# A Multi-Disciplinary Approach to Tracking the Downstream Impacts of Inadequate Sanitation in Central Appalachia

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ABSTRACT

Poor sanitation infrastructure in rural areas can often lead to high levels of fecal contamination in local waterbodies and subsequent exposure to waterborne disease can occur. Although standard water quality measures such as quantification of *E. coli* can reveal relative concentrations of fecal contamination, they do not pinpoint the sources of such contamination. Source assessment in rural areas affected by untreated household waste might be improved with the human-specific, microbial source tracking marker HF183. This study attempted to quantify HF183 in two particular Appalachia streams with known discharges of untreated household waste. Water samples were taken above and at multiple points below these discharges on 29 occasions between August 2012 and April 2016, and tested for both HF183 and *E. coli*. HF183 was detected consistently in one of the study streams, though the concentrations were generally much lower than those previously reported in raw sewage; in the other watershed, HF183 was never detected. Further analysis via a multiple linear regression model showed a positive correlation between the level of *E. coli* and the proximity and number of known waste discharge points upstream from each sampling site. Primary conclusions of this study include: 1) HF183 is not always detected, even in watersheds with known sources of human fecal contamination, 2) it may be a useful water quality assessment tool where such contamination is suspected, particularly in cases where contaminant source allocation is necessary for setting mitigation priorities.

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GENERAL AUDIENCE ABSTRACT

Poor wastewater infrastructure in rural areas can often lead to high levels of fecal contamination in local waterbodies and subsequent downstream exposure to waterborne disease can occur. The concentration of fecal contamination is currently measured in streams with microbial markers such as *E. coli*, however current methods do not pinpoint the sources of such contamination. Determining the source of contamination is important as it can improve the effectiveness of pollution mitigation strategies. Source assessment in rural areas affected by untreated household waste might be improved by testing water for a human-specific, genetic marker known as HF183. This study attempted to quantify HF183 in two particular Appalachia streams with known discharges of untreated household waste. Water samples were taken above and at multiple points below these discharges and tested for both HF183 and *E. coli*. HF183 was detected consistently in one of the study streams, though the concentrations were relatively low; in the other watershed, HF183 was never detected. Further statistical analysis showed a positive correlation between the level of *E. coli* and the known waste discharge points upstream from each sampling site. Primary conclusions of this study include: 1) HF183 is not always detected, even in watersheds with known sources of human fecal contamination, 2) HF183 may be a useful water quality assessment tool where such contamination is already suspected in order to justify setting mitigation priorities.

To Mike

“Everyone has a plan until they get punched in the face” – Mike Tyson

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## 1. Introduction: Importance of surface water quality

Despite major advancements in wastewater treatment in the 20th century, inadequate sanitation services remain an issue for some communities in the United States, particularly in rural areas. Although the World Health Organization, United Nations Children's Fund and the World Bank state that 100% of US households have adequate plumbing, 1.7 million (0.64% of the total population) Americans still are lacking of basic water and sanitation services (1). The perception of overall wealth in the US may blind the public from the realities that exist in many rural communities (2). According to a recent analysis of available US Census Data (1), rural communities are two times as likely to suffer from improper sanitation services as urban communities. Infrastructure for sanitation services is particularly lacking in remote communities (i.e., less than 1000 residents) where there are less users to cover infrastructure costs and technical assistance is not readily available locally (1, 3). This can result in inadequate sanitation practices (e.g. soakaways, straight pipes) that directs untreated household waste to local surface waters.

Human waste can be a major contributor to pathogen contamination of surface water (4). In areas with low population density, municipal collection and treatment of wastewater is not practical, and so wastewater management is generally handled on a per household basis. Onsite wastewater treatment is most commonly performed by a septic system, where treatment consists of sedimentation in a septic tank followed by biological degradation by soils in a drainfield. If not maintained, system failure can result in the discharge of poorly treated or untreated effluent that may still contain hazardous pathogens (5, 6) to groundwater or surface water. Under Virginia law, it is the owner's responsibility for operation and maintenance of private sewage systems (7), but some residents may be unable to do this – and municipalities are generally unable to finance private systems (2). In some rural areas where septics are impractical due to poor soils or lack of space, waste can be directly discharged into local waterways via "straight piping" (8, 9).

