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**Total Maximum Daily Loads of Fecal Bacteria  
for the Non-Tidal Dividing Creek Basin  
in Somerset, Wicomico, and Worcester Counties, Maryland**

**FINAL**



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## List of Abbreviations

ARCC	Average rates of correct classification
ARA	Antibiotic Resistance Analysis
BMP	Best Management Practice
BST	Bacteria Source Tracking
cfs	Cubic Feet per Second
CFR	Code of Federal Regulations
CFU	Colony Forming Units
COMAR	Code of Maryland Regulations
CWA	Clean Water Act
DNR	Maryland Department of Natural Resources
EPA	United States Environmental Protection Agency
GIS	Geographic Information System
LA	Load Allocation
MACS	Maryland Agricultural Cost Share Program
MDE	Maryland Department of the Environment
MDP	Maryland Department of Planning
MGD	Millions of Gallons per Day
ml	Milliliter(s)
MOS	Margin of Safety
MPN	Most Probable Number
MPR	Maximum Practicable Reduction
MS4	Municipal Separate Storm Sewer System
MST	Microbial Source Tracking
NPDES	National Pollutant Discharge Elimination System
RCC	Rates of Correct Classification
TMDL	Total Maximum Daily Load
USGS	United States Geological Survey
WQIA	Water Quality Improvement Act
WLA	Wasteload Allocation
WQLS	Water Quality Limited Segment
WWTP	Wastewater Treatment Plant

## EXECUTIVE SUMMARY

This document, upon approval by the U.S. Environmental Protection Agency (EPA), establishes a Total Maximum Daily Load (TMDL) for fecal bacteria in the non-tidal portion of the Dividing Creek watershed (basin number 02-13-02-04). Section 303(d)(1)(C) of the federal Clean Water Act (CWA) and the U.S. Environmental Protection Agency's (EPA) implementing regulations direct each state to identify and list waters, known as water quality limited segments (WQLSs), in which current required controls of a specified substance are inadequate to achieve water quality standards. For each WQLS, states are required to either establish a Total Maximum Daily Load (TMDL) of the specified substance that the waterbody can receive without violating water quality standards or demonstrate that water quality standards are being met.

The Maryland Department of the Environment (MDE) has identified the non-tidal portion of Dividing Creek and its tributaries in the State of Maryland's 303(d) List as impaired by fecal bacteria (listed in 1996) and by impacts to biological communities (listed in 2004 and 2006). The waterbodies of the Dividing Creek watershed have been designated as Use I - Water Contact Recreation, and Protection of Non-tidal Warmwater Aquatic Life (see Code of Maryland Regulations (COMAR) 26.08.02.07F(5)). This document proposes to establish a TMDL for fecal bacteria in the non-tidal Dividing Creek watershed that will allow for attainment of the beneficial use designation of primary water contact recreation. The listing for impacts to biological communities will be addressed separately at a future date. MDE monitored the non-tidal portion of the Dividing Creek watershed from 2003-2004 for fecal bacteria. A data solicitation for fecal bacteria was conducted by MDE in 2003, and all readily available data from the past five years were considered.

For this TMDL analysis, the non-tidal Dividing Creek basin has been divided into two subwatersheds, which include Dividing Creek and Pusey Branch. The pollutant loads set forth in this document are for these two subwatersheds. To establish baseline and allowable pollutant loads for this TMDL, a flow duration curve approach was employed, using bacteria data from MDE and flow strata estimated from United States Geological Survey (USGS) daily flow monitoring. The sources of fecal bacteria are estimated at two representative stations in the non-tidal Dividing Creek basin where samples were collected for one year. Multiple antibiotic resistance analysis (ARA) source tracking was used to determine the relative proportion of domestic (pets and human associated animals), human (human waste), livestock (agriculture-related animals), and wildlife (mammals and waterfowl) source categories.

The allowable load is determined by estimating a baseline load from current monitoring data. The baseline load is estimated using a long-term geometric mean and weighting factors from the flow duration curve. The TMDL for fecal bacteria is established after considering three different hydrological conditions: high flow and low flow annual conditions; and an average seasonal condition (the period between May 1<sup>st</sup> and September 30<sup>th</sup>, when water contact recreation is more prevalent). This allowable load is reported in units of Most Probable Number (MPN)/year and represents a long-term load estimated over a variety of hydrological conditions.

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Two scenarios were developed, with the first assessing if attainment of current water quality standards could be achieved by applying maximum practicable reductions (MPRs), and the second applying higher reductions than MPRs. Scenario solutions were based on an optimization method where the objective was to minimize the overall risk to human health, assuming that the risk varies over the four bacteria source categories. In both subwatersheds, it was estimated that water quality standards could not be attained with MPRs, thus higher maximum reductions were applied.

The fecal bacteria long-term annual average TMDL for the non-tidal Dividing Creek basin is 25,351 billion MPN *E. coli*/year, with a maximum daily load of 505 billion MPN/day. These total loads represent the sum of individual TMDLs for the two subwatersheds. The long-term annual average TMDL represents a reduction of approximately 65.6 % from the baseline load of 73,630 billion MPN/year. The TMDL is distributed between a load allocation (LA) for nonpoint sources and waste load allocations (WLAs) for point sources. Point sources include National Pollutant Discharge Elimination System (NPDES) wastewater treatment plants (WWTPs) and NPDES regulated stormwater discharges, including municipal separate storm sewer systems (MS4s). No such point sources exist in the non-tidal Dividing Creek basin; thus, the entire TMDL is assigned to the LA. The maximum daily load, estimated using predicted long-term annual average TMDL concentrations, is also assigned entirely to the LA.

The margin of safety (MOS) has been incorporated using a conservative assumption by estimating the loading capacity of the stream based on a water quality endpoint concentration more stringent than the applicable Maryland (MD) water quality standard criterion. The *E. coli* water quality criterion concentration was reduced by 5%, from 126 MPN/100ml to 119.7 MPN/100ml.

Once EPA has approved a TMDL, and it is known what measures must be taken to reduce pollution levels, implementation of best management practices (BMPs) is expected to take place. MDE intends for the required reductions to be implemented in an iterative process that first addresses those sources with the largest impacts to water quality and creating the greatest risks to human health, with consideration given to ease and cost of implementation. In addition, follow-up monitoring plans will be established to track progress and to assess the implementation efforts. As previously stated, water quality standards cannot be attained in either of the two subwatersheds using the MPR scenario. MPRs may not be sufficient in subwatersheds where wildlife is a significant component or where very high reductions of fecal bacteria loads are required to meet water quality standards. In these cases, it is expected that the MPR scenario will be the first stage of TMDL implementation. Progress will be made through the iterative implementation process described above, and the situation will be reevaluated in the future.



## 1.0 INTRODUCTION

This document, upon approval by the U.S. Environmental Protection Agency (EPA), establishes a Total Maximum Daily Load (TMDL) for fecal bacteria in the non-tidal portion of the Dividing Creek watershed (basin number 02-13-02-04). Section 303(d)(1)(C) of the federal Clean Water Act (CWA) and the U.S. Environmental Protection Agency's (EPA) implementing regulations direct each state to develop a TMDL for each impaired water quality limited segment (WQLS) on the Section 303(d) List, taking into account seasonal variations and a protective margin of safety (MOS) to account for uncertainty. A TMDL reflects the total pollutant loading of the impairing substance a waterbody can receive and still meet water quality standards.

TMDLs are established to achieve and maintain water quality standards. A water quality standard is the combination of a designated use for a particular body of water and the water quality criteria designed to protect that use. Designated uses include activities such as swimming, drinking water supply, and shellfish propagation and harvest. Water quality criteria consist of narrative statements and numeric values designed to protect the designated uses. Criteria may differ among waters with different designated uses.

The non-tidal portion of Dividing Creek and its tributaries have been identified in the State of Maryland's 303(d) List as impaired by fecal bacteria (listed in 1996) and by impacts to biological communities (listed in 2004 and 2006). The waterbodies of the Dividing Creek watershed have been designated as Use I - Water Contact Recreation, and Protection of Non-tidal Warmwater Aquatic Life (see Code of Maryland Regulations (COMAR) 26.08.02.07F(5)). This document proposes to establish a TMDL for fecal bacteria in the non-tidal Dividing Creek watershed that will allow for attainment of the beneficial use designation of primary water contact recreation. The listing for impacts to biological communities will be addressed separately at a future date. MDE monitored the non-tidal portion of the Dividing Creek watershed from 2003-2004 for fecal bacteria. A data solicitation for fecal bacteria was conducted by MDE in 2003, and all readily available data from the past five years were considered.

Fecal bacteria are microscopic single-celled organisms (primarily fecal coliform and fecal streptococci) found in the wastes of warm-blooded animals. Their presence in water is used to assess the sanitary quality of water for body-contact recreation, for consumption of molluscan bivalves (shellfish), and for drinking water. Excessive amounts of fecal bacteria in surface water used for recreation are known to indicate an increased risk of pathogen-induced illness to humans. Infections due to pathogen-contaminated recreation waters include gastrointestinal, respiratory, eye, ear, nose, throat, and skin diseases (US EPA 1986).

In 1986, EPA published "Ambient Water Quality Criteria for Bacteria," in which three indicator organisms were assessed to determine their correlation with swimming-associated illnesses. Fecal coliform, *E. coli* and enterococci were the indicators used in the analysis. Fecal coliform bacteria are a subgroup of total coliform bacteria and *E. coli* bacteria are a subgroup of fecal coliform bacteria. Most *E. coli* are harmless and are found in great quantities in the intestines of people and warm-blooded animals. However, certain pathogenic strains may cause illness.

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Enterococci are a subgroup of bacteria in the fecal streptococcus group. Fecal coliform, *E. coli* and enterococci can all be classified as fecal bacteria. The results of the EPA study demonstrated that fecal coliform showed less correlation to swimming-associated gastroenteritis than did either *E. coli* or enterococci.

Based on EPA's guidance (US EPA 1986), adopted by Maryland in 2004, the State has revised the bacteria water quality criteria and it is now based on water column limits for either *E. coli* or enterococci. Because multiple monitoring datasets are available within this watershed for various pathogen indicators, the general term fecal bacteria will be used to refer to the impairing substance throughout this document. The TMDL will be based on the pathogen indicator organisms specified in Maryland's current bacteria water quality criteria, either *E. coli* or enterococci. The indicator organism used in the non-tidal Dividing Creek TMDL analysis was *E. coli*.

## 2.0 SETTING AND WATER QUALITY DESCRIPTION

### 2.1 General Setting

#### Location

The Dividing Creek watershed is located in the Pocomoke River region of Maryland (See Figure 2.1.1). The watershed encompasses 39,699 acres (62.03 square miles) in Somerset, Wicomico and Worcester counties. Of this, the non-tidal portion accounts for 23,150 acres (36.17 square miles) of the northern half of the overall watershed. The headwaters of Dividing Creek, which is one of the major tributaries of the Pocomoke River, begin just above the Wicomico County border. Flowing south it becomes the border for Somerset and Worcester Counties and is joined by Pollitt's Branch. Near the town of Whitesburg, Dividing Creek is joined by its major tributary Pusey Branch, which also begins in Wicomico County.

#### Land Use

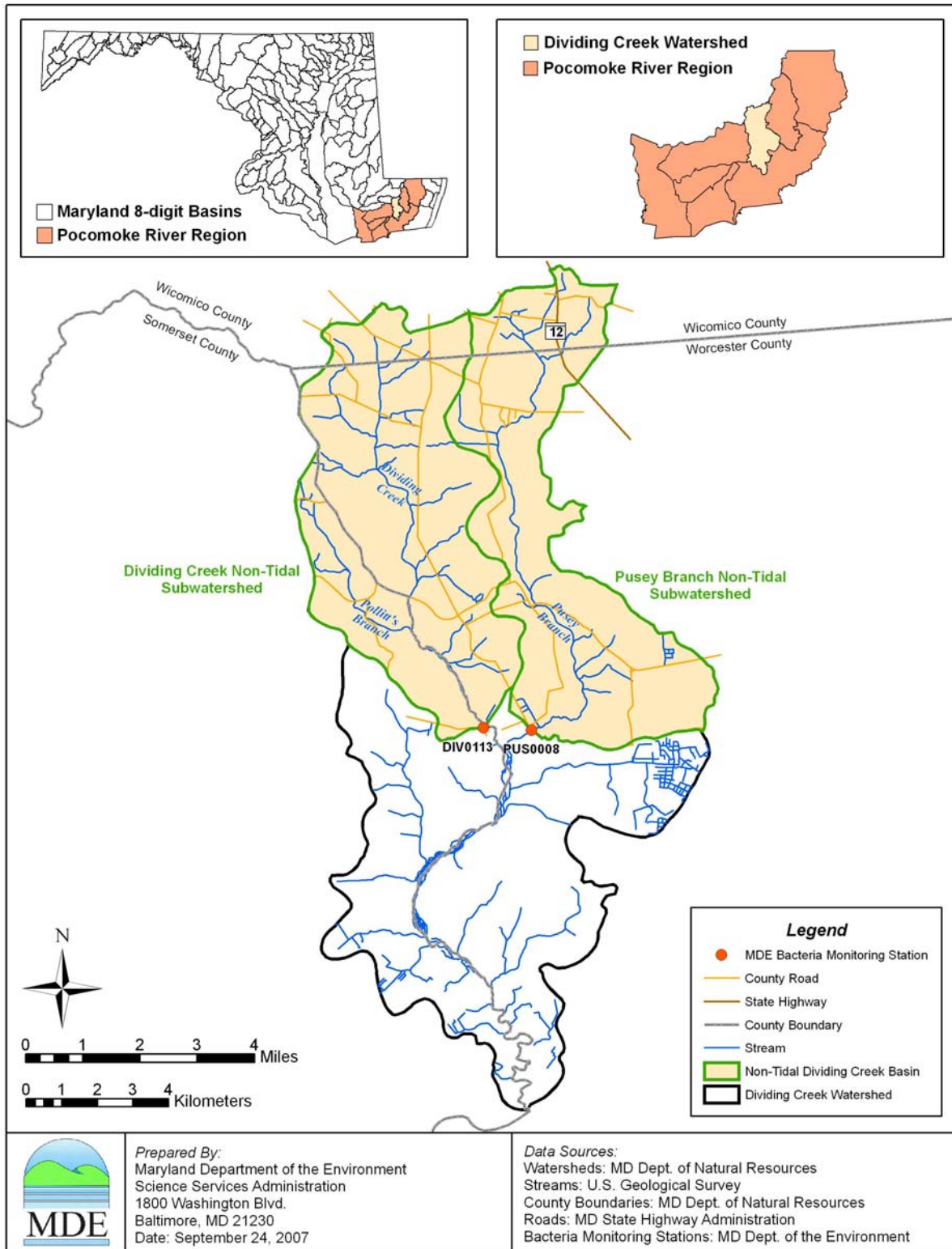
The non-tidal portion of the Dividing Creek watershed covers an area of 23,150 acres. Based on the 2002 Maryland Department of Planning (MDP) land use/land cover data the majority of this region, approximately 80%, is forested land. There is also occasional agricultural use and very little residential presence. The land use percentage distribution is shown in Table 2.1.1, and spatial distributions for each land use are shown in Figure 2.1.2. Table 2.1.2 shows the land use percentage distribution for each subwatershed considered in the analysis.

**Table 2.1.1: Land Use Distribution for the Non-Tidal Dividing Creek Basin**

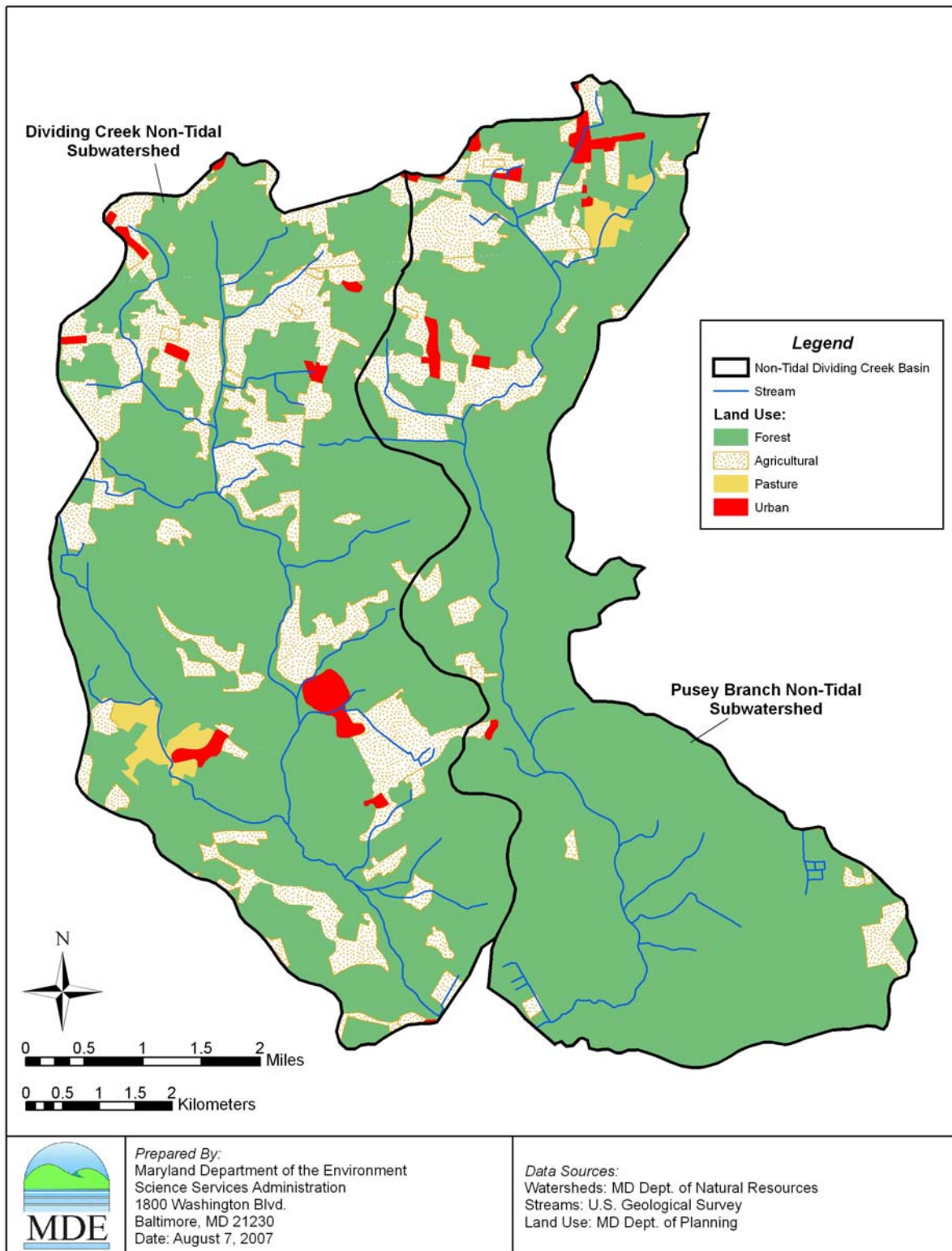
Land Type	Area (acres)	Percentage (%)
Forest	18,465	79.8
Agricultural	4,048	17.5
Urban	373	1.6
Pasture	264	1.1
<b>Total</b>	<b>23,150</b>	<b>100</b>

**Table 2.1.2: Land Use Distribution per Subwatershed in the Non-Tidal Dividing Creek Basin**

<b>Land Type</b>	<b>Dividing Creek / DIV0113</b>		<b>Pusey Branch / PUS0008</b>	
	<b>Area (acres)</b>	<b>Percentage (%)</b>	<b>Area (acres)</b>	<b>Percentage (%)</b>
Forest	9,372	75.0	9,093	85.3
Agricultural	2,729	21.8	1,319	12.4
Urban	223	1.8	150	1.4
Pasture	171	1.4	93	0.9
<b><i>Total</i></b>	<b>12,495</b>	<b>100</b>	<b>10,655</b>	<b>100</b>



**Figure 2.1.1: Location Map of the Non-Tidal Area in the Dividing Creek Watershed**



**Figure 2.1.2: Land Use of the Non-Tidal Dividing Creek Basin**

### **Population**

The total population in the non-tidal portion of the Dividing Creek watershed is estimated to be 690 people. Figure 2.1.3 depicts the population density in the region. The human population and the number of dwellings were estimated based on a weighted average from the 2000 Census GIS Block Groups (U.S. Department of Commerce 2000) and the 2002 MDP Land Use Land Cover. Since the boundaries of the watershed differ from the boundaries of the block groups, residential land use data were used to extract the necessary areas of the Census block groups. The MDP residential density designations shown in Table 2.1.3 were used for this estimation.

