		5/15/2006	DMT
3CLB000.50	6/21/2006	D6023	6/22/2006	400				56.5		6/26/2006	MAF
3CLB000.50	7/25/2006	D6164	7/26/2006	250				62.7		7/28/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.69 Bacterial Enumeration for Hazel Run at Station 3HAL000.57.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3HAL000.57	7/20/2005	D4577	7/21/2005	940	L	400		48.80		7/25/2005	DM
3HAL000.57	8/24/2005	D4705	8/25/2005	242	A	1750	A	61.70		8/29/2005	DMT
3HAL000.57	9/27/2005	D4824	9/28/2005	212	A	1500	B	65.70		9/30/2005	DMT
3HAL000.57	10/26/2005	D4917	10/27/2005	219	A			71.20		10/28/2005	DMT
3HAL000.57	11/29/2005	D5009	11/30/2005	54				56.80		12/2/2005	DMT
3HAL000.57	12/21/2005	D5105	12/22/2005	32	B			47.40		12/29/2005	DMT
3HAL000.57	1/24/2006	D5241	1/25/2006	64				70.00		1/27/2006	DMT
3HAL000.57	2/21/2006	D5378	2/22/2006	6	B			38.30		2/24/2006	DMT
3HAL000.57	3/28/2006	D5564	3/29/2006	32	B			38.00		3/31/2006	DMT
3HAL000.57	4/19/2006	D5701	4/20/2006	199	A			57.20		4/26/2006	DMT
3HAL000.57	5/9/2006	D5772	5/10/2006	230				73.50		5/15/2006	DMT
3HAL000.57	6/21/2006	D6025	6/22/2006	310				63.20		6/26/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.70 Bacterial Enumeration for Massaponax Creek at Station 3MAP002.61.

Station ID	Date of Sample	Lab ID	Lab-In Date	<i>E. coli</i> cfu/100 ml	Quality	Fecal Coliform cfu/100ml	Quality	Optical Brighteners /ppb	Comments	Lab-Out Date	Lab Personnel
3MAP002.61	7/20/2005	D4576	7/21/2005	880	L	1550	A	49.60		7/25/2005	DM
3MAP002.61	8/24/2005	D4704	8/25/2005	58		300		92.70		8/29/2005	DMT
3MAP002.61	9/27/2005	D4823	9/28/2005	24	B	70	B	75.90		9/30/2005	DMT
3MAP002.61	10/26/2005	D4918	10/27/2005	236	A			72.60		10/28/2005	DMT
3MAP002.61	11/29/2005	D5010	11/30/2005	4	B			55.90		12/2/2005	DMT
3MAP002.61	12/21/2005	D5106	12/22/2005	4	B			48.40		12/29/2005	DMT
3MAP002.61	1/24/2006	D5242	1/25/2006	254	A			75.90		1/27/2006	DMT
3MAP002.61	2/21/2006	D5377	2/22/2006	2	B			40.60		2/24/2006	DMT
3MAP002.61	3/28/2006	D5565	3/29/2006	12	B			47.80		3/31/2006	DMT
3MAP002.61	4/19/2006	D5700	4/20/2006	84				79.60		4/26/2006	DMT
3MAP002.61	5/9/2006	D5773	5/10/2006	1480				86.60		5/15/2006	DMT
3MAP002.61	6/21/2006	D6024	6/22/2006	114				92.70		6/26/2006	MAF

A: Value reported is the mean of two or more determinations.

B: Results based upon colony counts outside the acceptable range

U: Material was analyzed for, but not detected. Value stored is the limit of detection for the process in use.

Originally, fecal coliform enumerations were to be done; however, as of October 4, 2005, the client requested that fecal coliform enumerations no longer be analyzed and reported.