Improper sanitation of household wastewater has been long recognized as a public health hazard. In a recent national survey of health care providers in rural communities, surface water pollution was identified as a primary environmental health concern (10). Relative to less developed countries, the incidence of waterborne diseases such as hepatitis A, salmonellosis, and typhoid,

are not common, however risks are greater in areas without proper water and sanitation (1). Waste entering common waterways used for recreation or drinking water can be a point of human exposure and cause infection (11). Denno et. al. (2009) found that the use of water from a private supply in Washington was a risk factor for reportable enteric infections in children (12). DeFelice et. al. (2015) found that around 0.8% of the population in North Carolina was exposed to total coliform bacteria through private well water (13). Characterizing the burden of disease attributable to these issues is of considerable interest in prioritizing and justifying infrastructure improvements, as waterborne disease illnesses have multiple adverse economic effects including the cost of hospitalizations, losses in productivity at places of employment, and absences from school (1). Eliminating these sources of pathogen contamination could therefore result in significant public health and economic benefits.

Standard water quality assessments generally only include evaluation of total coliforms or *E. coli*. These bacteria are common to all warm-blooded animals, and so provide limited information regarding specific sources. Fecal contamination by human sources is generally considered a greater health concern than contamination by other animals since many waterborne pathogens (viruses in particular) exhibit host specificity with humans (14–16). The inclusion of library-independent, established source-specific markers in monitoring efforts can greatly aid in watershed assessment. Understanding the origin of fecal contamination can improve understanding of potential health risks and inform decisions about appropriate remediation actions to meet water quality goals (15, 17). The genetic marker HF183 from *Bacteroides* spp. has been well established as strongly indicative of the presence of human fecal contamination (14, 18). However to date, the majority of studies have only documented HF183 detection downstream from relatively large centralized wastewater treatment facilities (14). The usefulness of this type of monitoring strategy in rural watersheds is not well documented. The subsequent literature review (Chapter 2) discusses the most effective methods to detect human contamination in environmental waters, focusing on applications in rural areas with improper sanitation. More specifically, this review analyzes their usefulness in Appalachia in detecting and quantifying health risks from human contamination.

## 2. Literature Review

### *2.1 Fecal Indicator Bacteria*

Fecal indicator bacteria (FIB) originate from the gut of warm blooded animals, as do enteric pathogens. Although generally not harmful themselves, FIB may indicate pathogen presence when found in the environment. FIB are used as indicators of potential pathogen presence since monitoring for the entire suite of pathogenic bacteria, viruses, and protozoa potentially present in a water body is impractical in terms of time and costs (14, 19, 20). There are many different types of FIB, and each has different predictive strengths and weaknesses that inform how it can be best applied to sanitary surveys of water.

The most common fecal indicator organisms used to monitor waters in the United States are coliforms, *Escherichia coli* (*E. coli*), and enterococci (21). Regulatory limits for freshwater and saltwater microbial indicators as recommended by the USEPA are primarily based on epidemiological studies which drew relationships between concentrations of specific indicators observed in recreational waters in New York, Massachusetts, Louisiana, Pennsylvania and Oklahoma and the occurrence of gastroenteritis in swimmers. In these studies, relationships between exposure and illness were relatively weak for total and fecal coliforms, but strong for *E. coli* and enterococci. These bacteria are therefore recommended for use in state regulations by the USEPA in freshwater and saltwater, respectively (21, 22).