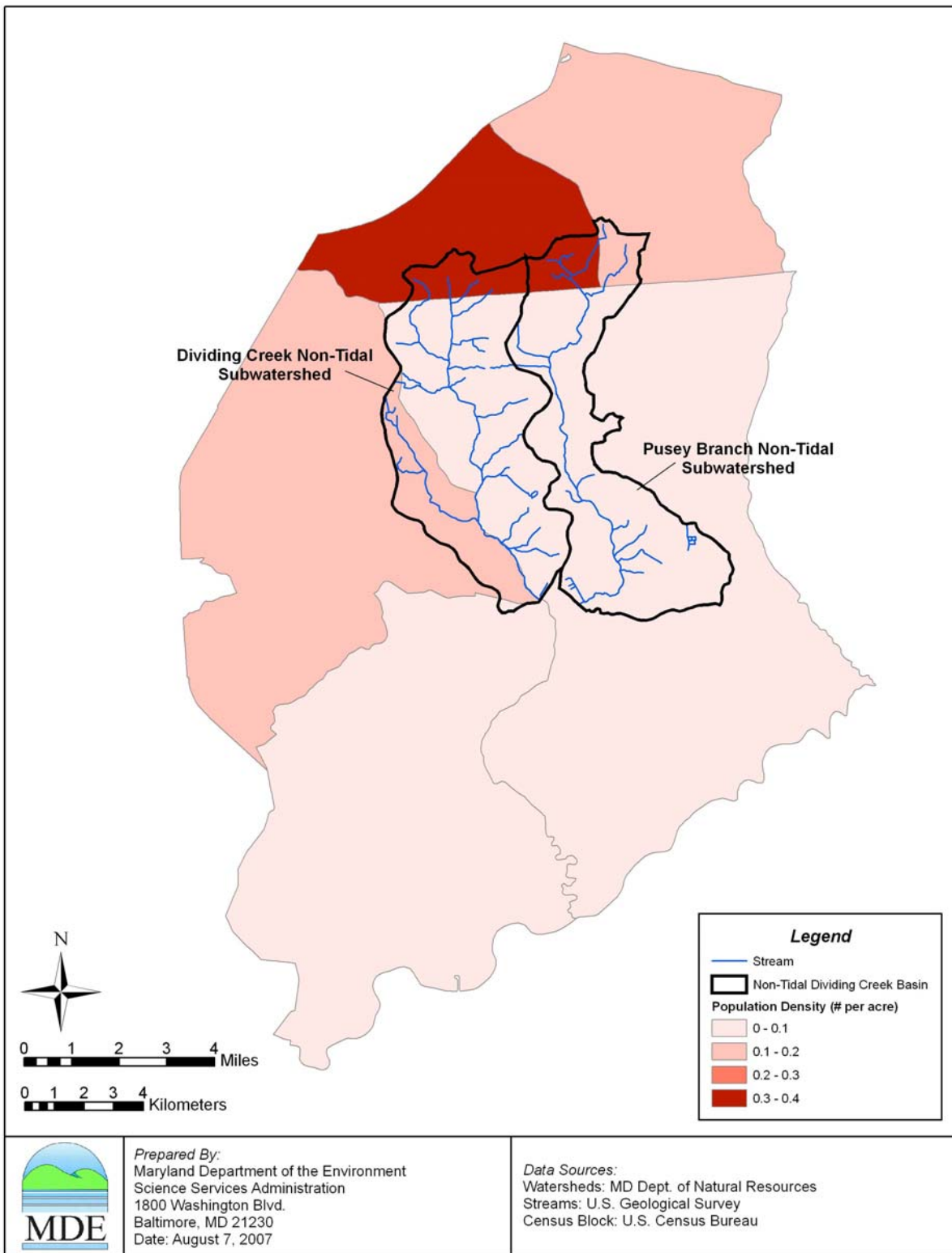
**Table 2.1.3: Number of Dwellings Per Acre**

<b>Land Use Code</b>	<b>Dwellings Per Acre</b>
11 Low Density Residential	1
12 Medium Density Residential	5
13 High Density Residential	8

Based on these densities, the number of dwellings in each census block group was determined. From this a ratio of people per dwelling was calculated for each block group. The residential density was again used to find the number of dwellings for the portion of each block group within the watershed. Multiplying this dwelling count by the people per dwelling ratio for the block group gives the population in the portion of the block group within the watershed. Summing the population for each block group gives the results presented in Table 2.1.4, which shows the total population for the two subwatersheds in the non-tidal Dividing Creek basin.

**Table 2.1.4: Total Population Per Subwatershed in the Dividing Creek Watershed**

<b>Tributary</b>	<b>Station</b>	<b>Population</b>
Dividing Creek	DIV0113	425
Pusey Branch	PUS0008	265
<b><i>Total</i></b>		<b>690</b>



**Figure 2.1.3: Population Density in the Non-Tidal Dividing Creek Basin**



## 2.2 Water Quality Characterization

EPA's guidance document, "Ambient Water Quality Criteria for Bacteria" (1986), recommended that states use *E. coli* (for fresh water) or enterococci (for fresh or salt water) as pathogen indicators. Fecal bacteria, *E. coli*, and enterococci were assessed as indicator organisms for predicting human health impacts. A statistical analysis found that the highest correlation to gastrointestinal illness was linked to elevated levels of *E. coli* and enterococci in fresh water (enterococci in salt water).

As per EPA's guidance, Maryland has adopted the new indicator organisms, *E. coli* and enterococci, for the protection of public health in Use I, II, and IV waters. These bacteria listings were originally assessed using fecal coliform bacteria. The analysis was based on a geometric mean of the monitoring data, where the result had to be less than or equal to 200 MPN/100ml. From EPA's analysis (US EPA 1986), this fecal coliform geometric mean target equates to an approximate risk of 8 illnesses per 1,000 swimmers at fresh water beaches and 19 illnesses per 1,000 swimmers at marine beaches (enterococci only), which is consistent with MDE's revised Use I bacteria criteria. Therefore, the original 303(d) List fecal coliform listings can be addressed using the refined bacteria indicator organisms to ensure that risk levels are acceptable.

### **Bacteria Monitoring**

Table 2.2.1 lists the historical monitoring data available for the non-tidal Dividing Creek basin. MDE conducted bacteria monitoring at two stations in the Dividing Creek watershed from November 2003 through October 2004. USGS gage station 01485500, located near the non-tidal Dividing Creek basin near Snow Hill, MD, was used in deriving the surface flow. The locations of these stations are shown in Tables 2.2.2 and 2.2.3 and in Figure 2.2.1. Observations recorded from the two MDE monitoring stations are provided in Appendix A.

Bacteria counts are highly variable which is typical due to the nature of bacteria and their relationship to flow. The *E. coli* counts for the two stations ranged between 10 and 2,005 MPN/100 ml.

**Table 2.2.1: Historical Monitoring Data in the Dividing Creek Watershed**

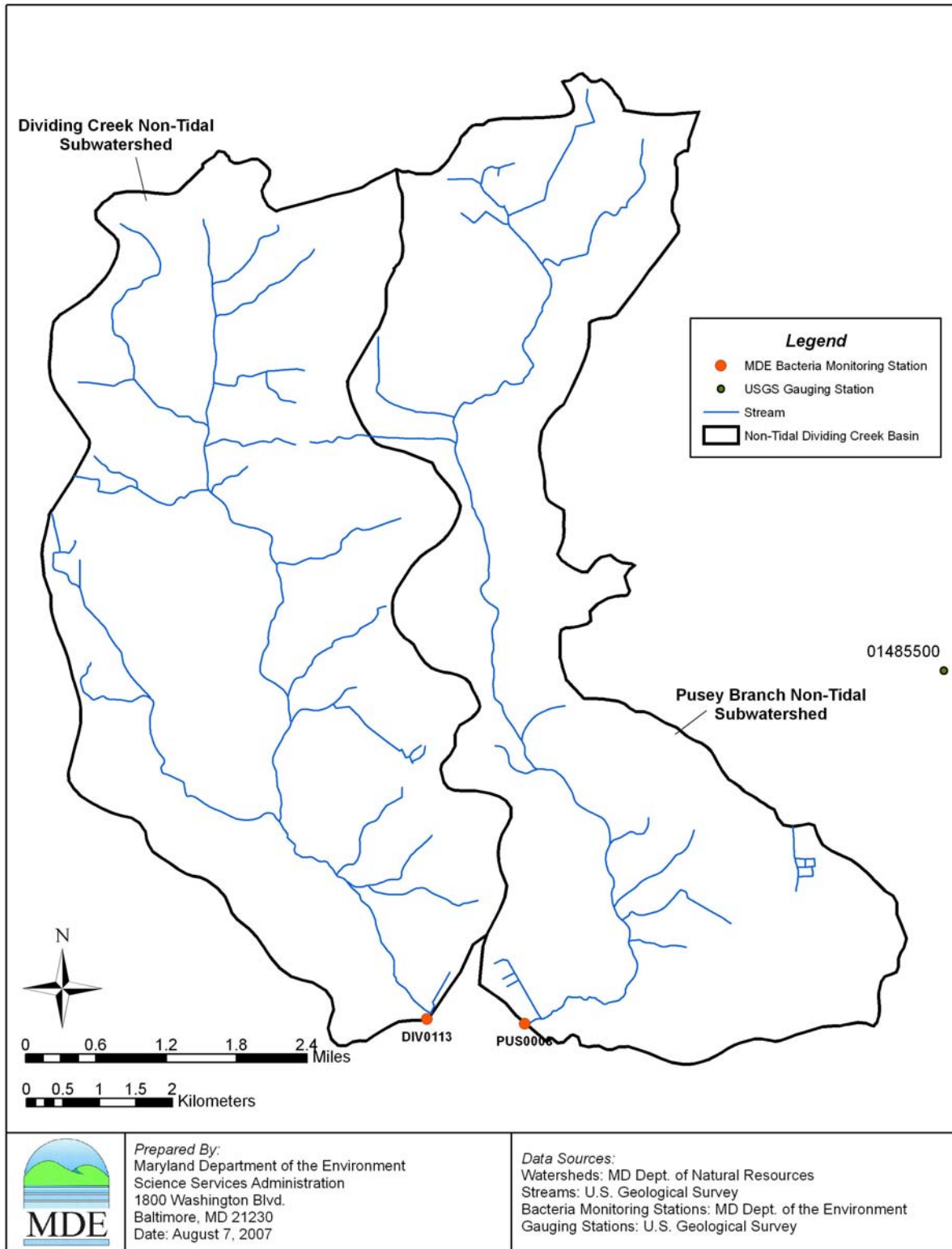
<b>Organization</b>	<b>Date</b>	<b>Parameter</b>	<b>Summary</b>
MDE	11/2003 through 10/2004	<i>E. coli</i>	2 stations 2 samples per month
MDE	11/2003 through 10/2004	BST ( <i>Enterococcus</i> )	2 stations 1 sample per month

**Table 2.2.2: Locations of MDE Monitoring Stations in the Dividing Creek Watershed**

<b>Tributary</b>	<b>Station Code</b>	<b>Observation Period</b>	<b>Total Observations</b>	<b>Latitude (Decimal Degrees)</b>	<b>Longitude (Decimal Degrees)</b>
Dividing Creek	DIV0113	2003-2004	24	38.187	-75.553
Pusey Branch	PUS0008	2003-2004	24	38.186	-75.538

**Table 2.2.3: Location of USGS Gauging Station nearest the Dividing Creek Watershed**

<b>Site Number</b>	<b>Observation Period Used</b>	<b>Total Observations</b>	<b>Latitude (Decimal Degrees)</b>	<b>Longitude (Decimal Degrees)</b>
01485500	1981-2006	9132	38.229	-75.471



**Figure 2.2.1: Monitoring Stations for the Non-Tidal Dividing Creek Basin**

## 2.3 Water Quality Impairment

### Designated Uses and Water Quality Standard

The Maryland water quality standards Surface Water Use Designation for the Dividing Creek watershed is Use I (Water Contact Recreation, and Protection of Non-tidal Warmwater Aquatic Life) [COMAR 26.08.02.07F(5)]. The non-tidal portion of the Dividing Creek watershed has been included on the final 2006 Integrated 303(d) List as impaired by fecal coliform bacteria.

#### *Water Quality Criteria*

The State water quality standard for bacteria (*E. coli*) used in this study is as follows:

**Table 2.3.1: Bacteria Criteria Values**

(Source: COMAR 26.08.02.03-3 Water Quality Criteria Specific to Designated Uses; Table 1)

Indicator	Steady State Geometric Mean Indicator Density
<i>Freshwater</i>	
<i>E. coli</i>	126 MPN/100ml

#### *Interpretation of Bacteria Data for General Recreational Use*

The relevant portion (for freshwater) of the listing methodology pursuant to the 2006 Integrated 303(d) List for all Use Waters - Water Contact Recreation and Protection of Aquatic Life is as follows:

##### Recreational Waters

A steady-state geometric mean will be calculated with available data where there are at least five representative sampling events. The data shall be from samples collected during steady-state conditions and during the beach season (Memorial Day through Labor Day) to be representative of the critical condition. If the resulting steady-state geometric mean is greater than 126 *E. coli* MPN/100 ml in freshwater, the waterbody will be listed as impaired. If fewer than five representative sampling events for an area being assessed are available, data from the previous two years will be evaluated in the same way. The single sample maximum criterion applies only to beaches and is to be used for closure and advisory decisions based on short-term exceedances of the geometric mean portion of the standard.

## **Water Quality Assessment**

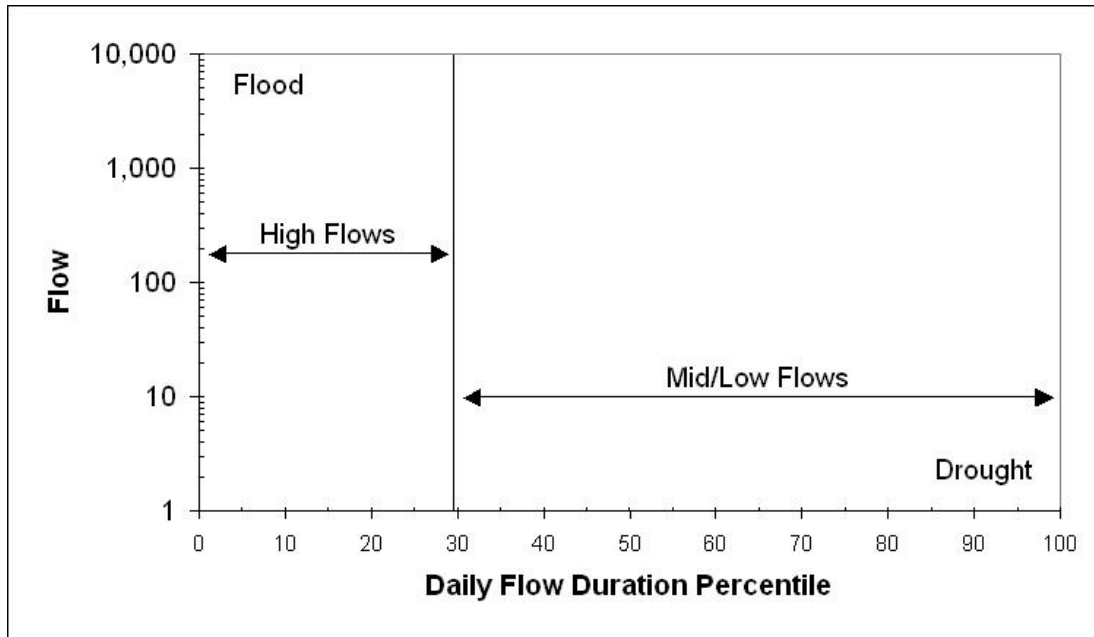
Bacteria water quality impairment in the non-tidal Dividing Creek basin was assessed by comparing both the annual and the seasonal (May 1<sup>st</sup>–September 30<sup>th</sup>) steady-state geometric means of *E. coli* concentrations with the water quality criterion.

The steady-state condition is defined as unbiased sampling targeting average flow conditions and/or equally sampling or providing for unbiased sampling of high and low flows. The 1986 EPA criteria document assumed steady-state flow in determining the risk at various bacterial concentrations, and therefore the chosen criterion value also reflects steady-state conditions (US EPA 1986). The steady-state geometric mean condition can be estimated either by monitoring design or more practically by statistical analysis as follows:

1. A stratified monitoring design is used where the number of samples collected is proportional to the duration of high flows, mid flows and low flows within the watershed. This sample design allows a geometric mean to be calculated directly from the monitoring data without bias.
2. Routine monitoring typically results in samples from varying hydrologic conditions (i.e., high flows, mid flows and low flows) where the numbers of samples are not proportional to the duration of those conditions. Averaging these results without consideration of the sampling conditions results in a biased estimate of the steady-state geometric mean. The potential bias of the steady-state geometric means can be reduced by weighting the samples' results collected during high flow, mid flow and low flow regimes by the proportion of time each flow regime is expected to occur. This ensures that the high flow and low flow conditions are proportionally balanced.
3. If (1) the monitoring design was not stratified based on flow regime or (2) flow information is not available to weight the samples accordingly, then a geometric mean of sequential monitoring data can be used as an estimate of the steady-state geometric mean condition for the specified period.