Table 5.71 Bacterial Source Tracking for Neabsco Creek at Station 1ANEA002.89.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
1ANEA002.89	7/20/05	D4578	A25	24	96	63%	0%	4%	33%
1ANEA002.89	8/24/05	D4706	A25	9	48	22%	0%	11%	67%
1ANEA002.89	9/27/05	D4825	A25	23	96	39%	9%	17%	35%
1ANEA002.89	10/26/05	D4921	A25	24	254	42%	4%	42%	12%
1ANEA002.89	11/29/05	D5013	A25	24	36	55%	33%	12%	0%
1ANEA002.89	12/21/05	D5103	A25	24	80	17%	25%	50%	8%
1ANEA002.89	1/24/06	D5245	A25	24	92	29%	0%	38%	33%
1ANEA002.89	2/21/06	D5381	A25	1	4	0%	0%	100%	0%
1ANEA002.89	3/28/06	D5568	A25	5	10	60%	0%	40%	0%
1ANEA002.89	4/19/06	D5704	A25	22	64	36%	5%	45%	14%
1ANEA002.89	5/9/06	D5776	A25	23	186	44%	9%	30%	17%
1ANEA002.89	6/21/06	D6026	A25	24	320	33%	4%	55%	8%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.72 Bacterial Source Tracking for Marsh Run at Station 3MAH000.19.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3MAH000.19	7/6/05	D4523	E08	24	72	12%	17%	38%	33%
3MAH000.19	8/17/05	D4659	E08	24	50	21%	4%	4%	71%
3MAH000.19	9/14/05	D4769	E08	10	12	10%	0%	0%	90%
3MAH000.19	10/18/05	D4877	E08	9	22	22%	0%	11%	67%
3MAH000.19	12/12/05	D5041	E08	24	14	25%	0%	25%	50%
3MAH000.19	1/9/06	D5137	E08	24	287	8%	8%	21%	63%
3MAH000.19	2/7/06	D5305	E08	24	157	8%	0%	0%	92%
3MAH000.19	3/14/06	D5496	E08	24	78	17%	25%	17%	41%
3MAH000.19	4/11/06	D5644	E08	24	56	33%	12%	4%	51%
3MAH000.19	5/2/06	D5756	E08	13	98	39%	15%	31%	15%
3MAH000.19	6/13/06	D5961	E08	24	74	38%	0%	8%	54%
3MAH000.19	7/6/05	D4523	E08	24	56	12%	17%	38%	33%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.73 Bacterial Source Tracking for Browns Run at Station 3BOS000.72.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3BOS000.72	7/6/05	D4520	E08	24	82	42%	4%	25%	29%
3BOS000.72	8/17/05	D4657	E08	24	125	17%	0%	29%	54%
3BOS000.72	10/18/05	D4874	E08	24	308	25%	0%	17%	58%
3-BOS000.72	11/15/05	D4977	E08	24	44	21%	4%	33%	42%
3BOS000.72	12/12/05	D5043	E08	24	348	25%	0%	4%	71%
3BOS000.72	1/9/06	D5138	E08	24	74	25%	12%	29%	34%
3BOS000.72	2/7/06	D5306	E08	24	80	12%	0%	12%	76%
3BOS000.72	3/14/06	D5497	E08	24	132	1%	33%	33%	33%
3BOS000.72	4/11/06	D5645	E08	24	104	42%	4%	4%	50%
3BOS000.72	5/2/06	D5757	E08	24	330	29%	0%	50%	21%
3BOS000.72	6/13/06	D5962	E08	24	700	4%	17%	17%	62%
3BOS000.72	7/25/06	D6165	E08	12	20	0%	0%	33%	67%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.74 Bacterial Source Tracking for Craig Run at Station 3CRA000.46.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3CRA000.46	7/6/05	D4521	E08	24	243	25%	0%	63%	12%
3CRA000.46	10/18/05	D4875	E08	24	54	8%	4%	8%	80%
3-CRA000.46	11/15/05	D4978	E08	7	20	29%	0%	57%	14%
3CRA000.46	12/12/05	D5039	E08	24	630	21%	12%	38%	29%
3CRA000.46	1/9/06	D5139	E08	24	76	12%	8%	4%	76%
3CRA000.46	2/7/06	D5307	E08	23	48	4%	0%	53%	43%
3CRA000.46	3/14/06	D5498	E08	24	68	4%	4%	54%	38%
3CRA000.46	4/11/06	D5646	E08	18	26	44%	0%	28%	28%
3CRA000.46	5/2/06	D5758	E08	7	24	29%	14%	0%	57%
3CRA000.46	6/13/06	D5963	E08	24	510	12%	25%	46%	17%
3CRA000.46	7/25/06	D6166	E08	24	310	25%	0%	12%	63%
3CRA000.46	10/3/06	D6451	E08	16	34	44%	6%	6%	44%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.75 Bacterial Source Tracking for Rappahannock River at Station 3RPP147.10.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3RPP147.10	7/6/05	D4522	E08	24	220	4%	4%	42%	50%
3RPP147.10	8/17/05	D4660	E08	24	84	38%	0%	8%	54%