For FIB to correlate with health hazards, it is important to understand how they behave in different environments in comparison to the pathogens they represent. Factors such as UV light, temperature, competition, and predation affect the survival rates of FIB in the environment, and may affect pathogens present in the immediate area differently (23, 24). FIB do not necessarily always co-occur in the environment with viruses and protozoa (25). This may in part be explained by a number of different physical, chemical and biological factors such as the organism size, density, charge, and resistance to UV radiation (26). Several studies have demonstrated that FIB can survive for varying periods of time in soil, may experience regrowth, then be suspended into the water column (27, 28). Lee et. al. (2006) studied FIB on beaches near Santa Monica and found FIB were able to grow in beach sediments through microcosm

experiments, and also observed higher levels of FIB in environmental waters after storm events (29). Anderson et. al. (2005) studied the differences in persistence of FIB in soil inocula and wastewater, and found differences in decay rates between fecal coliforms (90% identified as *E. coli*) and enterococci (30).

Although fecal indicator organisms provide useful information on microbial water quality, continual monitoring requires extensive water quality sampling. Linear regression is a technique that uses hydrologic, physiochemical and land use variables and can be used to predict the level of fecal contamination in rivers (31). Hampson et. al. (2010) used a linear regression model to predict the levels of fecal indicator organisms in the UK after implementation of pollution reduction strategies on dairy operations to determine the effectiveness of different remediation strategies. This statistical analysis may provide a link between upstream fecal contamination loadings with downstream measures of FIB concentration. This information can be useful in determining health hazards for specific areas.

### *2.2 Source Tracking*

Impaired waters frequently include fecal contamination from multiple sources (e.g. wildlife, domestic animals, humans). Multiple studies suggest contamination by human sources is of greatest public health concern given the host-specificity of many waterborne pathogens (14–16). Different health hazards from different sources may explain why some studies continue to find only weak correlations between community health outcomes and FIB exposure (14). When identification of the contamination source is critical, biological, physical or chemical techniques, collectively referred to as “source tracking”, may be used to track and discriminate between sources (15). If successful, source tracking can be used to effectively target and remove specific sources of fecal contamination, and has shown potential for evaluating placement of treatment systems in TMDL plans (32, 33). Though useful, the appropriateness of particular markers depends heavily on site specific factors, and considerable debate remains regarding markers that might be universally appropriate (15).

### *2.2.1 Chemical Source Tracking*

Chemical source tracking involves the recovery of chemical compounds that do not come from the natural environment; these markers thus can provide high source specificity (i.e., the rate of true positives to all positive results). Common targets include chemicals that are solely anthropogenic, such as pharmaceuticals and personal care products, caffeine, and optical brighteners (33), as well as fecal sterols or stanols. Pharmaceuticals, which pass through the body and into sewage following ingestion, are expected to have a large geographical range given their broad accessibility. Source tracking targets include common drugs such as ibuprofen and β-blockers, as well as common chemicals in household products (e.g. in mouthwash, toothpaste, deodorant, soaps and air fresheners) (15). In a study by Buser et. al. (1999), ibuprofen was consistently found in wastewater treatment influent, and when compared with other pharmaceuticals (i.e., clofibric acid and diclofenac), was more efficiently degraded during wastewater treatment (34). Degradation during treatment makes the marker more indicative of untreated waste if found in the environment, e.g. the presence of caffeine, a biodegradable compound, is a useful indicator of untreated wastewater (33, 35). Caffeine is found in drinks such as coffee, tea, soda and energy drinks, and the average American consumes 210 mg/day of caffeine (36); therefore caffeine is geographically widely dispersed. Many different pharmeceuticals have been successfully detected in sewage, however caffeine has shown detectability in environmental waters (37, 38). In a screening for 24 different chemicals in wastewater and environmental waters in northwest Washington, 16 compounds were detected in the effluent while only caffeine, metformin and nicotine were detected in the environmental samples, at much lower levels than in wastewater (38).