A routine monitoring design was used to collect bacteria data for the non-tidal portion of the Dividing Creek watershed. To estimate the steady-state geometric mean, the monitoring data were first reviewed by plotting the sample results versus their corresponding daily flow duration percentile. Graphs illustrating these results can be found in Appendix B.

To calculate the steady-state geometric mean with routine monitoring data, a conceptual model was developed by dividing the daily flow frequency for the stream segment into strata that are representative of hydrologic conditions. A conceptual continuum of flows is illustrated in Figure 2.3.1.



**Figure 2.3.1: Conceptual Diagram of Flow Duration Zones**

During high flows, a significant portion of the total stream flow is from surface flow contributions. Low flow conditions represent periods with minimal rainfall and surface runoff. There is typically a transitional mid flow period between the high and low flow durations, representative of varying contributions of surface flow inputs that result from differing rainfall volumes and antecedent soil moisture conditions. The division of the entire flow regime into strata enables the estimation of a less biased geometric mean from routine monitoring data that more closely approaches steady-state. Based on flow data of USGS gage 01485500 it was determined that the long-term average daily flow corresponds to a daily flow duration of 29.5%. Hence for this analysis it is defined that flows greater than the 29.5 percentile flow represent high flows, and flows less than the 29.5 percentile flow represent mid/low flows. A detailed method of how the flow strata were defined is presented in Appendix B.

Factors for estimating a steady-state geometric mean are based on the frequency of each flow stratum. The weighting factor accounts for the proportion of time each flow stratum represents. The weighting factors for an average hydrological year used in the non-tidal Dividing Creek TMDL analysis are presented in Table 2.3.2.

**Table 2.3.2: Weighting Factors for Average Hydrology Year Used for Estimation of Geometric Means in the Non-Tidal Dividing Creek Basin**

Flow Duration Zone	Duration Interval	Weighting Factor
High Flows	0 – 29.5%	0.295
Mid/Low Flows	29.5 – 100%	0.705

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Bacteria enumeration results for samples within a specified stratum will receive their corresponding weighting factor. The steady-state geometric mean is calculated as follows:

$$M = \sum_{i=1}^2 M_i * W_i \quad (1)$$

where,

$$M_i = \frac{\sum_{j=1}^{n_i} \log_{10}(C_{i,j})}{n_i} \quad (2)$$

- M = log weighted mean
- M<sub>i</sub> = log mean concentration for stratum *i*
- W<sub>i</sub> = proportion of stratum *i*
- C<sub>i,j</sub> = concentration for sample *j* in stratum *i*
- n<sub>i</sub> = number of samples in stratum

Finally, the steady-state geometric mean concentration is estimated using the following equation:

$$C_{gm} = 10^M \quad (3)$$

where,

C<sub>gm</sub> = Steady-state geometric mean concentration

Tables 2.3.3 and 2.3.4 present the maximum and minimum concentrations and the geometric means by stratum, and the overall steady-state geometric mean for the non-tidal Dividing Creek subwatersheds for the annual and seasonal (May 1<sup>st</sup> – September 30<sup>th</sup>) periods. For the seasonal period, no samples fell in the high flow zone. As such, for the seasonal analysis, only the overall geometric mean for the period was applied.

**Table 2.3.3: Non-Tidal Dividing Creek Basin Annual Steady-State Geometric Mean by Flow Stratum per Monitoring Station**

Station / Tributary	Flow Stratum	Number of Samples	<i>E. coli</i> Minimum Concentration (MPN/100ml)	<i>E. coli</i> Maximum Concentration (MPN/100ml)	Annual Steady State Geometric Mean (MPN/100ml)	Annual Weighted Geometric Mean (MPN/100ml)
DIV0113 Dividing Creek	High	8	10	364	89	238
	Low	16	20	2005	360	
PUS0008 Pusey Branch	High	8	10	113	47	74
	Low	16	10	2005	89	

**Table 2.3.4: Non-Tidal Dividing Creek Basin Seasonal Period (May 1 – September 30) Steady-State Geometric Mean per Monitoring Station**

Station / Tributary	Number of Samples	<i>E. coli</i> Minimum Concentration (MPN/100ml)	<i>E. coli</i> Maximum Concentration (MPN/100ml)	Seasonal Steady State Geometric Mean (MPN/100ml)
DIV0113 Dividing Creek	10	192	2005	515
PUS0008 Pusey Branch	10	31	2005	172

## 2.4 Source Assessment

### Nonpoint Source Assessment

Nonpoint sources of fecal bacteria do not have one discharge point but occur over the entire length of a stream or waterbody. During rain events, surface runoff transports water and fecal bacteria over the land surface and discharges to the stream system. This transport is dictated by rainfall, soil type, land use, and topography of the watershed. Many types of nonpoint sources introduce fecal bacteria to the land surface, including the manure spreading process, direct deposition from livestock during the grazing season, and excretions from pets and wildlife. The deposition of non-human fecal bacteria directly to the stream occurs when livestock or wildlife



have direct access to the waterbody. Nonpoint source contributions from human sources generally arise from failing septic systems and their associated drain fields or from leaking infrastructure (i.e., sewer systems).

### *Sewer Systems*

There is no sewage collection system in the non-tidal Dividing Creek basin.

### *Septic Systems*

All households in the non-tidal Dividing Creek basin rely on on-site disposal (septic) systems. Table 2.4.1 presents the number of septic systems in each subwatershed. Figure 2.4.1 displays the locations of these septic systems.

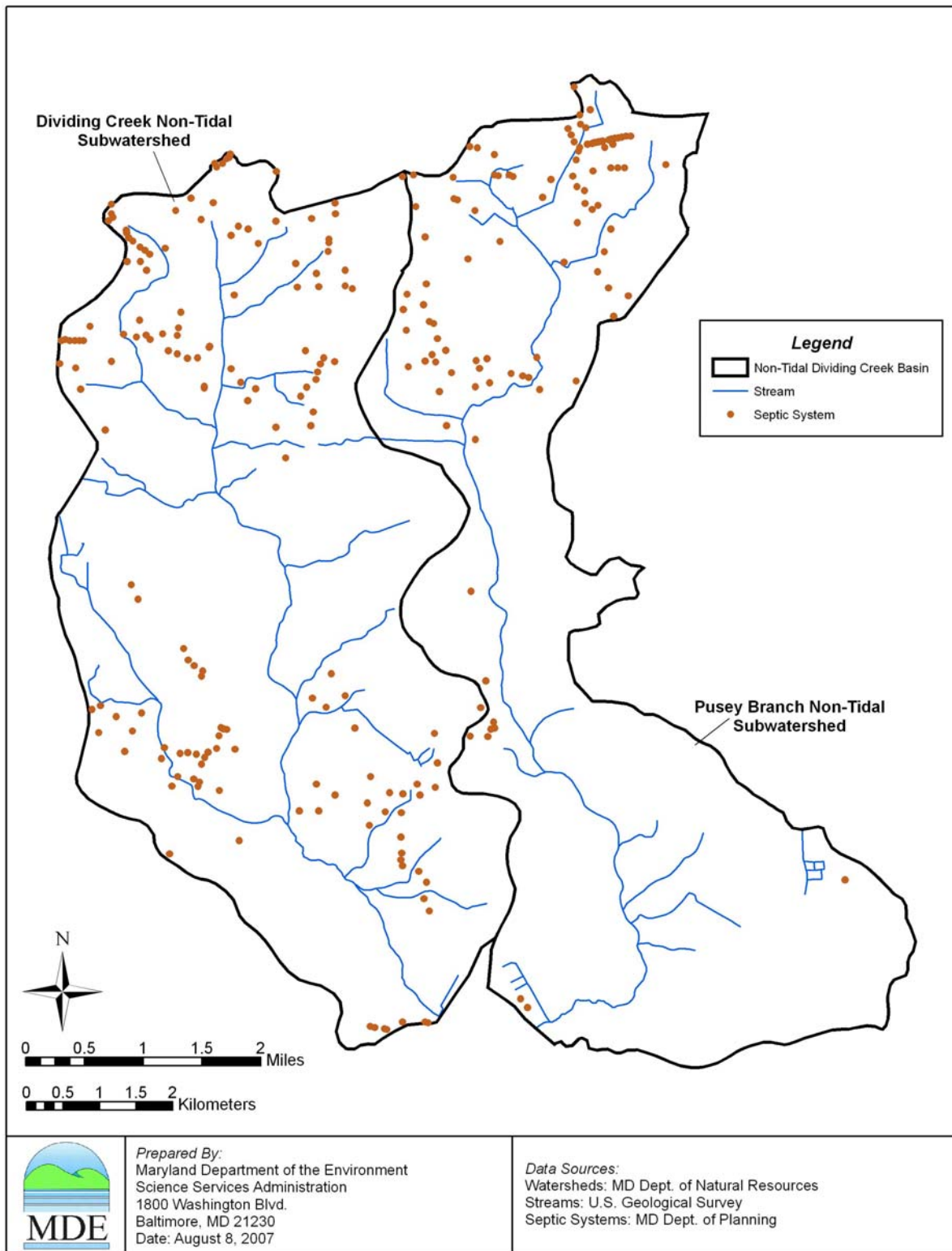
**Table 2.4.1: Septic Systems and Households Per Subwatershed in the Non-Tidal Dividing Creek Basin**

<b>Tributary / Station</b>	<b>Septic Systems</b>	<b>Households</b>
Dividing Creek DIV0113	164	149
Pusey Branch PUS0008	102	104
<b><i>Total</i></b>	<b>266</b>	<b>253</b>

### **Point Source Assessment**

There are two broad types of National Pollutant Discharge Elimination System (NPDES) permits considered in the analysis, individual and general. Both types of permits include industrial and municipal categories. Individual permits are issued for industrial and municipal WWTPs and Phase I municipal separate storm sewer systems (MS4s). MDE general permits have been established for surface water discharges from: Phase II and other MS4 entities, surface coal mines, mineral mines, quarries, borrow pits, ready-mix concrete, asphalt plants, seafood processors, hydrostatic testing of tanks and pipelines, marinas, concentrated animal feeding operations, and stormwater associated with industrial activities.

There are no MS4s, WWTPs, or other NPDES-regulated permit holders located in the non-tidal portion of the Dividing Creek watershed.



**Figure 2.4.1: Septic Systems in the Non-Tidal Dividing Creek Basin**

### **Bacteria Source Tracking**

Bacteria source tracking (BST) was used to identify the relative contribution of different sources of bacteria to in-stream water samples. BST monitoring was conducted at two stations in the non-tidal portion of the Dividing Creek watershed, where samples were collected once per month for a one-year duration. Sources are defined as domestic (pets and human associated animals), human (human waste), livestock (agricultural animals), and wildlife (mammals and waterfowl). To identify sources, samples are collected within the watershed from known fecal sources, and the patterns of antibiotic resistance of these known sources are compared to isolates of unknown bacteria from ambient water samples. Details of the BST methodology and data can be found in Appendix C.

An accurate representation of the expected contribution of each source at each station is estimated by using a stratified weighted mean of the identified sample results. The weighting factors are based on the  $\log_{10}$  of the bacteria concentration and the percent of time that represents the high stream flow or low stream flow (see Appendix B). The procedure for calculating the stratified weighted mean of the sources per monitoring station is as follows:

1. Calculate the percentage of isolates per source per each sample date (S).
2. Calculate the weighted percentage (MS) of each source per flow strata (high/low). The weighting is based on the  $\log_{10}$  bacteria concentration for the water sample.
3. The final weighted mean source percentage, for each source category, is based on the proportion of time in each flow duration zone.

The weighted mean for each source category is calculated using the following equations:

$$MS_k = \sum_{i=1}^2 MS_{i,k} * W_i \quad (4)$$

where,

$$MS_{i,k} = \frac{\sum_{j=1}^{n_i} \log_{10}(C_{i,j}) * S_{i,j,k}}{\sum_{j=1}^{n_i} \log_{10}(C_{i,j})} \quad (5)$$

where,

- $MS_k$  = weighted mean proportion of isolates of source k
- $MS_{i,k}$  = weighted mean proportion of isolates for source k in stratum i
- $W_i$  = proportion covered by stratum i
- i = stratum
- j = sample
- k = source category (1=human, 2=domestic, 3=livestock, 4=wildlife, 5=unknown)
- $C_{i,j}$  = concentration for sample j in stratum i
- $S_{i,j,k}$  = proportion of isolates for sample j, of source k in stratum i
- $n_i$  = number of samples in stratum i

The complete distributions of the annual and seasonal periods source loads are listed in Tables 2.4.2 and 2.4.3. Details of the BST data and tables with the BST analysis results can be found in Appendix C.

There are no potential human bacteria sources in the Dividing Creek watershed; therefore, human samples for BST analysis were not taken. Very few homes are present in the overall basin. Their locations are remote from either direct access or conduit connections to stream flow of the creek, meaning that septic system issues are not present. The locations of the septic systems are such that a failure would pose a localized nuisance and not a water quality load.

In the seasonal period, no samples fell in the high flow category in either subwatershed; therefore, a distribution by flow stratum was not calculated due to an insufficient number of samples. For the seasonal analysis, all samples between May 1<sup>st</sup> and September 30<sup>th</sup> were used to calculate an average seasonal distribution.

**Table 2.4.2: Distribution of Fecal Bacteria Source Loads in the Non-Tidal Dividing Creek Basin for the Average Annual Period**

Station	Flow Stratum	% Domestic Animals	% Human	% Livestock	% Wildlife	% Unknown
DIV0113	High	14.1	0.0	1.2	77.6	7.1
	Low	13.5	0.0	0.2	81.9	4.3
	Weighted	13.7	0.0	0.5	80.6	5.2
PUS0008	High	2.9	0.0	6.5	87.8	2.9
	Low	2.9	0.0	8.9	64.2	24.0
	Weighted	2.9	0.0	8.2	71.2	17.8

**Table 2.4.3: Distribution of Fecal Bacteria Source Loads in the Non-Tidal Dividing Creek Basin for the Seasonal Period (May 1<sup>st</sup> – September 30<sup>th</sup>)**

Station	% Domestic Animals	% Human	% Livestock	% Wildlife	% Unknown
DIV0113	12.9	0.0	0.0	87.1	0.0
PUS0008	2.2	0.0	0.9	79.3	17.6

### **3.0 TARGETED WATER QUALITY GOAL**

The overall objective of the fecal bacteria TMDL set forth in this document is to establish the loading caps needed to ensure attainment of water quality standards in the non-tidal portion of the Dividing Creek watershed. These standards are described fully in Section 2.3, “Water Quality Impairment.”

## 4.0 TOTAL MAXIMUM DAILY LOADS AND SOURCE ALLOCATION

### 4.1 Overview

This section provides an overview of the non-tidal fecal bacteria TMDL development, with a discussion of the many complexities involved in estimating bacteria concentrations, loads and sources. The second section presents the analysis framework and how the hydrological, water quality and BST data are linked together in the TMDL process. The third section describes the analysis for estimating a representative geometric mean fecal bacteria concentration and baseline loads. This analysis methodology is based on available monitoring data and is specific to a free-flowing stream system. The fourth section addresses the critical condition and seasonality. The fifth section presents the margin of safety. The sixth section discusses annual average TMDL loading caps and how maximum daily loads are estimated. The seventh section presents TMDL scenario descriptions. The eighth section presents the load allocations. Finally, in section nine, the TMDL equation is summarized.

To be most effective, the TMDL provides a basis for allocating loads among the known pollutant sources in the watershed so that appropriate control measures can be implemented and water quality standards achieved. By definition, the TMDL is the sum of the individual waste load allocations (WLAs) for point sources and load allocations (LAs) for nonpoint sources and natural background sources. A margin of safety (MOS) is also included and accounts for the uncertainty in the analytical procedures used for water quality modeling, and the limits in scientific and technical understanding of water quality in natural systems. Although this formulation suggests that the TMDL be expressed as a load, the Code of Federal Regulations (40 CFR 130.2(i)) states that the TMDL can be expressed in terms of “mass per time, toxicity or other appropriate measure.”

For many reasons, bacteria are difficult to simulate in water quality models. They reproduce and die off in a non-linear fashion as a function of many environmental factors, including temperature, pH, turbidity (UV light penetration) and settling. They occur in concentrations that vary widely (i.e., over orders of magnitude) and an accurate estimation of source inputs is difficult to develop. Finally, limited data are available to characterize the effectiveness of any program or practice at reducing bacteria loads (Schueler 1999).

Bacteria concentrations, determined through laboratory analysis of in-stream water samples for bacteria indicators (e.g., enterococci), are expressed in either colony forming units (CFU) or most probable number (MPN) of colonies. The first method (Method 1600) is a direct estimate of the bacteria colonies (US EPA 1985). The second method is a statistical estimate of the number of colonies (ONPG MUG Standard Method 9223B, AOAC 991.15). Sample results indicate the extreme variability in the total bacteria counts (see Appendix A). The distribution of the sample results tends to be lognormal, with a strong positive skew of the data. Estimating loads of constituents that vary by orders of magnitude can introduce much uncertainty and result in large confidence intervals around the final results.