3RPP147.10	9/14/05	D4770	E08	24	100	12%	0%	0%	88%
3RPP147.10	10/18/05	D4878	E08	24	74	8%	4%	25%	63%
3-RPP147.10	11/15/05	D4979	E08	24	60	4%	46%	33%	17%
3RPP147.10	12/12/05	D5040	E08	24	163	21%	4%	17%	58%
3RPP147.10	1/9/06	D5140	E08	24	60	8%	8%	38%	46%
3RPP147.10	2/7/06	D5308	E08	12	30	33%	0%	25%	42%
3RPP147.10	3/14/06	D5499	E08	24	56	12%	12%	38%	38%
3RPP147.10	4/11/06	D5647	E08	24	42	17%	25%	12%	46%
3RPP147.10	5/2/06	D5759	E08	23	94	13%	30%	13%	44%
3RPP147.10	6/13/06	D5964	E08	24	42	0%	0%	100%	0%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.76 Bacterial Source Tracking for Cedar Run at Station 3CED000.59.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3CED000.59	7/12/05	D4533	E16	24	60	0%	12%	59%	29%
3CED000.59	8/3/05	D4641	E16	24	36	21%	0%	8%	71%
3CED000.59	9/7/05	D4744	E16	15	18	7%	0%	0%	93%
3CED000.59	10/19/05	D4900	E16	13	34	0%	23%	15%	62%
3CED000.59	11/16/05	D4995	E16	21	48	33%	38%	19%	10%
3CED000.59	12/19/05	D5100	E16	24	210	21%	4%	29%	46%
3CED000.59	1/10/06	D5147	E16	16	32	12%	38%	6%	44%
3CED000.59	2/14/06	D5354	E16	13	28	31%	0%	31%	38%
3CED000.59	3/22/06	D5536	E16	7	8	29%	0%	14%	57%
3CED000.59	4/26/06	D5733	E16	24	134	0%	0%	8%	92%
3CED000.59	5/31/06	D5894	E16	8	22	12%	12%	0%	76%
3CED000.59	7/11/06	D6081	E16	24	88	45%	17%	17%	21%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.77 Bacterial Source Tracking for Rapidan River at Station 3RAP006.53.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3RAP006.53	7/12/05	D4532	E18	24	96	0%	0%	54%	46%
3RAP006.53	8/3/05	D4640	E18	24	30	21%	0%	21%	58%
3RAP006.53	9/7/05	D4743	E18	21	28	24%	0%	33%	43%
3RAP006.53	10/19/05	D4899	E18	24	56	0%	12%	0%	88%
3RAP006.53	11/16/05	D4994	E18	23	60	17%	31%	26%	26%
3RAP006.53	12/19/05	D5101	E18	24	223	17%	0%	62%	21%
3RAP006.53	1/10/06	D5146	E18	24	140	4%	33%	51%	12%
3RAP006.53	2/14/06	D5353	E18	24	60	54%	0%	21%	25%
3RAP006.53	3/22/06	D5535	E18	6	6	17%	0%	0%	83%
3RAP006.53	4/26/06	D5732	E18	20	310	20%	10%	5%	65%
3RAP006.53	5/31/06	D5893	E18	5	24	20%	0%	0%	80%
3RAP006.53	7/11/06	D6080	E18	24	58	21%	4%	25%	50%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.78 Bacterial Source Tracking for Rappahannock River at Station 3RPP110.57.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3RPP110.57	7/20/05	D4574	E20	24	92	96%	0%	4%	0%
3RPP110.57	8/24/05	D4702	E20	16	56	69%	19%	0%	12%
3RPP110.57	9/27/05	D4821	E20	24	68	0%	8%	8%	84%
3RPP110.57	10/26/05	D4920	E20	24	328	25%	42%	4%	29%
3RPP110.57	11/29/05	D5012	E20	24	62	76%	12%	0%	12%
3RPP110.57	12/21/05	D5104	E20	24	60	55%	12%	4%	29%
3RPP110.57	1/24/06	D5244	E20	24	356	25%	21%	8%	46%
3RPP110.57	2/21/06	D5380	E20	2	4	0%	100%	0%	0%
3RPP110.57	3/28/06	D5567	E20	2	8	0%	0%	100%	0%
3RPP110.57	4/19/06	D5703	E20	24	116	21%	21%	41%	17%
3RPP110.57	5/9/06	D5775	E20	10	22	0%	30%	60%	10%
3RPP110.57	6/21/06	D6022	E20	16	82	6%	12%	82%	0%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.79 Bacterial Source Tracking for Rappahannock River at Station 3RPP104.47.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3RPP104.47	7/7/05	D4526		24	42	72%	12%	8%	8%
3RPP104.47	9/6/05	D4740		8	22	76%	0%	12%	12%
3RPP104.47	10/18/05	D4879		24	256	0%	4%	8%	88%
3RPP104.47	11/1/05	D4937		13	28	15%	15%	55%	15%
3RPP104.47	12/12/05	D5045		24	388	17%	21%	25%	37%
3RPP104.47	2/8/06	D5314		20	46	20%	35%	10%	35%
3RPP104.47	3/8/06	D5452		3	6	0%	67%	33%	0%