The primary disadvantages to targeting chemical compunds as a primary source tracking strategy are the generally intricate and expensive analytical procedures involved. For example, testing for pharmaceuticals and caffeine requires liquid or gas chromatography and sensitive detection techniques, which requires specialized equipment and experienced analysts (15). Optical brighteners have been proposed as an alternative target as they are comparatively cheap and simple to detect. These chemicals are found in home products such as detergents, toilet paper and dishwashing soap, and are discharged into the environment along with waste. Optical brighteners

have a strong transmittance of blue light (39) and the fluorenscense can be detected via multiple means including by flurometer. Flurometers can be hand-held devices with the ability to operate in a continuous flow mode. Although targeting optical brighteners is cost effective and very simple to test for, they are limited by a poor level of detection, and interference from organic substances (15). A study in southern California tested optical brighteners from a variety of detergents, sewage and septic samples, and although there were no false positives (i.e., results were highly specific), the method missed many samples with lower concentrations of human sewage. That study concluded that detection of optical brighteners can provide rapid, low-cost, preliminary field results, but are not suitable for surveys requiring a high level of accuracy or low detection limits (39). Detection limits is a common issue with chemical markers, as they are often in the environment in very low, yet still hazardous concentrations (15).

Field and Samadpour (2007) state that the survival and relationship of indicators and pathogens may be complicated by differences in persistence and degradation patterns. Survival of pathogens may be based on settling rate, UV radiation and predation. These factors will affect chemical indicators much differently, as they are not live cells (40). Therefore even if detected downstream from sources, the physical and biological environment may need to be taken into account to determine potential for pathogen presence. Markers within microbial hosts are more physically similar to pathogens than chemical markers, therefore it is thought that they will likely better reflect downstream survival/decay patterns.

### 2.2.2. *Microbial Source Tracking*

Microbial source tracking is based on the concept that certain microbial species and/or genetic sequences are specific to different animal hosts. If these markers are recovered in the environment, their origin can therefore be identified. Microbial source tracking is generally categorized into library-dependent and library-independent methods. Library dependent methods identify host-associated microorganisms by comparing metabolic characteristics to a regional library of behavior while library independent methods rely on detecting genetic markers specific to a targeted host (14, 41).

*Library-Dependent Microbial Source Tracking*

Library dependent methods (LDM) rely on a dataset of source profiles (i.e., library) established from samples of known fecal origin. Libraries consist of characteristics in host-associated microorganisms (e.g. *E. coli*, enterococci, and total coliforms) such as antibiotic resistance, carbon source utilization, genetic fingerprinting, or ribotyping (14). After libraries are formed, statistical analyses can be performed to compare unknown samples to the known source profiles to identify a most likely source. LDM therefore requires the establishment of large libraries that are not generally geographically stable; the amount of isolates needed to form a successful library depends on the method and organism used and can be both time-consuming and expensive (15). Wiggins et. al. (2003) found that in antibiotic resistance analysis, larger libraries tend to provide more accurate source identification, i.e. over 2,300 samples may be required to consider a library sufficiently representative. Since fecal isolate profiles between different watersheds vary, establishing libraries can be quite complicated (42). In addition, as microbial communities constantly evolve, libraries may not be temporally stable (18, 43). Given the effort to establish these libraries, which may only be applicable to certain regions, library-independent methods are increasingly popular and more widely applied for tracking human fecal contamination.

*Library-Independent Microbial Source Tracking*

Library-independent analyses focus on specific genetic markers in host-associated microorganisms. Ideally, these markers do not vary over a large geographical range. There are many different types of markers that identify different host-associated organisms, and each has a different analytical procedure (14). In 2003, the Southern California Coastal Water Research Project (SCCWRP) and the USEPA sponsored a multi-laboratory source tracking study, where 27 different laboratories were charged to determine the source of blinded fecal samples representing human, cow, deer, pig, chicken, pigeon, gull, horse and goose sources (40). The best performing markers (i.e., highest sensitivity and specificity) were HF183, BacH, HumM2, nifH and polyomaviruses. These markers were recovered directly from human feces as well as from raw sewage. Although this study provided a comprehensive analysis of different genetic markers,

it did not test recoverability of these markers from field samples. Given sufficient recovery and reliability in field samples, microbial methods have potential for use in practice in detecting human sources of contamination. This may prove particularly useful in areas with poor sanitation attempting to track the influence of untreated wastewater discharges and/or prioritize infrastructure upgrades.