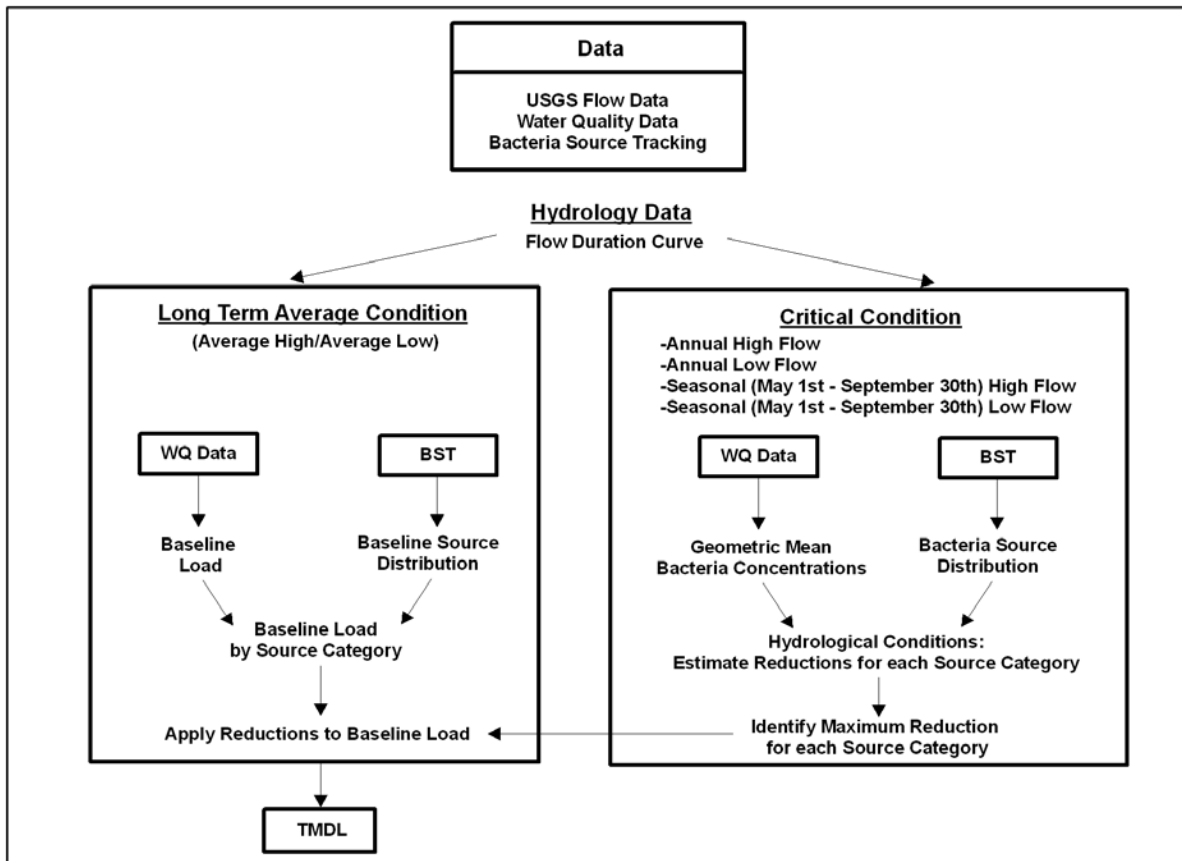
Estimating bacteria sources can also be problematic due to the many assumptions required and the limited data available. Lack of specific numeric and spatial location data for several source categories, from failing septic systems to domestic animals, livestock, and wildlife populations, can create many potential uncertainties in traditional water quality modeling. For this reason, MDE applies an analytical method combined with the bacteria source tracking described above for the calculation of this TMDL.

## **4.2 Analytical Framework**

This TMDL analysis uses flow duration curves to identify flow intervals that are used as indicators of hydrological conditions (i.e., annual average and critical conditions). This analytical method, combined with water quality monitoring data and BST, provides reasonable results (Cleland 2003), a better description of water quality than traditional water quality modeling, and also meets TMDL requirements.

In brief, baseline loads are estimated first for each subwatershed by using bacteria monitoring data and long-term flow data. These baseline loads are divided into four bacteria source categories using the results of BST analysis. Next, the percent reduction required to meet the water quality criterion in each subwatershed is estimated from the observed bacteria concentrations after determining the critical condition and accounting for seasonality. Critical condition and seasonality are determined by assessing annual and seasonal hydrological conditions for high flow and low flow periods. Finally, TMDLs for each subwatershed are estimated by applying these percent reductions.

Figure 4.2.1 illustrates how the hydrological (flow duration curve), water quality and BST data are linked together for the TMDL development.



**Figure 4.2.1: Diagram of the Non-Tidal Bacteria TMDL Analysis Framework**

### 4.3 Estimating Baseline Loads

Baseline loads estimated in this TMDL analysis are reported as long-term average annual loads. These loads are estimated using geometric mean concentrations and bias correction factors (calculated from bacteria monitoring data) and daily average flows (estimated from long-term flow data).

The geometric mean concentration is calculated from the log transformation of the raw data. Statistical theory tells us that when back-transformed values are used to calculate average daily loads or total annual loads, the loads will be biased low (Richards 1998). To avoid this bias, a factor should be added to the log-concentration before it is back-transformed. There are several methods of determining this bias correction factor, ranging from parametric estimates resulting from the theory of the log-normal distribution to non-parametric estimates using a bias correction factor [Ferguson 1986; Cohn et al. 1989; Duan 1983]. There is much literature on the applicability and results from these various methods with a summary provided in Richards (1998). Each has advantages and conditions of applicability. A non-parametric estimate of the bias correction factor (Duan 1983) was used in this TMDL analysis.



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With calculated geometric means and arithmetic means for each flow stratum, the bias correction factors are estimated as follows:

$$F_{1i} = A_i/C_i \quad (6)$$

where,

- $F_{1i}$  = bias correction factor for stratum  $i$
- $A_i$  = long-term annual arithmetic mean for stratum  $i$
- $C_i$  = long-term annual geometric mean for stratum  $i$

Daily average flows are estimated for each flow stratum using the watershed area ratio approach, since nearby long-term monitoring data are available.

The loads for each stratum are estimated as follows:

$$L_i = Q_i * C_i * F_{1i} * F_2 \quad (7)$$

where,

- $L_i$  = daily average load (Billion MPN/day) at monitoring station for stratum  $i$
- $Q_i$  = daily average flow (cfs) for stratum  $i$
- $C_i$  = geometric mean for stratum  $i$
- $F_{1i}$  = bias correction factor for stratum  $i$
- $F_2$  = unit conversion factor (0.0245)

Finally, for each subwatershed, the baseline load is estimated as follows:

$$L = \sum_{i=1}^2 L_i * W_i \quad (8)$$

where,

- $L$  = daily average load at station (MPN/day)
- $W_i$  = proportion of stratum  $i$

In the non-tidal Dividing Creek basin, weighting factors of 0.295 for high flow and 0.705 for low/mid flows were used to estimate the annual baseline load expressed as Billion MPN *E. coli*/year. Results of the baseline load calculations are presented in Table 4.3.1.

**Table 4.3.1: Baseline Load Calculations**

Station	Area (mi <sup>2</sup> )	High Flow (Unit Flow = 3.16 cfs/mi <sup>2</sup> )			Low Flow (Unit Flow = 0.41 cfs/mi <sup>2</sup> )			<i>E. coli</i> Baseline Load (Billion MPN/year)
		Average Flow (cfs)	<i>E. coli</i> Concentration. (MPN/100ml)	Bias Correction Factor	Average Flow (cfs)	<i>E. coli</i> Concentration (MPN/100ml)	Bias Correction Factor	
DIV0113	19.5	61.7	89	1.83	7.9	360	1.54	54,011
PUS0008	16.6	52.6	47	1.30	6.8	89	2.95	19,618

#### 4.4 Critical Condition and Seasonality

Federal regulations (40 CFR 130.7(c)(1)) require TMDLs to take into account critical conditions for stream flow, loading, and water quality parameters. The intent of this requirement is to ensure that the water quality of the waterbody is protected during times when it is most vulnerable.

For this TMDL the critical condition is determined by assessing annual and seasonal hydrological conditions for high flow and low flow periods. Seasonality is assessed as the time period when water contact recreation is expected, specifically May 1<sup>st</sup> through September 30<sup>th</sup>. For this TMDL analysis, the average hydrological condition over a 25-year period has been estimated as 29.5% high flow and 70.5% low flow as defined in Appendix B. Using the definition of a high flow condition as occurring when the daily flow duration interval is less than 29.5% and a low flow condition as occurring when the daily flow duration interval is greater than 29.5%, critical hydrological condition can be estimated by the percent of high or low flows during a specific period.

Using long-term flow data from USGS station 01485500, critical condition and seasonality has been determined by assessing various hydrological conditions to account for seasonal and annual averaging periods. The four conditions listed in Table 4.4.1 were used to account for the annual, critical, and seasonal conditions.

**Table 4.4.1: Hydrological Conditions Used to Account for Critical Condition and Seasonality**

Hydrological Condition		Averaging Period	Water Quality Data Used	Fraction High Flow	Fraction Low Flow	Condition Period
Annual	Average	365 days	All	0.295	0.705	Long-Term Average
	Wet (High Flow)	365 days	All	0.582	0.418	Feb. 2003 – Feb. 2004
	Dry (Low Flow)	365 days	All	0.030	0.970	Aug. 2001 – Aug. 2002
Seasonal	Average	May 1 <sup>st</sup> – Sept. 30 <sup>th</sup>	May 1 <sup>st</sup> – Sept. 30 <sup>th</sup>	N/A	N/A	Long-Term Average For May – Sept. Period

The critical condition requirement is met by determining the maximum reduction per bacteria source that satisfies all hydrological conditions and meets the water quality standard, thereby minimizing the risk to water contact recreation. It is assumed that the reduction applied to a bacteria source category will be constant through all conditions.

The bacteria monitoring data for the two stations in the non-tidal Dividing Creek basin cover a sufficient temporal span (at least one year) to estimate annual conditions. However, sufficient data were not available for the seasonal period to consider high flow and low flow conditions. Since all samples of the seasonal period were taken during low flow, a geometric mean cannot be established for the high flow condition. Therefore an average geometric mean and average flow were used for the seasonal analysis.

The reductions of fecal bacteria required to meet water quality standards in each subwatershed of the non-tidal Dividing Creek basin are shown in Table 4.4.2.

**Table 4.4.2: Required Fecal Bacteria Reductions (by Hydrological Condition per Subwatershed) to Meet Water Quality Standards**

Station / Tributary	Hydrological Condition		Domestic Animals %	Human %	Livestock %	Wildlife %
<b>DIV0113</b> Dividing Creek	Annual	Average	95.0	0.0	0.0	42.3
		Wet	95.0	0.0	0.0	12.8
		Dry	95.0	0.0	0.0	60.6
	Seasonal	Average	95.0	0.0	0.0	74.0
	<b>Maximum Source Reduction</b>		<b>95.0</b>	<b>0.0</b>	<b>0.0</b>	<b>74.0</b>
<b>PUS0008</b> Pusey Branch	Annual	Average	0.0	0.0	0.0	0.0
		Wet	0.0	0.0	0.0	0.0
		Dry	0.0	0.0	0.0	0.0
	Seasonal	Average	75.0	0.0	75.0	28.8
	<b>Maximum Source Reduction</b>		<b>75.0</b>	<b>0.0</b>	<b>75.0</b>	<b>28.8</b>

#### 4.5 Margin of Safety

A margin of safety (MOS) is required as part of this TMDL in recognition of the many uncertainties in the understanding and simulation of bacteriological water quality in natural systems and in statistical estimates of indicators. As mentioned in Section 4.1, it is difficult to estimate stream loadings for fecal bacteria due to the variation in loadings across sample locations and time. Load estimation methods should be both precise and accurate to obtain the true estimate of the mean load. Refined precision in the load estimation is due to using a stratified approach along the flow duration intervals, thus reducing the variation in the estimates. Moreover, Richards (1998) reports that averaging methods are generally biased, and the bias increases as the size of the averaging window increases. Finally, accuracy in the load estimation is based on minimal bias in the final result when compared to the true value.

Based on EPA guidance, the MOS can be achieved through two approaches (US EPA 1991a). One approach is to reserve a portion of the loading capacity as a separate term in the TMDL (i.e.,  $TMDL = LA + WLA + MOS$ ). The second approach is to incorporate the MOS as conservative assumptions used in the TMDL analysis. The second approach was used for this TMDL by estimating the loading capacity of the stream based on a reduced (more stringent) water quality criterion concentration. The *E. coli* water quality criterion concentration was reduced by 5%, from 126 *E. coli* MPN/100ml to 119.7 *E. coli* MPN/100ml.

## 4.6 Scenario Descriptions

### Source Distribution

The final bacteria source distribution and corresponding baseline loads are derived from the source proportions listed in Table 2.4.2. For the purposes of the TMDL analysis and allocations, the percentage of sources identified as “unknown” was removed and redistributed proportionally among the known sources to total 100%. The source distribution and baseline loads used in the TMDL scenarios are presented in Table 4.6.1.

**Table 4.6.1: Bacteria Source Distributions and Corresponding Baseline Loads Used in the Annual Average TMDL Analysis**

Station / Tributary	Domestic		Human		Livestock		Wildlife		Total Load (Billion <i>E. coli</i> MPN/year)
	%	Load (Billion <i>E. coli</i> MPN/year)	%	Load (Billion <i>E. coli</i> MPN/year)	%	Load (Billion <i>E. coli</i> MPN/year)	%	Load (Billion <i>E. coli</i> MPN/year)	
<b>DIV0113</b> Dividing Creek	14.4	7,800	0.0	0.0	0.6	299	85.0	45,912	54,011
<b>PUS0008</b> Pusey Branch	3.5	691	0.0	0.0	9.9	1,947	86.6	16,980	19,618

### First Scenario: Fecal Bacteria Practicable Reduction Targets

The maximum practicable reduction (MPR) for each of the four source categories is listed in Table 4.6.2. These values are based on review of the available literature and best professional judgment. It is assumed that human sources would potentially have the highest risk of causing gastrointestinal illness and therefore should have the highest reduction. If a domestic WWTP is located in the upstream watershed, this is considered in the MPR so as to not violate the permitted loads. The domestic animal category includes sources from pets (e.g., dogs) and the MPR is based on an estimated success of education and outreach programs.

**Table 4.6.2: Maximum Practicable Reduction Targets**

Max Practicable Reduction per Source	Human	Domestic	Livestock	Wildlife
	95%	75%	75%	0%
<b>Rationale</b>	(a) Direct source inputs. (b) Human pathogens more prevalent in humans than animals. (c) Enteric viral diseases spread from human to human. <sup>1</sup>	Target goal reflects uncertainty in effectiveness of urban BMPs <sup>2</sup> and is also based on best professional judgment	Target goal based on sediment reductions from BMPs <sup>3</sup> and best professional judgment	No programmatic approaches for wildlife reduction to meet water quality standards.  Waters contaminated by wild animal wastes offer a public health risk that is orders of magnitude less than that associated with human waste. <sup>4</sup>

<sup>1</sup>Health Effects Criteria for Fresh Recreational Waters. EPA-600/1-84-004. U.S. Environmental Protection Agency, Washington, DC. EPA. 1984.

<sup>2</sup>Preliminary Data Summary of Urban Storm Water Best Management Practices. EPA-821-R-99-012. U.S. Environmental Protection Agency, Washington, DC. EPA. 1999.

<sup>3</sup>Agricultural BMP Descriptions as Defined for The Chesapeake Bay Program Watershed Model. Nutrient Subcommittee Agricultural Nutrient Reduction Workshop. EPA. 2004.

<sup>4</sup>Environmental Indicators and Shellfish Safety. 1994. Edited by Cameron, R., Mackeney and Merle D. Pierson, Chapman & Hall.

As previously stated, these maximum practicable reduction targets are based on the available literature and best professional judgment. There is much uncertainty with estimated reductions from best management practices (BMP). The BMP efficiency for bacteria reduction ranged from -6% to +99% based on a total of 10 observations (US EPA 1999). The MPR to agricultural lands was based on sediment reductions identified by EPA (US EPA 2004).

The practicable reduction scenario was developed based on an optimization analysis whereby a subjective estimate of risk was minimized and constraints were set on maximum reduction and allowable background conditions. Risk was defined on a scale of one to five, where it was assumed that human sources had the highest risk (5), domestic animals and livestock next (3), and wildlife the lowest (1) (See Table 4.6.2). The model was defined as follows:

$$\text{Risk Score} = \text{Min} \sum_{i=1}^4 P_j * W_j \quad (9)$$

where,

$$P_j = \frac{(1 - R_i) * P b_j}{1 - TR} \quad (10)$$

and,

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$$TR = \frac{C - C_{cr}}{C} \quad (11)$$

Therefore the risk score can be represented as:

$$Risk\ Score = Min \sum_{i=1}^4 \left[ \frac{(1 - R_j) * P_{b_j}}{\left(1 - \frac{C - C_{cr}}{C}\right)} * W_j \right] \quad (12)$$

where,

- $i$  = hydrological condition
- $j$  = bacteria source category = human, domestic animal, livestock and wildlife
- $P_j$  = % of each source category (human, domestic animals, livestock and wildlife) in final allocation
- $W_j$  = weight of risk per source category = 5, 3 or 1
- $R_j$  = percent reduction applied by source category (human, domestic animals, livestock and wildlife) for the specified hydrological condition (variable)
- $P_{b_j}$  = original (baseline) percent distribution by source category (variable)
- TR = total reduction (constant within each hydrological condition) = Target reduction
- C = in-stream concentration
- Ccr = water quality criterion

The model is subject to the following constraints:

$$\begin{aligned} C &= C_{cr} \\ 0 \leq R_{human} &\leq 95\% \\ 0 \leq R_{pets} &\leq 75\% \\ 0 \leq R_{livestock} &\leq 75\% \\ R_{wildlife} &= 0 \\ P_j &\geq 1\% \end{aligned}$$

In both subwatersheds, the constraints of this scenario could not be satisfied, indicating there was not a practicable solution. A summary of the first scenario analysis results is presented in Table 4.6.3. No reduction was applied to human sources because human sources were not considered in the BST analysis.

**Table 4.6.3: Maximum Practicable Reduction Scenario Results**

Station / Tributary	Applied Reductions				Total Reduction %	Target Reduction %	Achievable?
	Domestic Animals %	Human %	Livestock %	Wildlife %			
<b>DIV0113</b> Dividing Creek	75.0	0.0	75.0	0.0	11.2	76.7	No
<b>PUS0008</b> Pusey Branch	75.0	0.0	75.0	0.0	10.3	35.1	No

### **Second Scenario: Fecal Bacteria Reductions Higher Than MPRs**

The TMDL must specify load allocations that will meet the water quality standards. In the practicable reduction targets scenario, neither of the two subwatersheds could meet water quality standards based on MPRs.

To further develop the TMDL, a second scenario was analyzed in which the constraints of the MPRs were relaxed. For both subwatersheds, the percentage reduction from baseline loads for all sources including wildlife was allowed to increase, as necessary to achieve water quality standards, up to a maximum of 95%. A similar optimization procedure as before was used to minimize risk. Again, the objective is to minimize the sum of the risk for all conditions while meeting the scenario reduction constraints. The model was defined in the same manner as considered in the practicable reduction scenario but subject to the following constraints:

$$\begin{aligned}
 C &= C_{cr} \\
 0 \leq R_j &\leq 95\% \\
 P_j &\geq 1\%
 \end{aligned}$$

A summary of the results from this second scenario analysis is presented in Table 4.6.4.