3RPP104.47	4/5/06	D5595		4	6	25%	0%	50%	25%
3RPP104.47	5/3/06	D5763		24	70	26%	33%	8%	33%
3RPP104.47	6/20/06	D5978		2	2	0%	50%	0%	50%
3RPP104.47	8/9/06	D6230		1	2	100%	0%	0%	0%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.80 Bacterial Source Tracking for Rappahannock River at Station 3RPP080.19.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3RPP080.19	7/7/05	D4524	E21	5	6	20%	40%	20%	20%
3RPP080.19	8/9/05	D4642	E21	10	22	10%	0%	50%	40%
3RPP080.19	9/6/05	D4741	E21	5	16	80%	0%	0%	20%
3RPP080.19	10/18/05	D4873	E21	24	180	4%	0%	38%	58%
3RPP080.19	11/1/05	D4935	E21	24	66	29%	0%	21%	50%
3RPP080.19	12/12/05	D5042	E21	24	108	17%	25%	21%	37%
3RPP080.19	1/4/06	D5122	E21	24	76	42%	17%	12%	29%
3RPP080.19	2/8/06	D5312	E21	24	136	8%	71%	4%	17%
3RPP080.19	3/8/06	D5450	E21	1	2	0%	100%	0%	0%
3RPP080.19	4/5/06	D5593	E21	6	8	50%	0%	50%	0%
3RPP080.19	5/3/06	D5761	E21	12	24	0%	67%	25%	8%
3RPP080.19	6/20/06	D5976	E21	3	6	0%	67%	33%	0%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.81 Bacterial Source Tracking for Rappahannock River at Station 3RPP091.55.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3RPP091.55	7/7/2005	D4525	E21	7	10	43%	14%	29%	14%
3RPP091.55	8/9/2005	D4643	E21	6	12	0%	17%	66%	17%
3RPP091.55	9/6/2005	D4742	E21	13	28	54%	0%	31%	15%
3RPP091.55	10/18/2005	D4876	E21	24	74	4%	8%	12%	76%
3RPP091.55	11/1/2005	D4936	E21	24	114	46%	8%	17%	29%
3RPP091.55	12/12/2005	D5044	E21	24	246	8%	17%	33%	42%
3RPP091.55	1/4/2006	D5123	E21	23	141	26%	22%	30%	22%
3RPP091.55	2/8/2006	D5313	E21	24	210	0%	45%	17%	38%
3RPP091.55	3/8/2006	D5451	E21	*NVI	1	*NVI	*NVI	*NVI	*NVI
3RPP091.55	4/5/2006	D5594	E21	6	12	67%	0%	33%	0%
3RPP091.55	5/3/2006	D5762	E21	9	20	0%	67%	0%	33%
3RPP091.55	6/20/2006	D5977	E21	4	20	0%	50%	50%	0%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.82 Bacterial Source Tracking for Claiborne Run at Station 3CLB000.50.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3CLB000.50	7/20/05	D4575	E20	24	279	76%	12%	12%	0%
3CLB000.50	8/24/05	D4703	E20	24	114	83%	0%	17%	0%
3CLB000.50	9/27/05	D4822	E20	24	142	0%	4%	17%	79%
3CLB000.50	10/26/05	D4919	E20	24	300	4%	42%	0%	54%
3CLB000.50	11/29/05	D5011	E20	24	90	71%	4%	17%	8%
3CLB000.50	1/24/06	D5243	E20	24	66	21%	4%	67%	8%
3CLB000.50	2/21/06	D5379	E20	5	12	20%	80%	0%	0%
3CLB000.50	3/28/06	D5566	E20	19	34	52%	26%	11%	11%
3CLB000.50	4/19/06	D5702	E20	24	94	29%	17%	17%	37%
3CLB000.50	5/9/06	D5774	E20	23	128	22%	22%	22%	34%
3CLB000.50	6/21/06	D6023	E20	24	400	8%	0%	84%	8%
3CLB000.50	7/25/06	D6164	E20	24	250	33%	0%	67%	0%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.83 Bacterial Source Tracking for Hazel Run at Station 3HAL000.57.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3HAL000.57	7/20/05	D4577	E20	24	940	83%	0%	17%	0%
3HAL000.57	8/24/05	D4705	E20	24	242	54%	0%	46%	0%
3HAL000.57	9/27/05	D4824	E20	24	212	0%	0%	4%	96%
3HAL000.57	10/26/05	D4917	E20	24	219	0%	12%	59%	29%
3HAL000.57	11/29/05	D5009	E20	24	54	72%	4%	12%	12%
3HAL000.57	12/21/05	D5105	E20	15	32	67%	0%	20%	13%
3HAL000.57	1/24/06	D5241	E20	24	64	59%	4%	25%	12%
3HAL000.57	2/21/06	D5378	E20	3	6	33%	67%	0%	0%
3HAL000.57	3/28/06	D5564	E20	17	32	18%	18%	64%	0%
3HAL000.57	4/19/06	D5701	E20	24	199	42%	21%	25%	12%
3HAL000.57	5/9/06	D5772	E20	23	230	9%	9%	43%	39%
3HAL000.57	6/21/06	D6025	E20	24	310	8%	8%	76%	8%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