*2.3 Poverty and Sanitation in Central Appalachia, and Applicability of Source Tracking*

The Appalachian region is defined by the Appalachian Regional Commission (ARC) as a 205,000 square mile region that extends from New York to Mississippi, following the spine of the Appalachian mountains (44). Central Appalachia is a region that includes parts of Kentucky, Maryland, Ohio, Pennsylvania, Tennessee, and Virginia, and all of West Virginia. This region is often characterized by its significant coal and, more recently, natural gas production (45). Although the land is rich in resources, socioeconomic depression is another defining characteristic. In 2009, per capita income in Appalachia overall was 25% lower than the national average; and in Central Appalachia, it was 32% lower (46). Given the lack of economic resources at the state or local levels, infrastructure such as roads, municipal drinking water, and sanitation services is sometimes lacking, and there may be insufficient technical expertise or resources to sufficiently address these problems (47).

With regard to the particular issue of sanitation, on-site wastewater treatment is commonly present as septic tanks; however, karst soils in Central Appalachia are generally inadequate for treatment. Furthermore, in the narrow Appalachian "hollows", residences are often located too close to streams for the construction of proper drain fields (45). Other on-site solutions, such as package treatment plants or sand filters have seen limited use, but are known to be precluded by local challenges in numerous cases (e.g., the necessity of shared responsibility for installation and maintenance in already resource-stressed communities) (3).

When other opportunities are not readily available, untreated household waste may simply be "straight piped" into nearby surface waters. Although the number of straight pipes in the entire region is unknown, the practice is not uncommon, e.g. in Letcher County, KY there are an

estimated 3,000 straight pipes serving 12,000 of the county's 30,000 residents (8). These types of problems undoubtedly contribute to fecal contamination in many Appalachian water bodies (45).

*E. coli* is commonly used to identify impaired waterways in the Appalachian region that require remediation, although the detection of *E. coli* does not reveal the source of contamination, and in and of itself suggest the most effective means to improve water quality and protect public health. Ideally, source tracking techniques could be used to track the persistence of this contamination beyond these small communities to motivate wider investment; however, available source tracking markers have not been as widely applied in rural areas, and there is no consensus on which genetic markers may have the most potential for use. Markers with low sensitivity or distribution in the human population may be hard to detect in rural environments where wastewater discharges are contributing from individual or small community residences rather than large municipalities. Tambalo et. al. (2012) detected low levels of BacH (also found in *Bacteroides*) in stream samples near homes without municipal wastewater treatment in the rural Canadian watershed of Qu'Appelle Valley (48). BacH was also successfully found in a river below a wastewater treatment plant in Eastern Austria (49). However Ridley et. al. (2014) could not recover BacH from streams samples in an agricultural watershed in Nova Scotia, Canada, with known water quality influences from on-site water treatment. The study concluded that BacH did not have a low enough level of detection for assessment of faulty septic systems (50). The nifH gene sequence from *Methanobacter Smithii*, was found by Ufnar et. al. (2006) in 100% of water samples downstream from leaking sewers in Gulfport, Mississippi. However *M. smithii* is only found in one third of individual fecal samples (51). Sidhu et. al. (2013) was only able to detect the nifH marker in 56% of samples from urban runoff with failing sewage infrastructure in Melbourne, Australia (29). Similarly, polyomaviruses have shown very high specificity to humans, however are in low abundance in human feces and are difficult to detect (14, 18, 52). Given that these markers are not consistently recovered in even more high density, urban waters, their likely utility in detecting more dilute wastewater in rural areas is low.