**Table 4.6.4: Reduction Results Based on Optimization Model Allowing up to 95% Reduction of All Sources**

Station / Tributary	Applied Reductions				Total Reduction %	Target Reduction %
	Domestic %	Human %	Livestock %	Wildlife %		
<b>DIV0113</b> Dividing Creek	95.0	0.0	0.0	74.0	76.7	76.7
<b>PUS0008</b> Pusey Branch	75.0	0.0	75.0	28.8	35.1	35.1

#### 4.7 TMDL Loading Caps

The TMDL loading cap is an estimate of the assimilative capacity of the monitored watershed. Estimation of the TMDL requires knowledge of how bacteria concentrations vary with flow rate or the flow duration interval. This relationship between concentration and flow is established using the strata defined by the flow duration curve.

The TMDL loading caps are provided in billion MPN *E. coli*/day. These loading caps are for the two subwatersheds located upstream of their respective monitoring stations: DIV0113 and PUS0008.

#### Annual Average TMDL Loading Caps

As explained in the sections above, the annual average TMDL loading caps are estimated by first determining the baseline or current condition loads for each subwatershed and the associated geometric mean from the available monitoring data. This annual average baseline load is estimated using the geometric mean concentration and the long-term annual average daily flow for each flow stratum. The loads from these two strata are then weighted to represent average conditions (see Table 4.3.1), based on the proportion of each stratum, to estimate the total long-term loading rate.

Next, the percent reduction required to meet the water quality criterion is estimated from the observed bacteria concentrations accounting for the critical conditions (See Section 4.4). A reduction in concentration is proportional to a reduction in load; thus the TMDL is equal to the current baseline load multiplied by one minus the required reduction. This reduction, estimated as explained in Section 4.4, represents the maximum reduction per source that satisfies all hydrological conditions in each subwatershed, and that is required to meet water quality standards.

$$\text{TMDL Loading Cap} = L_b * (1 - R) \quad (13)$$

where,

- $L_b$  = current or baseline load estimated from monitoring data  
 $R$  = reduction required from baseline to meet water quality criterion.

The annual average bacteria TMDL loading caps for the subwatersheds are shown in Tables 4.7.1 and 4.7.2.

**Table 4.7.1: Annual Average TMDL Loading Caps**

Station / Tributary	<i>E. coli</i>	Long-Term	% Target Reduction
	Baseline Load (Billion MPN/year)	Average <i>E. coli</i> TMDL Load (Billion MPN/year)	
<b>DIV0113</b> Dividing Creek	54,011	12,609	76.7
<b>PUS0008</b> Pusey Branch	19,618	12,741	35.1
<b>Total</b>	<b>73,630</b>	<b>25,341</b>	<b>65.6</b>

**Table 4.7.2: Annual Average TMDL Loading Caps by Source Category**

Station / Tributary	Domestic		Human		Livestock		Wildlife		Total Load (Billion <i>E. coli</i> MPN/year)
	%	Load (Billion <i>E. coli</i> MPN/year)	%	Load (Billion <i>E. coli</i> MPN/year)	%	Load (Billion <i>E. coli</i> MPN/year)	%	Load (Billion <i>E. coli</i> MPN/year)	
<b>DIV0113</b> Dividing Creek	3.1	390	0.0	0	2.4	299	94.5	11,921	12,609
<b>PUS0008</b> Pusey Branch	1.4	173	0.0	0	3.8	487	94.8	12,082	12,741

### **Maximum Daily Loads**

Selection of an appropriate method for translating a TMDL based on a longer time period into one using a daily time period requires decisions regarding 1) the level of resolution, and 2) the level of protection. The level of resolution pertains to the amount of detail used in specifying the maximum daily load. The level of protection represents how often the maximum daily load (MDL) is expected to be exceeded. Draft EPA/TetraTech guidance on daily loads (Limno-Tech 2007) provides three categories of options for both level of resolution and level of protection, and discusses these categories in detail.

For the non-tidal Dividing Creek MDLs, a “representative daily load” option was selected as the level of resolution, and a value “that will be exceeded with a pre-defined probability” was selected as the level of protection. In these options, the MDLs are two single daily loads that correspond to the two flow strata, with an upper bound percentile that accounts for the variability of daily loads. The upper bound percentile and the MDLs were estimated following EPA’s “*Technical Support Document for Water Quality-Based Toxics Control*” (1991 TSD) (US EPA 1991); and “*Approaches For Developing a Daily Load Expression for TMDLs Computed for Longer Term Averages*” (US EPA 2006).

There are three steps to the overall process of estimating these MDLs. First, all the data available from each monitoring station are examined together by stratum and the percentile rank of the highest observed concentration (for each stratum at each station) is computed. The highest computed percentile rank is the upper bound percentile to be used in estimating the MDLs.

Secondly, the long-term annual average TMDL (see Table 4.7.1) concentrations are estimated for both high flow and low flow strata. This is conducted for each station using a statistical methodology (the “Statistical Theory of Rollback,” or “STR,” described more fully in Appendix D).

Third, based on the estimated long-term average (LTA) TMDL concentrations, the MDL for each flow stratum at each station is estimated using the upper boundary percentile computed in the first step above. Finally, MDLs are computed from these MDL concentrations and their corresponding flows.

Results of the fecal bacteria MDL analysis for the non-tidal Dividing Creek subwatersheds are shown in Table 4.7.3.

**Table 4.7.3: Maximum Daily Loads Summary**

Station / Tributary	Flow Stratum	Maximum Daily Load (Billion <i>E. coli</i> MPN/day)	
		by Stratum	Weighted by Stratum
<b>DIV0113</b> Dividing Creek	<b>High</b>	610	285
	<b>Low</b>	149	
<b>PUS0008</b> Pusey Branch	<b>High</b>	231	220
	<b>Low</b>	215	

See Appendix D for a more detailed explanation of the procedure for obtaining these daily loads.

#### 4.8 TMDL Allocation

The non-tidal Dividing Creek fecal bacteria TMDL is composed of the following components:

$$\text{TMDL} = \text{LA} + \text{WLA} + \text{MOS} \quad (14)$$

where,

LA = Load Allocation  
WLA = Waste Load Allocation  
MOS = Margin of Safety

The TMDL allocation includes load allocations (LA) for nonpoint sources and waste load allocations (WLA) for point sources including WWTPs and NPDES-regulated stormwater discharges. The Stormwater (SW) WLA includes any nonpoint source loads deemed to be transported and discharged by regulated stormwater systems. An explanation of the distribution of nonpoint source loads and point source loads to the LA and to the SW-WLA and WWTP-WLA is provided in the subsections that follow.

The margin of safety (MOS) is explicit and is incorporated in the analysis using a conservative assumption; it is not specified as a separate term. The assumption is that a 5% reduction of the criterion concentration established by MD to meet the applicable water quality standard will result in more conservative allowable loads of fecal bacteria, and thus provide the MOS. The final loads are based on average hydrological conditions, with reductions estimated based on critical hydrological conditions. The load reduction scenario results in load allocations that will achieve water quality standards. The State reserves the right to revise these allocations provided such revisions are consistent with the achievement of water quality standards.

### **Bacteria Source Categories and Allocation Distributions**

The bacteria sources are grouped into four categories that are also consistent with divisions for various management strategies. The categories are human, domestic animal, livestock and wildlife. TMDL allocation rules are presented in Table 4.8.1. This table identifies how the TMDL will be allocated among the LA (those nonpoint sources or portions thereof not transported and discharged by stormwater systems) and the WLA (point sources including WWTPs and NPDES regulated stormwater discharges). Only the final LA or WLA is reported in this TMDL.

**Table 4.8.1: Potential Source Contributions for TMDL Allocation Categories**

Source Category	TMDL Allocation Categories		
	LA	WLA	
		WWTP	Stormwater
Human			
Domestic	X		
Livestock	X		
Wildlife	X		

#### **LA**

Human bacteria contributions were not considered in this TMDL analysis since there are no potential sources of human bacteria in the non-tidal Dividing Creek basin.

Livestock loads are all assigned to the LA. Since there are no MS4s or other NPDES-regulated stormwater systems, the domestic animals (pets) and wildlife allocations are also assigned to the LA.

#### **WLA**

##### ***NPDES Regulated Stormwater***

There are no MS4s permits or other NPDES-regulated Phase I or Phase II stormwater discharges in the non-tidal Dividing Creek basin.

***Municipal and Industrial WWTPs***

There are no municipal or industrial WWTPs with NPDES permits in the non-tidal Dividing Creek basin.

**4.9 Summary**

The long-term annual average TMDL and TMDL allocations are presented in Table 4.9.1. Table 4.9.2 presents the maximum daily loads for the subwatersheds.

**Table 4.9.1: Annual Average TMDL**

Station / Tributary	Total Allocation	LA	SW-WLA	WWTP-WLA
		(Billion MPN <i>E. coli</i> /year)		
<b>DIV0113</b> Dividing Creek	12,609	12,609	N/A	N/A
<b>PUS0008</b> Pusey Branch	12,741	12,741	N/A	N/A
<b>Total</b>	<b>25,351</b>	<b>25,351</b>	<b>0</b>	<b>0</b>

**Table 4.9.2: Maximum Daily Loads**

Station / Tributary	Total Allocation	LA	SW-WLA	WWTP-WLA
		(Billion MPN <i>E. coli</i> /day)		
<b>DIV0113</b> Dividing Creek	285	285	N/A	N/A
<b>PUS0008</b> Pusey Branch	220	220	N/A	N/A
<b>Total</b>	<b>505</b>	<b>505</b>	<b>0</b>	<b>0</b>

The long-term annual average fecal bacteria TMDL summary for the entire non-tidal Dividing Creek basin is presented in Table 4.9.3.

**Table 4.9.3: Annual Average TMDL Summary**

<b>TMDL</b>	<b>=</b>	<b>LA</b>	<b>+</b>	<b>WLA</b>	<b>+</b>	<b>MOS</b>
<b>(Billion MPN <i>E. Coli</i> /year)</b>						
<b>25,351</b>	<b>=</b>	<b>25,351</b>	<b>+</b>	<b>0</b>	<b>+</b>	<b>Incorporated</b>

The fecal bacteria MDL summary for the entire non-tidal Dividing Creek basin is presented in Table 4.9.4.

**Table 4.9.4: MDL Summary**

<b>TMDL</b>	<b>=</b>	<b>LA</b>	<b>+</b>	<b>WLA</b>	<b>+</b>	<b>MOS</b>
<b>(Billion MPN <i>E. Coli</i> /day)</b>						
<b>505</b>	<b>=</b>	<b>505</b>	<b>+</b>	<b>0</b>	<b>+</b>	<b>Incorporated</b>

In certain watersheds, the goal of meeting water quality standards may require very high reductions that are not achievable with current technologies and management practices. In this situation, where there is no feasible TMDL scenario, MPRs are increased to provide estimates of the reductions required to meet water quality standards. In the subwatersheds of the non-tidal Dividing Creek basin, water quality standards cannot be achieved with the maximum practicable reduction rates specified in Table 4.6.3. The TMDLs shown in Tables 4.9.1 and 4.9.2 represent reductions from current bacteria loadings that are beyond practical reductions. In cases where such high reductions are required to meet standards, it is expected that the first stage of implementation will be to carry out the MPR scenario.

## 5.0 ASSURANCE OF IMPLEMENTATION

Section 303(d) of the Clean Water Act and current EPA regulations require reasonable assurance that the TMDL load and waste load allocations can and will be implemented. In the non-tidal portion of the Dividing Creek watershed, the TMDL analysis indicates that, for both of the two subwatersheds, the reduction of fecal bacteria loads from wildlife are beyond the MPR targets. And for one of the two subwatersheds, the reduction of fecal bacteria loads from domestic animals is beyond the MPR target. These MPR targets were defined based on a literature review of BMPs effectiveness and assuming a zero reduction for wildlife sources. The non-tidal Dividing Creek, Pusey Branch and their tributaries may not be able to attain water quality standards. The fecal bacteria load reductions required to meet water quality criteria in the non-tidal portion of the Dividing Creek basin are not feasible by implementing effluent limitations and cost-effective, reasonable BMPs to nonpoint sources. Therefore, MDE proposes a staged approach to implementation beginning with the MPR scenario, with regularly scheduled follow-up monitoring to assess the effectiveness of the implementation plan.

Additional reductions will be achieved through the implementation of BMPs; however, the literature reports considerable uncertainty concerning the effectiveness of BMPs in treating bacteria. As an example, pet waste education programs have varying results based on stakeholder involvement. Additionally, the extent of wildlife reduction associated with various BMPs methods (e.g., structural, non-structural, etc.) is uncertain. Therefore, MDE intends for the required reductions to be implemented in an iterative process that first addresses those sources with the largest impact on water quality and human health risk, with consideration given to ease of implementation and cost. The iterative implementation of BMPs in the watershed has several benefits: tracking of water quality improvements following BMP implementation through follow-up stream monitoring; providing a mechanism for developing public support through periodic updates on BMP implementation; and helping to ensure that the most cost-effective practices are implemented first.

Potential funding sources for implementation include the Maryland's Agricultural Cost Share Program (MACS), which provides grants to farmers to help protect natural resources, and the Environmental Quality and Incentives Program, which focuses on implementing conservation practices and BMPs on land involved with livestock and production. Though not directly linked, it is assumed that the nutrient management plans from the Water Quality Improvement Act of 1998 (WQIA) will have some reduction of bacteria from manure application practices.

### **Implementation and Wildlife Sources**

It is expected that, in some waters for which TMDLs will be developed, the bacteria source analysis indicates that after controls are in place for all anthropogenic sources, the waterbody will not meet water quality standards. However, while neither Maryland nor EPA is proposing the elimination of wildlife to allow for the attainment of water quality standards, managing the overpopulation of wildlife remains an option for state and local stakeholders.



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After developing and implementing, to the maximum extent possible, a reduction goal based on the anthropogenic sources identified in the TMDL, Maryland anticipates that implementation to reduce the controllable nonpoint sources may also reduce some wildlife inputs to the waters.

## **6.0 PUBLIC PARTICIPATION**

Stakeholders were informed by an October 18, 2007 MDE mailing of a notice of intent to develop a fecal bacteria TMDL for the non-tidal Dividing Creek basin. The notice letters provided MDE contact information and offered upon request an informational briefing on the proposed TMDL. MDE received no requests for a briefing. A follow-up notification was mailed to stakeholders on February 20, 2008 to announce the imminent release of the TMDL documents for public review and to offer again the opportunity for an informational briefing. No requests for a briefing were received.

A public notice of intent to establish the Dividing Creek fecal bacteria TMDL, announcing the opening and closing dates of the formal 30-day Public Comment Period, was published in the Somerset County Herald, the Wicomico Daily Times, and the Worcester County Times. The notice was also sent to MDE's stakeholder distribution list for the three counties in the Dividing Creek watershed and all other interested parties. All were invited to send written comments on the draft TMDL to MDE. The public notice announced the availability of the draft TMDL documents, which were placed in identified public libraries located in each of the three counties. The 30-day public notice also provided information on how to access the draft TMDL documents on MDE's website.

All written comments received by the close of the comment period are recorded and formally responded to in a Comment Response Document (CRD), to be included in the draft final TMDL documentation package submitted to EPA for the Agency's approval. Receipt of each set of comments is acknowledged by MDE, either by letter or email to comment authors. Following EPA approval of the TMDL, the responses are made available when the CRD is posted on MDE's website, together with the final approved TMDL documentation. The CRD is also mailed to stakeholders, including all those who sent comments to MDE, along with an approval notification letter.

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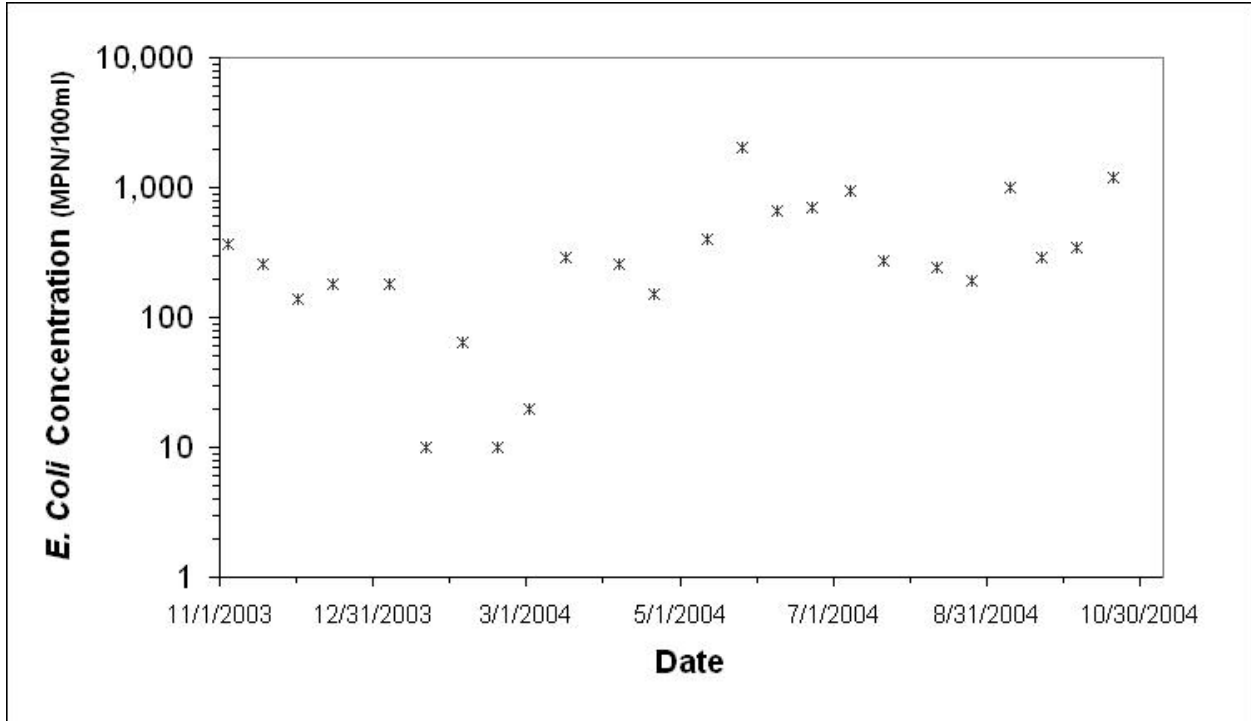
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## Appendix A – Bacteria Data