Table 5.84 Bacterial Source Tracking for Massaponax Creek at Station 3MAP002.61.

VADEQ ID	Date of Sample	Lab ID	HUP ID	Number of Isolates	<i>E. coli</i> (cfu/100 ml)	Wildlife	Human	Livestock	Pet
3MAP002.61	7/20/05	D4576	E20	24		75%	21%	4%	0%
3MAP002.61	8/24/05	D4704	E20	21		76%	0%	19%	5%
3MAP002.61	9/27/05	D4823	E20	11		0%	0%	9%	91%
3MAP002.61	10/26/05	D4918	E20	24		0%	12%	63%	25%
3MAP002.61	11/29/05	D5010	E20	4		50%	25%	25%	0%
3MAP002.61	12/21/05	D5106	E20	3		67%	0%	33%	0%
3MAP002.61	1/24/06	D5242	E20	24		21%	4%	63%	12%
3MAP002.61	2/21/06	D5377	E20	2		0%	100%	0%	0%
3MAP002.61	3/28/06	D5565	E20	6		33%	0%	67%	0%
3MAP002.61	4/19/06	D5700	E20	24		80%	8%	12%	0%
3MAP002.61	5/9/06	D5773	E20	22		9%	18%	32%	41%
3MAP002.61	6/21/06	D6024	E20	24		0%	0%	92%	8%