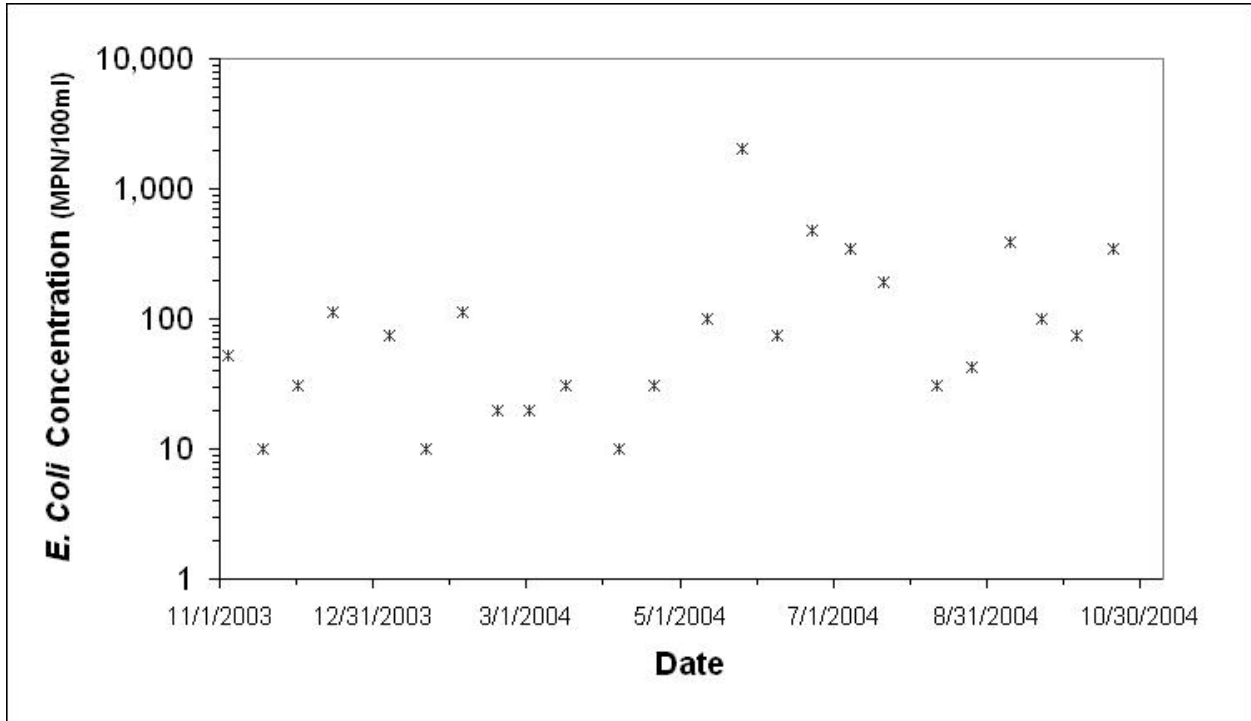
Table A-1: Measured Bacteria Concentration and Daily Flow Frequency

Station	Date	Daily Flow Frequency	<i>E. coli</i> Concentration (MPN/100ml)
DIV0113	11/04/2003	25.8213	364
	11/18/2003	30.7162	254
	12/02/2003	32.8734	137
	12/16/2003	0.8432	178
	01/07/2004	5.4314	178
	01/22/2004	11.7280	10
	02/05/2004	4.5445	64
	02/19/2004	25.1971	10
	03/03/2004	40.0241	20
	03/17/2004	35.1621	288
	04/07/2004	41.5572	254
	04/21/2004	24.6386	150
	05/12/2004	53.1318	406
	05/26/2004	70.7184	2,005
	06/09/2004	76.5878	659
	06/23/2004	84.8117	697
	07/08/2004	96.5944	945
	07/21/2004	71.9996	271
	08/11/2004	46.4082	244
	08/25/2004	47.3938	192
09/09/2004	50.6132	1013	
09/22/2004	44.5795	288	
10/06/2004	26.7740	344	
10/20/2004	53.1318	1,184	

<b>Station</b>	<b>Date</b>	<b>Daily Flow Frequency</b>	<b><i>E. coli</i> Concentration (MPN/100ml)</b>
<b>PUS0008</b>	11/04/2003	25.8213	53
	11/18/2003	30.7162	10
	12/02/2003	32.8734	31
	12/16/2003	0.8432	111
	01/07/2004	5.4314	75
	01/22/2004	11.7280	10
	02/05/2004	4.5445	113
	02/19/2004	25.1971	20
	03/03/2004	40.0241	20
	03/17/2004	35.1621	31
	04/07/2004	41.5572	10
	04/21/2004	24.6386	31
	05/12/2004	53.1318	99
	05/26/2004	70.7184	2,005
	06/09/2004	76.5878	75
	06/23/2004	84.8117	478
	07/08/2004	96.5944	344
	07/21/2004	71.9996	192
	08/11/2004	46.4082	31
	08/25/2004	47.3938	42
09/09/2004	50.6132	384	
09/22/2004	44.5795	99	
10/06/2004	26.7740	75	
10/20/2004	53.1318	344	



**Figure A-1: *E. coli* Concentration vs. Time for MDE Monitoring Station DIV0113**



**Figure A-2: *E. coli* Concentration vs. Time for MDE Monitoring Station PUS0008**

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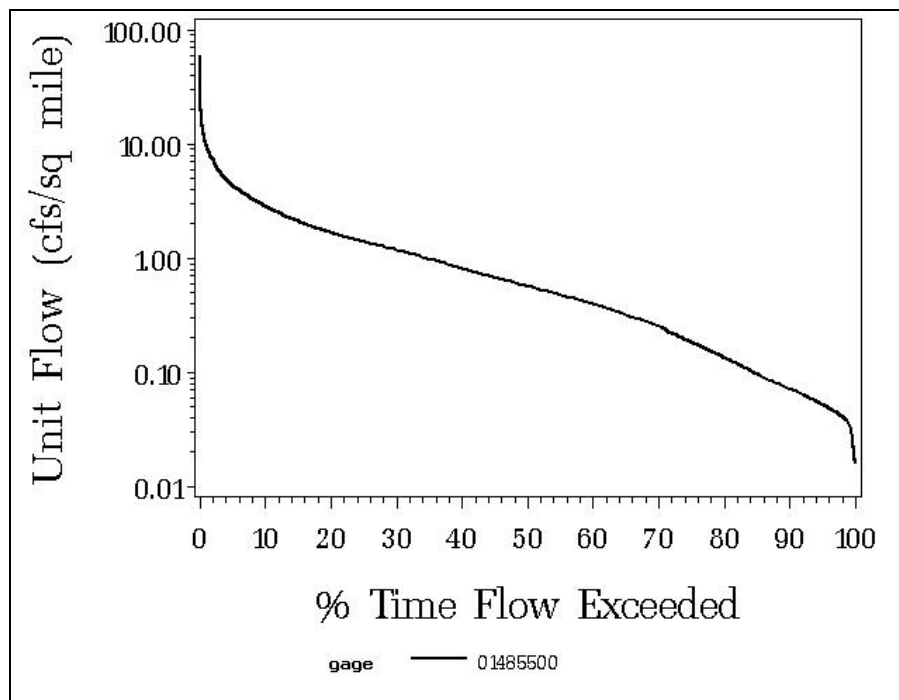
## Appendix B – Flow Duration Curve Analysis to Define Strata

The non-tidal Dividing Creek basin was assessed to determine hydrologically significant strata. The purpose of these strata is to apply weights to monitoring data and thus (1) reduce bias associated with the monitoring design and (2) approximate a critical condition for TMDL development. The strata group hydrologically similar water quality samples and provide a better estimate of the mean concentration at the monitoring station.

The flow duration curve for a watershed is a plot of all possible daily flows, ranked from highest to lowest, versus their probability of exceedance. In general, the higher flows will tend to be dominated by excess runoff from rain events and the lower flows will result from drought type conditions. The mid-range flows are a combination of high base flow with limited runoff and lower base flow with excess runoff. The range of these mid-level flows will vary with soil antecedent conditions. The purpose of the following analysis is to identify hydrologically significant groups, based on the previously described flow regimes, within the flow duration curve.

### Flow Analysis

A USGS gage station is not present within the non-tidal Dividing Creek basin. Thus the nearby gage station, #01485500 at Nassawango Creek near Snow Hill, MD was used for the analysis. The dates of information used were from October 1, 1981 to September 30, 2006. A flow duration curve for this gage station is presented in Figure B-1.



**Figure B-1: Flow Duration Curve for USGS Gage 01485500**

Based on the flow data from the Nassawango Creek gage station the long-term average daily unit flow is 1.21 cfs/sq. mile, which corresponds to a flow frequency of 29.5%. Using the definition of a high flow condition as occurring when flows are higher than the long-term average flow and a low flow condition as occurring when flows are lower than the long-term average flow, the 29.5 percentile threshold was selected to define the limits between high flows and low flows in this watershed. Therefore, a high flow condition will be defined as occurring when the daily flow duration percentile is less than 29.5% and a low flow condition will be defined as occurring when the daily flow duration percentile is greater than 29.5%. Definitions of high and low range flows are presented in Table B-1.

**Table B-1: Definition of Flow Regimes**

<b>High Flow</b>	Represents conditions where stream flow tends to be dominated by surface runoff.
<b>Low Flow</b>	Represents conditions where stream flow tends to be more dominated by groundwater flow.

### *Flow Data Analysis*

The final analysis to define the daily flow duration intervals (flow regions, strata) includes the bacteria monitoring data. Bacteria (*E. coli*) monitoring data are “placed” within the regions (strata) based on the daily flow duration percentile of the date of sampling. Figures B-2 to B-5 show the non-tidal Dividing Creek basin *E. coli* monitoring data with corresponding flow frequency for the average annual and the seasonal conditions.

Maryland’s water quality standards for bacteria state that, when available, the geometric mean indicator should be based on at least five samples taken representatively over 30 days. Therefore, in situations in which fewer than five samples “fall” within a particular flow regime interval, the interval and the adjacent interval will be joined. In the non-tidal Dividing Creek basin, for the annual average flow condition, there are sufficient samples in both the high flow and low flow strata to estimate the geometric means. However, in the seasonal (May 1<sup>st</sup> – September 30<sup>th</sup>) flow condition, there are no samples within the high flow strata; therefore, for this condition an average seasonal geometric mean will be calculated.

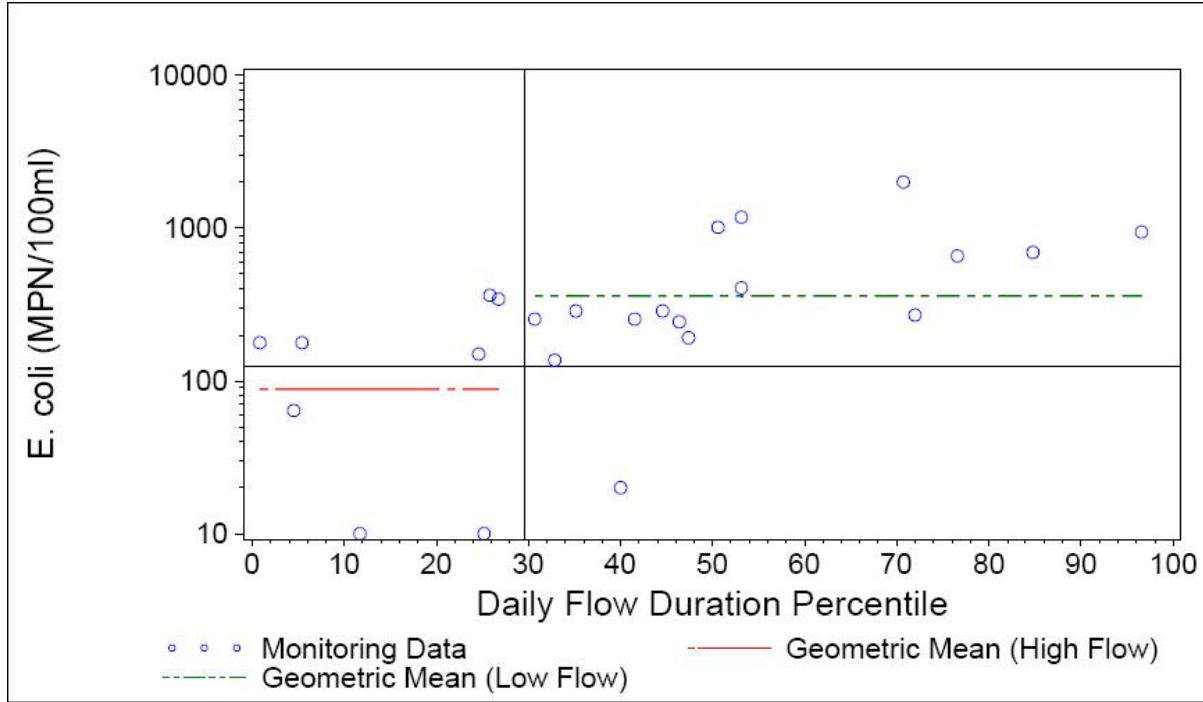
Weighting factors for estimating a weighted geometric mean are based on the frequency of each flow stratum during the averaging period. The weighting factors for the averaging periods and hydrological conditions are presented in Table B-2. Averaging periods are defined in this report as:

- (1) Average Annual Hydrological Condition
- (2) Annual High Flow Condition
- (3) Annual Low Flow Condition
- (4) Seasonal (May 1st – September 30th) Average Flow Condition

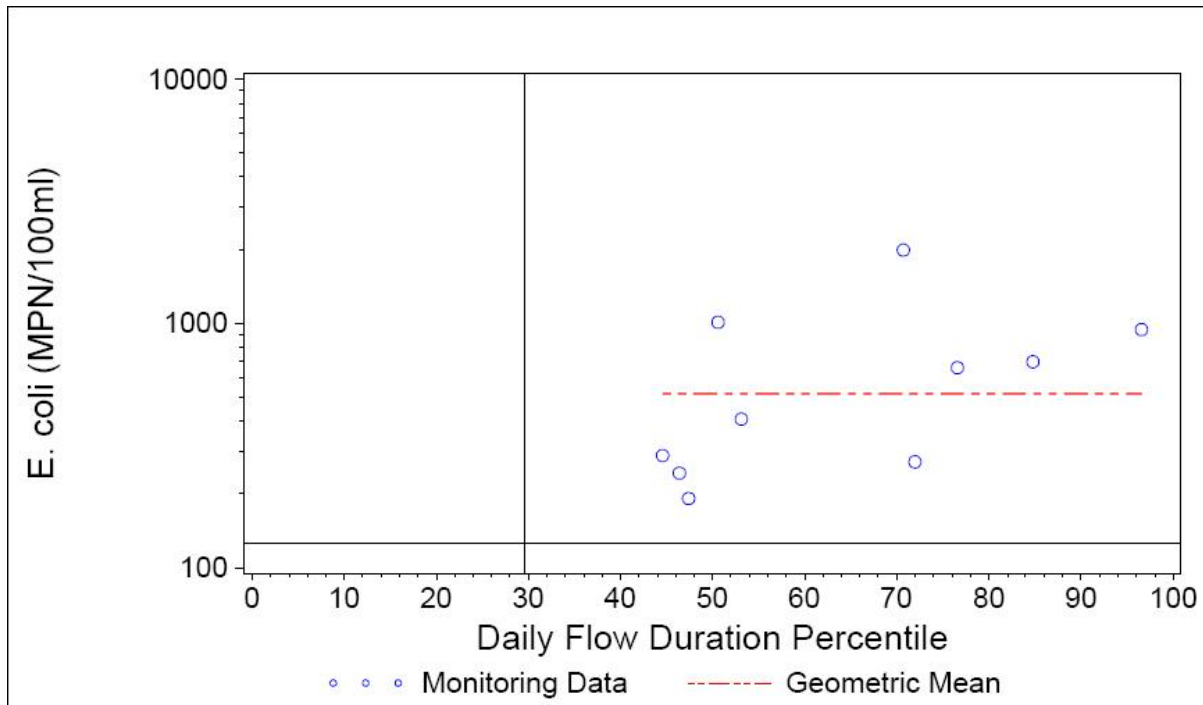
Weighted geometric means for the average annual and the seasonal conditions are plotted with the monitoring data on Figures B-2 to B-5.

**Table B-2: Weighting Factors for Estimation of Geometric Mean**

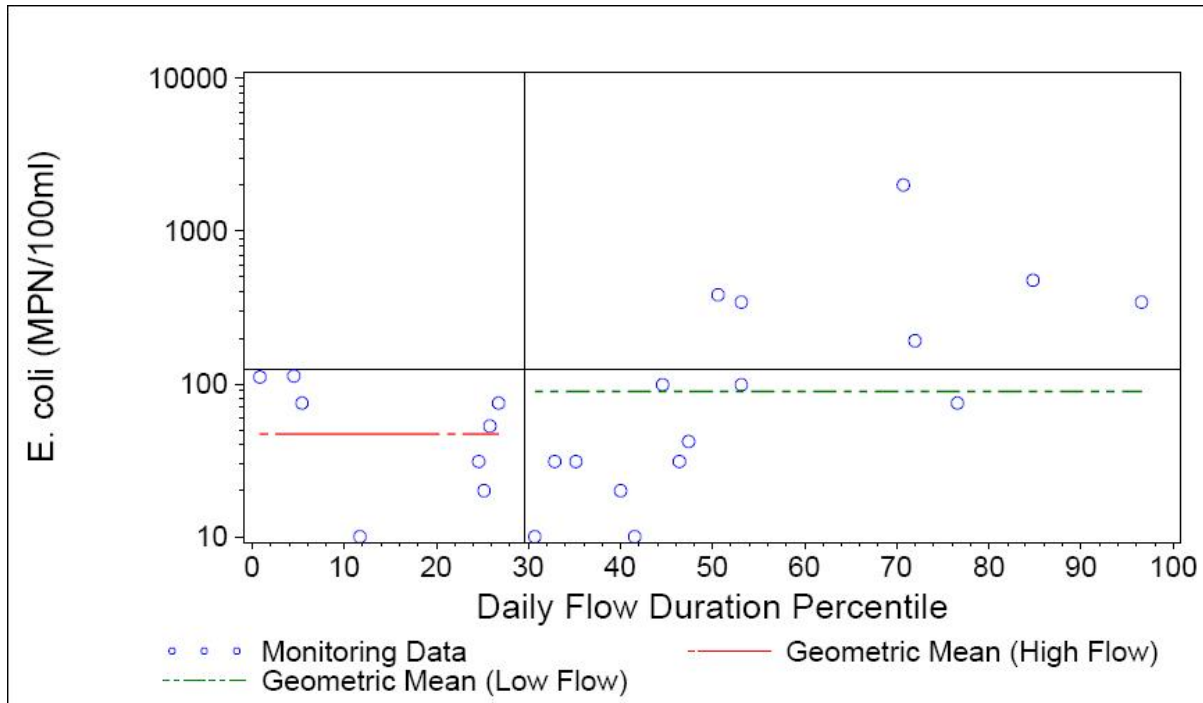
Hydrological Condition		Averaging Period	Water Quality Data Used	Fraction High Flow	Fraction Low Flow	Condition Period
Annual	Average	365 days	All	0.295	0.705	Long-term Average
	Wet	365 days	All	0.582	0.418	Feb. 2003 – Feb. 2004
	Dry	365 days	All	0.030	0.970	Aug. 2001 – Aug. 2002
Seasonal	Average	May 1 <sup>st</sup> – Sep. 30 <sup>th</sup>	May 1 <sup>st</sup> – Sep. 30 <sup>th</sup>	N/A		N/A



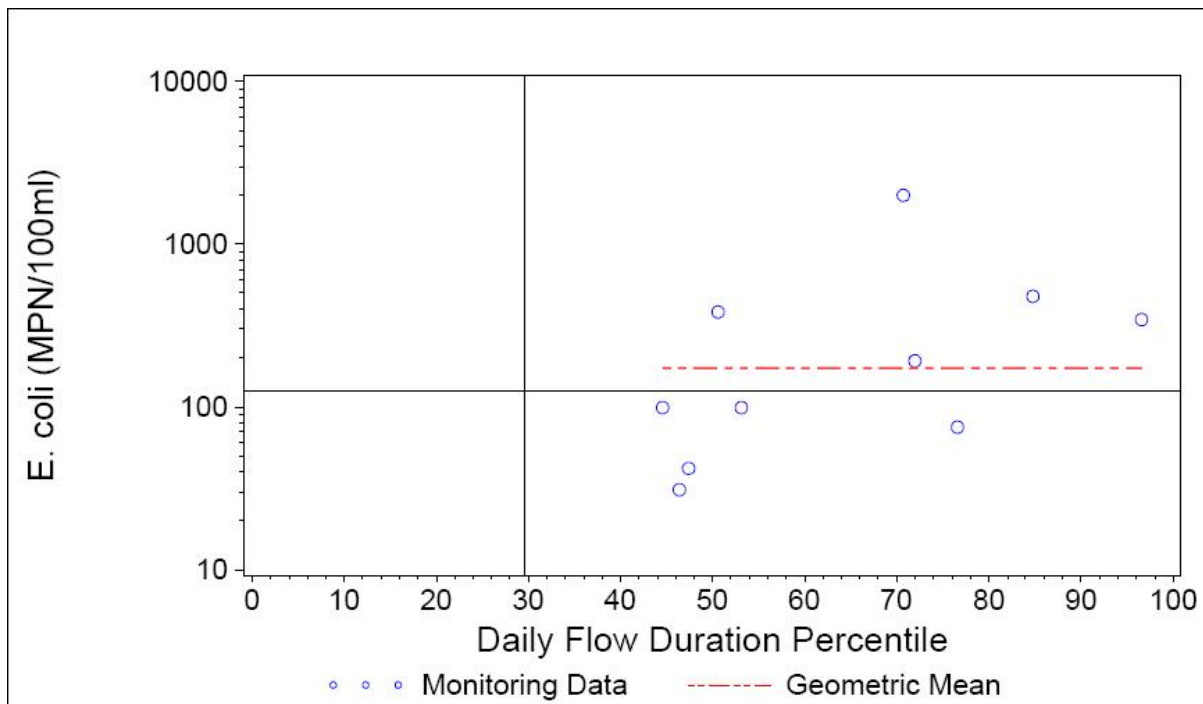
**Figure B-2: *E. coli* Concentration vs. Flow Duration for Monitoring Station DIV0113 (Annual Condition)**



**Figure B-3: *E. coli* Concentration vs. Flow Duration for Monitoring Station DIV0113 (Seasonal Condition)**



**Figure B-4: *E. coli* Concentration vs. Flow Duration for Monitoring Station PUS0008 (Annual Condition)**



**Figure B-5: *E. coli* Concentration vs. Flow Duration for Monitoring Station PUS0008 (Seasonal Condition)**

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**Appendix C – BST Report**

**Maryland Department of the Environment**

**Identifying Sources of Fecal Pollution in  
Shellfish and Nontidal Waters in  
Maryland Watersheds**

**November 2005 – June 2007**

**Mark F. Frana, Ph.D. and Elichia A. Venso, Ph.D.  
Department of Biological Sciences and Environmental Health Science  
Salisbury University, Salisbury, MD**

**Final Report  
June 30, 2007**

*Dividing Creek (Non-tidal) TMDL Fecal Bacteria  
Document version: March 5, 2008*

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## INTRODUCTION

**Microbial Source Tracking.** Microbial Source Tracking (MST) is a relatively recent scientific and technological innovation designed to distinguish the origins of enteric microorganisms found in environmental waters. Several different methods and a variety of different indicator organisms (both bacteria and viruses) have successfully been used for MST, as described in recent reviews (Scott *et al.*, 2002; Simpson *et al.*, 2002). When the indicator organism is bacteria, the term Bacterial Source Tracking (BST) is often used. Some common bacterial indicators for BST analysis include: *E. coli*, *Enterococcus* spp., *Bacteroides-Prevotella*, and *Bifidobacterium* spp.

Techniques for MST can be grouped into one of the following three categories: molecular (genotypic) methods, biochemical (phenotypic) methods, or chemical methods. Ribotyping, Pulsed-Field Gel Electrophoresis (PFGE), and Randomly-Amplified Polymorphic DNA (RAPD) are examples of molecular techniques. Biochemical methods include Antibiotic Resistance Analysis (ARA), F-specific coliphage typing, and Carbon Source Utilization (CSU) analysis. Chemical techniques detect chemical compounds associated with human activities, but do not provide any information regarding nonhuman sources. Examples of this type of technology include detection of optical brighteners from laundry detergents or caffeine (Simpson *et al.*, 2002).

Many of the molecular and biochemical methods of MST are “library-based,” requiring the collection of a database of fingerprints or patterns obtained from indicator organisms isolated from known sources. Statistical analysis determines fingerprints/patterns of known sources species or categories of species (*i.e.*, human, livestock, pets, wildlife). Indicator isolates collected from water samples are analyzed using the same MST method to obtain their fingerprints or patterns, which are then statistically compared to those in the library. Based upon this comparison, the final results are expressed in terms of the “statistical probability” that the water isolates came from a given source (Simpson *et al.* 2002).

**Antibiotic Resistance Analysis.** A variety of different host species can potentially contribute to the fecal contamination found in natural waters. Many years ago, scientists speculated on the possibility of using resistance to antibiotics as a way of determining the sources of this fecal contamination (Bell *et al.*, 1983; Krumpferman, 1983). In ARA, the premise is that bacteria isolated from different hosts can be discriminated based upon differences in the selective pressure of microbial populations found in the gastrointestinal tract of those hosts (humans, livestock, pets, wildlife) (Wiggins, 1996). Microorganisms isolated from the fecal material of wildlife would be expected to have a much lower level of resistance to antibiotics than isolates collected from the fecal material of humans, livestock and pets. In addition, depending upon the specific antibiotics used in the analysis, isolates from humans, livestock and pets could be differentiated from each other.

In ARA, isolates from known sources are tested for resistance or sensitivity against a panel of antibiotics and antibiotic concentrations. This information is then used to construct a library of antibiotic resistance patterns from known-source bacterial isolates. Microbial isolates collected from water samples are then tested and their resistance results are recorded. Based upon a

comparison of resistance patterns of water and library isolates, a statistical analysis can predict the likely host source of the water isolates. (Hagedorn 1999; Wiggins 1999).

## LABORATORY METHODS

**Isolation of *Enterococcus* from Known-Source Samples.** Fecal samples, identified to source, were delivered to the Salisbury University (SU) BST lab by Maryland Department of the Environment (MDE) personnel. Fecal material suspended in phosphate buffered saline was plated onto selective m-Enterococcus agar. After incubation at 37° C, up to eight (8) *Enterococcus* isolates were randomly selected from each fecal sample for ARA testing.

**Isolation of *Enterococcus* from Water Samples.** Water samples were collected by MDE staff and shipped overnight to MapTech Inc, Blacksburg, Va. Bacterial isolates were collected by membrane filtration. Up to 24 randomly selected *Enterococcus* isolates were collected from each water sample and all isolates were then shipped to the SU BST lab.

**Antibiotic Resistance Analysis.** Each bacterial isolate from both water and scat were grown in Enterococcosel<sup>®</sup> broth (Becton Dickinson, Sparks, MD) prior to ARA testing. *Enterococci* are capable of hydrolyzing esculin, turning this broth black. Only esculin-positive isolates were tested for antibiotic resistance.

Bacterial isolates were plated onto tryptic soy agar plates, each containing a different concentration of a given antibiotic. Plates were incubated overnight at 37° C and isolates then scored for growth (resistance) or no growth (sensitivity). Data consisting of a “1” for resistance or “0” for sensitivity for each isolate at each concentration of each antibiotic was then entered into a spread-sheet for statistical analysis.

The following table includes the antibiotics and concentrations used for isolates in analyses for all the study watersheds.

**Table C-1: Antibiotics and concentrations used for ARA.**

<u>Antibiotic</u>	<u>Concentration (µg/ml)</u>
Amoxicillin	0.625
Cephalothin	10, 15, 30, 50
Chloramphenicol	10
Chlortetracycline	60, 80, 100
Erythromycin	10
Gentamycin	5, 10, 15
Neomycin	40, 60, 80
Oxytetracycline	20, 40, 60, 80, 100
Salinomycin	10
Streptomycin	40, 60, 80, 100
Tetracycline	10, 30, 50, 100
Vancomycin	2.5

### KNOWN-SOURCE LIBRARY

**Construction and Use.** Fecal samples (scat) from known sources in each watershed were collected during the study period by MDE personnel and delivered to the BST Laboratory at SU. *Enterococcus* isolates were obtained from known sources (e.g., human, cow, goat, horse, dog, bear, beaver, deer, duck, fox, goose, heron, opossum, rabbit, raccoon, and squirrel). For each watershed, a library of patterns of *Enterococcus* isolate responses to the panel of antibiotics was analyzed using the statistical software CART<sup>®</sup> (Salford Systems, San Diego, CA).

*Enterococcus* isolate response patterns were also obtained from bacteria in water samples collected at the monitoring stations in each basin. Using statistical techniques, these patterns were then compared to those in the appropriate library to identify the probable source of each water isolate.

## STATISTICAL ANALYSIS

We applied a tree classification method, <sup>1</sup>CART<sup>®</sup>, to build a model that classifies isolates into source categories based on ARA data. CART<sup>®</sup> builds a classification tree by recursively splitting the library of isolates into two nodes. Each split is determined by the antibiotic variables (antibiotic resistance measured for a collection of antibiotics at varying concentrations). The first step in the tree-building process splits the library into two nodes by considering every binary split associated with every variable. The split is chosen that maximizes a specified index of homogeneity for isolate sources within each of the nodes. In subsequent steps, the same process is applied to each resulting node until a *stopping* criterion is satisfied. Nodes where an additional split would lead to only an insignificant increase in the *homogeneity index* relative to the *stopping* criterion are referred to as *terminal* nodes.<sup>2</sup> The collection of *terminal* nodes defines the classification model. Each *terminal* node is associated with one source, the source isolate with an unknown source), based that is most populous among the library isolates in the node. Each water sample isolate (*i.e.*, an on its antibiotic resistance pattern, is identified with one specific *terminal* node and is assigned the source of the majority of library isolates in that *terminal* node.<sup>3</sup>

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<sup>1</sup> The Elements of Statistical Learning: Data Mining, Inference, and Prediction. Hastie T, Tibshirani R, and Friedman J. Springer 2001.

<sup>2</sup> An ideal split, *i.e.*, a split that achieves the theoretical maximum for homogeneity, would produce two nodes each containing library isolates from only one source.

<sup>3</sup> The CART<sup>®</sup> tree-classification method we employed includes various features to ensure the development of an optimal classification model. For brevity in exposition, we have chosen not to present details of those features, but suggest the following sources: Breiman L, et al. *Classification and Regression Trees*. Pacific Grove: Wadsworth, 1984; and Steinberg D and Colla P. *CART—Classification and Regression Trees*. San Diego, CA: Salford Systems, 1997.

## ARA RESULTS

### Dividing Creek Watershed ARA Results

**Known-Source Library.** A 533 known-source isolate library was constructed from sources in the Dividing Creek Watershed. The number of unique antibiotic resistance patterns was calculated, and the known sources in the combined library were grouped into three categories: human, livestock (horse), pet (dog), and wildlife (beaver, deer, duck, fox, goose, raccoon) (Table C-2). There were no human sources collected. The library was analyzed for its ability to take a subset of the library isolates and correctly predict the identity of their host sources when they were treated as unknowns. Average rates of correct classification (ARCC) for the library were found by repeating this analysis using several probability cutoff points, as described above. The number-not-classified for each probability was determined. From these results, the percent unknown and percent correct classification (RCCs) was calculated (Table C-3).

**Table C-2: Category, total number, and number of unique patterns in the Dividing Creek known-source library.**

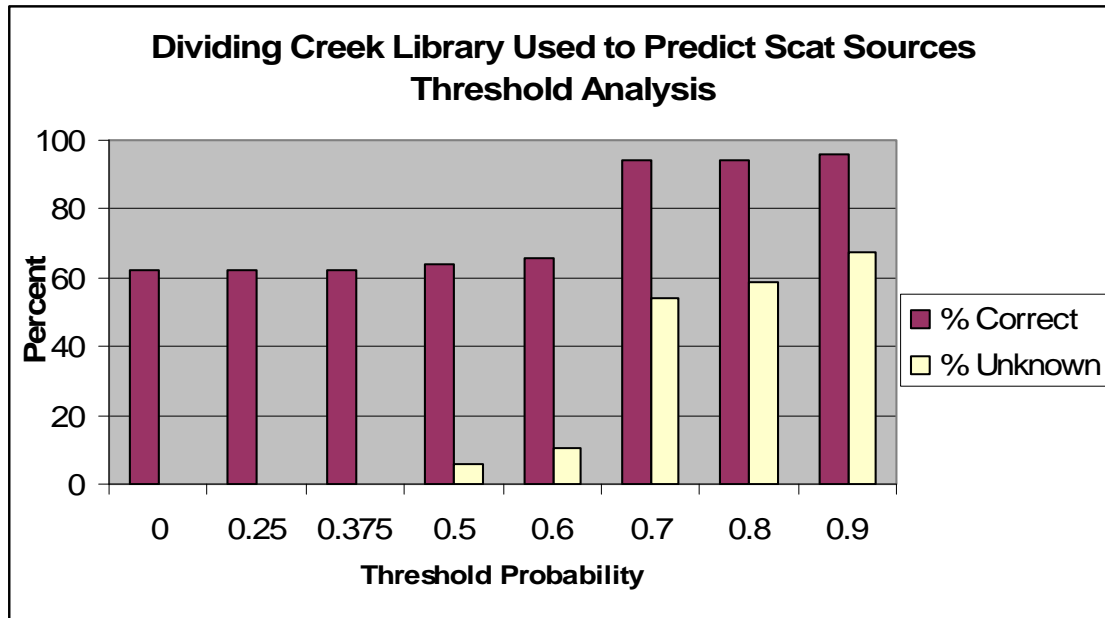
Category	Potential Sources	Total Isolates	Unique Patterns
human		0	0
livestock	horse	104	14
pet	dog	85	34
wildlife	beaver, deer, duck, fox, goose, raccoon	344	74
Total		533	122

For Dividing Creek Watershed, a cutoff probability of 0.70 (70%) was shown to yield an overall rate of correct classification of 94% (Table C-3). The resulting rates of correction classification (RCCs) for the four categories of sources in the Dividing Creek portion of the library are shown in Table C-4.

**Table C-3: Number of isolates not classified, percent unknown, and percent correct for eight (8) cutoff probabilities for Dividing Creek known-source isolates using the Dividing Creek known-source library.**

Threshold	0	0.25	0.375	0.5	0.6	<b>0.7</b>	0.8	0.9
% correct	62.3%	62.3%	62.3%	63.9%	65.7%	<b>93.9%</b>	94.1%	96.0%
% unknown	0.0%	0.0%	0.0%	5.8%	10.3%	<b>54.2%</b>	58.5%	67.4%
# not classified	0	0	0	31	55	<b>289</b>	312	359

**Figure C-1: Dividing Creek Classification Model: Percent Correct versus Percent Unknown using the Dividing Creek library.**



**Table C-4: Actual species categories versus predicted categories, at 70% probability cutoff, with rates of correct classification (RCC) for each category.**

Actual	Predicted					Total	RCC*
	Human	Livestock	Pet	Wildlife	Unknown		
Human	0	0	0	0	0	0	0.0%
Livestock	0	10	1	3	90	104	71.4%
Pet	0	0	62	1	22	85	98.4%
Wildlife	0	0	10	157	177	344	94.0%
Total	0	10	73	161	289	533	

\*RCC = Actual number of predicted species category / Total number predicted.

Example: 163 pet correctly predicted / 175 total number predicted for pet = 163/175 = 93%.

**Dividing Creek Water Samples.** Monthly monitoring from two (2) monitoring stations on Dividing Creek was the source of water samples. The maximum number of *Enterococcus* isolates per water sample was 24, although the number of isolates that actually grew was sometimes less than 24. A total of 442 *Enterococcus* isolates were analyzed by statistical analysis. The BST results by species category, shown in Table C-5, indicate that 94% of the

water isolates were able to be classified to a probable host source when using a 0.70 (70%) probability threshold.

**Table C-5: Probable host sources of water isolates by species category, number of isolates, and percent isolates classified at a cutoff probability of 70%.**

Source	Count	Percent	Percent Without Unknowns
human	0	0.0%	0.0%
livestock	9	2.0%	2.2%
pet	42	9.5%	10.1%
wildlife	363	82.1%	87.7%
unknown	28	6.3%	
Total	442	100.0%	100.0%

**% classified 93.7%**

\*Percentages may not add up to 100% due to rounding.

The seasonal distribution of water isolates from samples collected at each sampling station is shown below in Table C-6.

**Table C-6: *Enterococcus* isolates obtained from water collected during the spring, summer, fall, and winter seasons for Dividing Creek's two (2) monitoring stations.**

Station	Spring	Summer	Fall	Winter	Total
DIV0113	71	72	70	70	283
PUS0008	27	49	49	34	159
Total	98	121	119	104	442

## FINAL

Tables C-7 and C-8 on the following pages show the number and percent of the probable sources for each monitoring station by month.