BOLD type indicates a statistically significant value.

*NVI - No Viable isolates

6. DISCUSSION

Results of the 2005-2006 VADEQ BST program have been presented in this report. The ARCCs achieved during the library development stage are acceptable and there does not appear to be a high level of over-fitting. Based on the sample size targeted in each sample (*i.e.*, 24 isolates), there is 90% confidence that the proportions measured in each sample are within 15% of the actual proportions in the sampled population (*i.e.*, all bacteria in the stream at the time of sampling). Because a fixed-frequency sampling scheme was used, samples are not biased toward a particular flow regime and can therefore be combined to estimate the actual proportions contributed by the different sources over the entire year with greater precision (*i.e.*, 90% confidence that the estimate is within 5% of the actual proportions). Additionally, the statistical analyses applied to determine a significant difference from zero give a good indication of presence and absence of each source in each sample. All of these data are valuable for use in improving public awareness of the problem, improving model calibration/validation, and providing a more equitable allocation of loads to source classes.

Since the presence of optical brighteners does not always coincide with the presence of human fecal bacteria. These two indicators should be interpreted together. The consistent presence of optical brighteners with little or no indication of human fecal bacteria indicates likely gray-water discharge(s). The consistent presence of human fecal bacterial without the presence of high optical brightener concentrations may indicate failing septic systems-since optical brighteners photo degrade-or straight pipes in an area where washing machines are not typically owned or operated during the typical sample times. The presence of both indicators typically signifies a leaking/overflowing sewer system or a high density of failing on-site systems where the failure is directly discharged to the stream without the need of a washoff event.

In spite of the high quality of the data collected, care should be taken in using these data. These data represent, at most, 12 instantaneous observations at each station and may not be representative of long-term conditions. The hydrologic conditions during this period may not reflect either average or critical conditions. Additionally, the dynamics of the bacterial community are not well understood, so care should be taken in extrapolating from the in-stream condition to activities in the watershed. As with any other monitoring program, the data should

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not be viewed in a vacuum. Local knowledge of the sources involved, historical water quality records, and the hydrologic conditions during sampling should all be considered in any interpretation of this data.

REFERENCES

- Hagedorn, C., S. L. Robinson, J. R. Filtz, S. M. Grubbs, T. A. Angier, and R. B. Reneau, Jr. 1999. Using antibiotic resistance patterns in the fecal streptococci to determine sources of fecal pollution in a rural Virginia watershed. *Appl. Environ. Microbiol.* 65:5522-5531.
- USEPA. 1999. Guidance for Water Quality-Based Decisions: The TMDL Process. <http://www.epa.gov/OWOW/tmdl/decisions/dec1c.html>

APPENDIX A

Bacterial Source Tracking Analyses Supplemental Report

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Table A.1 False-positive and correct classification rates for twelve BST libraries developed in support of VADEQ's Phase-IV BST Program.

Library	False-Positive Rates				Rate of Correct Classification			
	Wildlife	Human	Livestock	Pet	Wildlife	Human	Livestock	Pet
02080103	4.5%	6.5%	13.6%	7.2%	69.5%	84.5%	84.1%	66.7%
02070010	11.9%	5.9%	11.9%	7.1%	69.9%	83.9%	65.2%	70.5%
02080104	18.2%	5.0%	9.6%	7.7%	77.6%	78.9%	46.7%	75.0%
02080203	9.1%	5.3%	12.6%	5.8%	67.9%	74.6%	80.0%	78.9%
02080205	9.9%	5.5%	11.0%	9.2%	68.7%	83.8%	70.0%	70.4%
02080206	12.7%	6.8%	11.1%	8.9%	67.0%	82.8%	61.1%	70.6%
02080202	12.2%	10.7%	13.2%	7.3%	67.8%	73.0%	64.4%	64.7%
03010105	8.8%	12.4%	10.2%	13.2%	64.5%	72.7%	59.3%	68.8%
06010205	7.9%	9.0%	9.3%	8.5%	73.3%	85.5%	70.4%	66.8%
05050001	5.8%	3.3%	7.7%	6.7%	80.0%	78.8%	83.9%	86.9%

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Table A.2 Species sampled for 12 libraries developed in support of VADEQ's Phase-IV BST Program.

Source Category	Species	02080103	02070010	02080104	02080203	0280205	02080206	02080202	03010105	06010205	05050001
Human	Human	x	x	x	x	x	x	x	x	x	x
Livestock	Beef	x	x	x	x	x	x	x	x	x	x
	Dairy				x	x	x	x	x		x
	Mule										x
	Goat					x	x		x	x	x
	Horse	x	x	x	x	x	x	x		x	x
	Llama									x	
	Poultry	x			x	x	x	x			
	Sheep	x					x				x
	Swine	x			x	x	x	x	x		
Pet	Cat	x	x		x	x	x	x	x	x	x
	Dog	x	x	x	x	x	x	x	x	x	x
Wildlife	Bear				x	x	x	x	x		
	Bobcat			x							
	Coyote										x
	Deer	x	x	x	x	x	x	x	x	x	x
	Duck				x						x
	Fox	x			x	x	x	x	x	x	
	Goose		x	x	x	x	x	x	x		x
	Groundhog										x
	Muskrat				x		x	x	x		
	Opossum	x	x	x					x	x	
	Otter				x	x	x	x	x		
	Pigeon									x	
	Quail					x					
	Rabbit	x		x						x	x
	Raccoon	x	x	x	x	x	x	x	x	x	x
	Sea Gull										
Skunk				x	x	x	x	x			
Squirrel				x	x	x	x	x			
Wild Turkey							x		x		