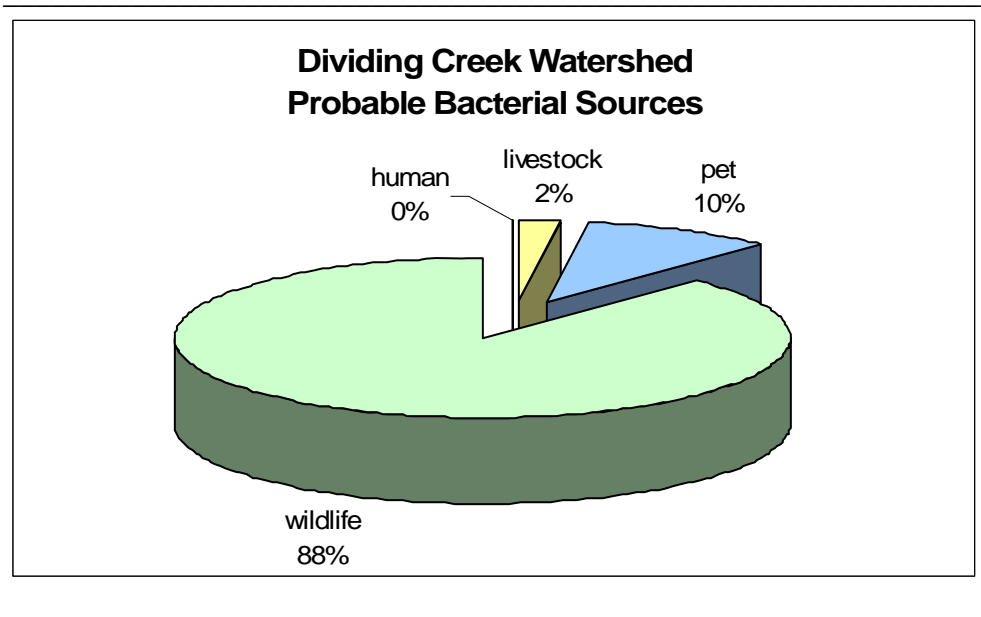
<b>Table C-7: BST Analysis: Number of Isolates per Station per Date.</b>							
Station	Date	Predicted Source					Total
		Human	Livestock	Pet	Wildlife	Unknown	
DIV0113	11/18/03	0	0	10	11	1	22
PUS0008	11/18/03	0	0	1	7	2	10
DIV0113	12/02/03	0	0	0	20	4	24
PUS0008	12/02/03	0	1	1	11	2	15
DIV0113	01/07/04	0	0	6	17	0	23
PUS0008	01/07/04	0	2	0	8	0	10
DIV0113	02/05/04	0	1	3	13	6	23
PUS0008	02/05/04	0	0	1	21	1	23
DIV0113	03/03/04	0	1	0	14	9	24
PUS0008	03/04/04	0	0	0	0	1	1
DIV0113	04/07/04	0	0	1	22	0	23
PUS0008	04/07/04	0	3	0	0	0	3
DIV0113	05/12/04	0	0	3	21	0	24
DIV0113	06/09/04	0	0	2	22	0	24
PUS0008	06/09/04	0	1	1	22	0	24
DIV0113	07/08/04	0	0	1	23	0	24
PUS0008	07/08/04	0	0	0	24	0	24
DIV0113	08/11/04	0	0	1	23	0	24
PUS0008	08/11/04	0	0	0	0	1	1
DIV0113	09/09/04	0	0	8	16	0	24
PUS0008	09/09/04	0	0	1	23	0	24
DIV0113	10/06/04	0	0	1	23	0	24
PUS0008	10/06/04	0	0	1	22	1	24
<b>Total</b>		<b>0</b>	<b>9</b>	<b>42</b>	<b>363</b>	<b>28</b>	<b>442</b>



**Table C-8: BST Analysis: Percent of Isolates per Station per Date.**

Station	Date	Predicted Source					Total
		Human	Livestock	Pet	Wildlife	Unknown	
DIV0113	11/18/03	0.0%	0.0%	45.5%	50.0%	4.5%	100.0%
PUS0008	11/18/03	0.0%	0.0%	10.0%	70.0%	20.0%	100.0%
DIV0113	12/02/03	0.0%	0.0%	0.0%	83.3%	16.7%	100.0%
PUS0008	12/02/03	0.0%	6.7%	6.7%	73.3%	13.3%	100.0%
DIV0113	01/07/04	0.0%	0.0%	26.1%	73.9%	0.0%	100.0%
PUS0008	01/07/04	0.0%	20.0%	0.0%	80.0%	0.0%	100.0%
DIV0113	02/05/04	0.0%	4.3%	13.0%	56.5%	26.1%	100.0%
PUS0008	02/05/04	0.0%	0.0%	4.3%	91.3%	4.3%	100.0%
DIV0113	03/03/04	0.0%	4.2%	0.0%	58.3%	37.5%	100.0%
PUS0008	03/04/04	0.0%	0.0%	0.0%	0.0%	100.0%	100.0%
DIV0113	04/07/04	0.0%	0.0%	4.3%	95.7%	0.0%	100.0%
PUS0008	04/07/04	0.0%	100.0%	0.0%	0.0%	0.0%	100.0%
DIV0113	05/12/04	0.0%	0.0%	12.5%	87.5%	0.0%	100.0%
DIV0113	06/09/04	0.0%	0.0%	8.3%	91.7%	0.0%	100.0%
PUS0008	06/09/04	0.0%	4.2%	4.2%	91.7%	0.0%	100.0%
DIV0113	07/08/04	0.0%	0.0%	4.2%	95.8%	0.0%	100.0%
PUS0008	07/08/04	0.0%	0.0%	0.0%	100.0%	0.0%	100.0%
DIV0113	08/11/04	0.0%	0.0%	4.2%	95.8%	0.0%	100.0%
PUS0008	08/11/04	0.0%	0.0%	0.0%	0.0%	100.0%	100.0%
DIV0113	09/09/04	0.0%	0.0%	33.3%	66.7%	0.0%	100.0%
PUS0008	09/09/04	0.0%	0.0%	4.2%	95.8%	0.0%	100.0%
DIV0113	10/06/04	0.0%	0.0%	4.2%	95.8%	0.0%	100.0%
PUS0008	10/06/04	0.0%	0.0%	4.2%	91.7%	4.2%	100.0%
<b>Total</b>		<b>0.0%</b>	<b>2.0%</b>	<b>9.5%</b>	<b>82.1%</b>	<b>6.3%</b>	<b>100.0%</b>

**Figure C-2: Dividing Creek Watershed relative contributions by probable sources of *Enterococcus* contamination.**



## SUMMARY

### Dividing Creek Summary

The use of ARA was successful for identification of probable bacterial sources in the Dividing Creek Watershed. When water isolates were compared to the library and potential sources predicted, 94% of the isolates were classified as to category by statistical analysis. The highest RCC for the library was 98% (for pet), with 94% (for wildlife) the second highest. Livestock had an RCC of 71%.

The largest category of potential sources in the watershed as a whole was wildlife (88% of classified water isolates), followed by pet (10%) and livestock (2%). The latter two contributed a relatively low percentage of probable bacterial contributions of 12% (Fig. 2-DIV).

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FINAL

## Appendix D – Estimating Maximum Daily Loads

This appendix documents the technical approach used to define maximum daily loads of fecal bacteria consistent with the annual average TMDL which, when met, are protective of water quality standards in the non-tidal portion of the Dividing Creek watershed. The approach builds upon the TMDL analysis that was conducted to ensure that compliance with the annual average target will result in compliance with the applicable water quality standards. The annual average loading target was converted into allowable *daily* values by using the loadings developed from the TMDL analysis. The approach is consistent with available EPA guidance on generating daily loads for TMDLs.

The available guidance for developing daily loads does not specify a single allowable approach; it contains a range of options. Selection of a specific method for translating a time-series of allowable loads into expression of a TMDL requires decisions regarding both the level of resolution (e.g., single daily load for all conditions vs. loads that vary with environmental conditions) and level of probability associated with the TMDL.

### Level of Resolution

The level of resolution pertains to the amount of detail used in specifying the maximum daily load. The draft EPA guidance on daily loads provides three categories of options for level of resolution.

1. **Representative daily load:** In this option, a single daily load (or multiple representative daily loads) is specified that covers all time periods and environmental conditions.
2. **Flow-variable daily load:** This option allows the maximum daily load to vary based upon the observed flow condition.
3. **Temporally-variable daily load:** This option allows the maximum daily load to vary based upon seasons or times of varying source or water body behavior.

### Probability Level

Essentially all TMDLs have some probability of being exceeded, with the specific probability being either explicitly specified or implicitly assumed. This level of probability reflects, directly or indirectly, two separate phenomena:

1. Water quality criteria consist of components describing acceptable magnitude, duration, and frequency. The frequency component addresses how often conditions can allowably surpass the combined magnitude and duration components.
2. Pollutant loads, especially from wet weather sources, typically exhibit a large degree of variability over time. It is rarely practical to specify a “never to be exceeded value” for a daily load, as essentially any loading value has some finite probability of being exceeded.

The draft daily load guidance states that the probability component of the maximum daily load should be “based on a representative statistical measure” that is dependent upon the specific TMDL and best professional judgment of the developers. This statistical measure represents

how often the maximum daily load is expected/allowed to be exceeded. The primary options for selecting this level of protection would be:

1. **The maximum daily load reflects some central tendency:** In this option, the maximum daily load is based upon the mean or median value of the range of loads expected to occur. The variability in the actual loads is not addressed.
2. **The maximum daily load reflects a level of protection implicitly provided by the selection of some “critical” period:** In this option, the maximum daily load is based upon the allowable load that is predicted to occur during some critical period examined during the analysis. The developer does not explicitly specify the probability of occurrence.
3. **The maximum daily load is a value that will be exceeded with a pre-defined probability:** In this option, a “reasonable” upper bound percentile is selected for the maximum daily load based upon a characterization of the variability of daily loads. For example, selection of the 95<sup>th</sup> percentile value would result in a maximum daily load that would be exceeded 5% of the time.

***Selected Approach for Defining Maximum Daily Loads for Nonpoint Sources and MS4***

To calculate the non-tidal Dividing Creek basin MDL for nonpoint sources (there are no MS4s in the watershed), a “representative daily load” option was selected as the level of resolution, and a value “that will be exceeded with a pre-defined probability” was selected as the level of protection. In these options, the maximum daily load is one single daily load that covers the two flow strata, with an upper bound percentile that accounts for the variability of daily loads. The upper bound percentile and the maximum daily loads were estimated following EPA’s “Technical Support Document for Water Quality-Based Toxics Control” (1991 TSD) (EPA 1991); and “Approaches For Developing a Daily Load Expression for TMDLs Computed for Longer Term Averages” (EPA 2006).

The 1991 TSD illustrates a way to identify a target maximum daily concentration from a long-term average concentration (LTA) based on a coefficient of variation (CV) and the assumption of a log-normal distribution of the data. The equations for determining both the upper boundary percentile and corresponding maximum daily load described in the TSD are as follows:

$$MDLC = LTA * e^{[Z\sigma - 0.5\sigma^2]} \tag{D1}$$

and,

$$MDL = MDLC * Q * F \tag{D2}$$

where,

- MDLC = maximum daily load concentration (MPN/100ml)
- LTAC = long-term average TMDL concentration (MPN/100ml)
- MDL = Maximum Daily Load (MPN/day)

- Z = z-score associated with upper bound percentile (unitless)
- $\sigma^2$  =  $\ln(\text{CV}^2 + 1)$
- CV = coefficient of variation
- Q = flow (cfs)
- F = conversion factor

The first step is to use the bacteria monitoring data to estimate the upper bound percentile as the percentile of the highest observed bacteria concentration in each of the two monitoring stations of the non-tidal Dividing Creek basin. Using the maximum value of *E. coli* observed in each monitoring station, and solving for the z-score using the above formula, the value of “z” and its corresponding percentile is found as shown below. The percentile associated with the particular value of z can be found in tables in statistics books or using the function NORMSINV(%) in EXCEL<sup>®</sup>.

$$Z = [\log_{10}(\text{MOC}) - \log(\text{AM}) + 0.5\sigma^2]/\sigma \quad (\text{D3})$$

where,

- Z = z-score associated with upper bound percentile
- MOC = maximum observed bacteria concentration (MPN/100ml)
- AM = arithmetic mean observed bacteria concentrations (MPN/100ml)
- $\sigma^2$  =  $\ln(\text{CV}^2 + 1)$
- CV = coefficient of variation (arithmetic)

Note that these equations use arithmetic parameters, not geometric parameters as used in the calculations of the long-term annual average TMDL. Therefore, bias correction factors are not necessary to estimate the loads as will be explained below.

The highest percentile of all the stations analyzed by stratum will define the upper bound percentile to be used in estimating the maximum daily limits. In the case of non-tidal Dividing Creek basin, a value measured during low flow conditions at the PUS0008 station resulted in the highest percentile of both strata of the two stations. This value translates to the 98.0<sup>th</sup> percentile, which is the upper boundary percentile to be used in the computation of the maximum daily limits (MDLs) throughout this analysis. Results of the analysis to estimate the recurrence or upper boundary percentile are shown in Table D-1.

**Table D-1: Percentiles of Maximum Observed Bacteria Concentrations**

<b>Station / Tributary</b>	<b>Flow Stratum</b>	<b>Maximum Observed <i>E. coli</i> Concentration (MPN/100ml)</b>	<b>Percentile (%)</b>
<b>DIV0113</b> Dividing Creek	<b>High</b>	364	83.5
	<b>Low</b>	2,005	94.4
<b>PUS0008</b> Pusey Branch	<b>High</b>	113	84.5
	<b>Low</b>	2,005	98.0

The 98.0<sup>th</sup> percentile value results in a maximum daily load that would not be exceeded 98.0% of the time, as, in a similar manner, a TMDL that represents the long-term average condition would be expected to be exceeded half the time even after all required controls were implemented.

The MDLCs are estimated based on a statistical methodology referred to as “Statistical Theory of Rollback (STR)”. This method predicts concentrations of a pollutant after its sources have been controlled (post-control concentrations), in this case after annual average TMDL implementation. Using STR, the daily TMDLs are calculated as presented below.

First, the long-term average TMDL concentrations ( $C_{LTA}$ ) by stratum are estimated by applying the required percent reduction to the baseline (monitoring data) concentrations ( $C_b$ ) by stratum as follows:

From Section 4.3, equations (8) and (9):

$$L_b = L_{b-H} + L_{b-L}$$

$$L_b = Q_H * C_{bH} * F_{IH} * W_H + Q_L * C_{bL} * F_{IL} * W_L$$

And from equation (10):

$$\text{Annual Average TMDL} = L_b * (1 - R)$$

Therefore,

$$L_b * (1 - R) = Q_H * C_H * F_{IH} * W_H * (1 - R) + Q_L * C_L * F_{IL} * W_L * (1 - R) \quad (D4)$$

As explained before, a reduction in concentration is proportional to a reduction in load, thus the bacteria concentrations expected after reductions are applied are equal to the baseline concentrations multiplied by one minus the required reduction:



$$C_{LTA-H} = C_{b-H} * (1 - R_H) \quad (D5)$$

$$C_{LTA-L} = C_{b-L} * (1 - R_L) \quad (D6)$$

The TMDL concentrations estimated as explained above are shown in Table D-2.

**Table D-2: Long-term Annual Average (LTA) TMDL Bacteria Concentrations**

Station / Tributary	Flow Stratum	LTA Geometric Mean <i>E. coli</i> Concentration (MPN/100ml)	LTA Arithmetic Mean* <i>E. coli</i> Concentration (MPN/100ml)
<b>DIV0113</b> Dividing Creek	<b>High</b>	21	59
	<b>Low</b>	84	150
<b>PUS0008</b> Pusey Branch	<b>High</b>	30	44
	<b>Low</b>	58	183

\*Only arithmetic parameters are used in the daily loads analysis.

The next step is to calculate the 98.0<sup>th</sup> percentile (the MDL concentrations) of these expected concentrations (LTA concentrations) using the coefficient of variation of the baseline concentrations. Based on a general rule for coefficient of variations, the coefficient of variation of the distribution of pollutant concentrations does not change after these concentrations have been reduced or controlled by a fixed proportion (Ott 1995). Therefore, the coefficient of variation estimated using the monitoring data concentrations does not change, and it can be used to estimate the 98.0<sup>th</sup> percentile of the long-term average TMDL concentrations (LTAC) using equation (D1). These values are shown in Table D-3.

**Table D-3: Maximum Daily Load (MDL) Concentrations**

Station / Tributary	Flow Stratum	Coefficient of Variation	MDL <i>E. coli</i> Concentration (MPN/100ml)
<b>DIV0113</b> Dividing Creek	<b>High</b>	2.68	404
	<b>Low</b>	1.48	766
<b>PUS0008</b> Pusey Branch	<b>High</b>	1.06	180
	<b>Low</b>	3.01	1,302

With the 98.0<sup>th</sup> percentiles of LTA TMDL bacteria concentrations estimated for both high flow and low flow strata as explained above, the maximum daily load for nonpoint sources for each subwatershed can be now estimated as:

$$\text{Daily TMDL (MPN/day)} = Q_H * (98.0^{\text{th}} C_{LTA-H}) * F_{IH} * W_H + Q_L * (98.0^{\text{th}} C_{LTA-L}) * F_{IL} * W_L \quad (D7)$$

***Selected Approach for Defining Maximum Daily Loads for Other Point Sources***

The TMDL also considers contributions from other point sources (i.e., municipal and industrial WWTP) in watersheds that have NPDES permits with fecal bacteria limits. The TMDL analysis that defined the average annual TMDL holds each of these sources constant at their existing NPDES permit limit (daily or monthly) for the entire year. The approach used to determine maximum daily loads is dependent upon whether a maximum daily load is specified within the permit. If a maximum daily load is specified within the permit, then the maximum design flow is multiplied by the maximum daily limit to obtain a maximum daily load. If a maximum daily limit is not specified in the permit, then the maximum daily loads are calculated from guidance in the TSD for Water Quality-based Toxics Control (EPA 1991).

There are no NPDES-regulated point sources to consider in the non-tidal portion of the Dividing Creek watershed.

The Maximum Daily Loads for the non-tidal Dividing Creek basin are presented in Table D-4 below.

**Table D-4: Maximum Daily Loads Summary**

Station / Tributary	Flow Stratum	Maximum Daily Load (Billion <i>E. coli</i> MPN/day)	
		by Stratum	Weighted
DIV0113 Dividing Creek	High	610	285
	Low	149	
PUS0008 Pusey Branch	High	231	220
	Low	215	

**Maximum Daily Loads Allocations**

Using the MDLs estimated as explained above, loads are allocated following the same methodology as the annual average TMDL (See section 4.8). The maximum daily load allocations for the non-tidal Dividing Creek basin are presented in Table D-5.

**Table D-5: Maximum Daily Loads**

<b>Station / Tributary</b>	<b>Total Allocation</b>	<b>LA</b>	<b>SW-WLA</b>	<b>WWTP-WLA</b>
<b>(Billion MPN <i>E. coli</i> /day)</b>				
<b>DIV0113</b> Dividing Creek	285	285	N/A	N/A
<b>PUS0008</b> Pusey Branch	220	220	N/A	N/A
<b>Total</b>	<b>505</b>	<b>505</b>	<b>0</b>	<b>0</b>

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