Comparatively, the HF183 marker shows potential for application in lower population density areas. Many studies report high sensitivity and specificity by HF183 in the detection of human wastewater influence (14, 18, 33, 52, 53). A study in Melbourne, Australia detected HF183 in

91% of the urban runoff samples (33). Seurinck was able to find HF183 in freshwater samples with a detection efficiency estimated between 78% and 91%, downstream from a wastewater treatment plant in Coupure, Gent, Belgium (53). Although these detections were successful, there is evidence that the amount of HF183 may vary per individual, and is more likely to be found where more individuals are contributing to the contamination (54). Therefore while the marker may be consistently found from the effluent of large wastewater facilities (14, 33, 53), the evidence linking HF183 and contamination from septic system discharges and straight pipes in rural areas is very limited. It is also unknown how the density of the marker relates to human health risk. In the aforementioned study by Tambalo et. al., as well as in a study by Kapoor et. al. (2013) located in an urban creek in Cincinnati, OH affected by combined sewage overflows, only a weak correlation was able to be drawn between human associated *Bacteroides* and *E. coli* (48, 55). More research needs to be done to better understand the relationship between FIB, source tracking markers, and health risks (16, 52), particularly in rural areas.

Of the many source tracking markers described, the microbial marker HF183 is most applicable in the Appalachian region. Although chemical markers may be highly specific and geographically disperse, they are expensive and would likely not follow decay patterns of organisms. The quick, cost effective methods of optical brighteners may prove useful in economically depressed areas, however in rural areas with disperse residents contributing waste to local waterways, this method may not have a low enough limit of detection. HF183 is the most appropriate source tracking marker, however it is not yet understood if it can be detected in an environment with a low population density, and after quantification whether HF183 could be related to levels of *E. coli* and health risks.

*2.4 Study Objectives*

**Infectious diseases in areas with poor wastewater treatment can be prevented, although justification of these investments may require demonstration of the degree of contamination associated with seemingly isolated community problems.** Justification of investment in sanitation infrastructure is particularly critical in areas with few financial resources. This study aims to directly link degraded water quality with the prevalence of straight pipes in Central Appalachian watersheds with known sanitation challenges through:

1. Development of a spatial statistical strategy to link upstream influences to *E. coli* levels longitudinally in Appalachian streams;

2. Detection of the currently most popular human source tracking marker (HF183) in environmental waters polluted by untreated household waste from rural communities; and

3. Assessment of the co-occurrence of HF183 with *E. coli*, the current state regulatory target used in identifying surface water impairments.

## 3. Methods

### 3.1 *Site Description*

The study area consists of two, 12-digit hydrological unit code (HUC) watersheds on the border of Kentucky and Virginia both of which are tributaries to the Powell River basin in Virginia: Callahan Creek and Roaring Fork (Figure 3.1). The watersheds have areas between 25 and 30 square miles, and are characterized by steep mountainous slopes and karst geology mainly composed of Mississippian (360-320 million years) and Pennsylvanian (320-296 million years) aged limestone, with deeper formations of shale, coal and sandstone (56). From analysis of the land cover dataset, both watersheds consists mainly of hardwood deciduous forest (66%) and surface mining activities (17%). Development and pasture land only account for 2% and 0.5% of land cover respectively. Development in both watersheds is mainly concentrated adjacent to the streams in the flatter, more habitable areas known as mountain "hollows" (45, 57). These watersheds were selected following input by the local Departments of Health; Environmental Quality; Mining, Minerals and Energy; and also local consultants with detailed knowledge of discharges of untreated household waste (58).

Each watershed contains fourteen in-stream sampling sites. The sites were selected directly above and below residential areas or any known discharges of fecal contamination, as well as at the top of the watershed and watershed outlet (58). At one particular site in Callahan Creek, samples were taken directly from the combined sewage effluent of multiple residences.

### 3.2 *Sampling Regimen*

Water samples were collected monthly from August 2012 until August 2014 and from January 2016 to April 2016. In all months and at all sites, samples were tested for *E. coli* (n = 28), and between April 2014 and August 2014 and January 2016 to April 2016 an additional sample aliquot was preserved for HF183 (n = 9) from 5 sites in each watershed. Temperature, dissolved oxygen, pH and specific conductivity were measured on site via an YSI Quattro Pro Plus (YSI, Yellow Springs, OH; USA). Samples for microbiological analysis were collected at the thalweg in 250 mL sterilized bottles, and transported on ice back to the Biological Systems Engineering Seitz Hall laboratory at Virginia Tech for prompt analysis (average transport time of 2-3 hours).

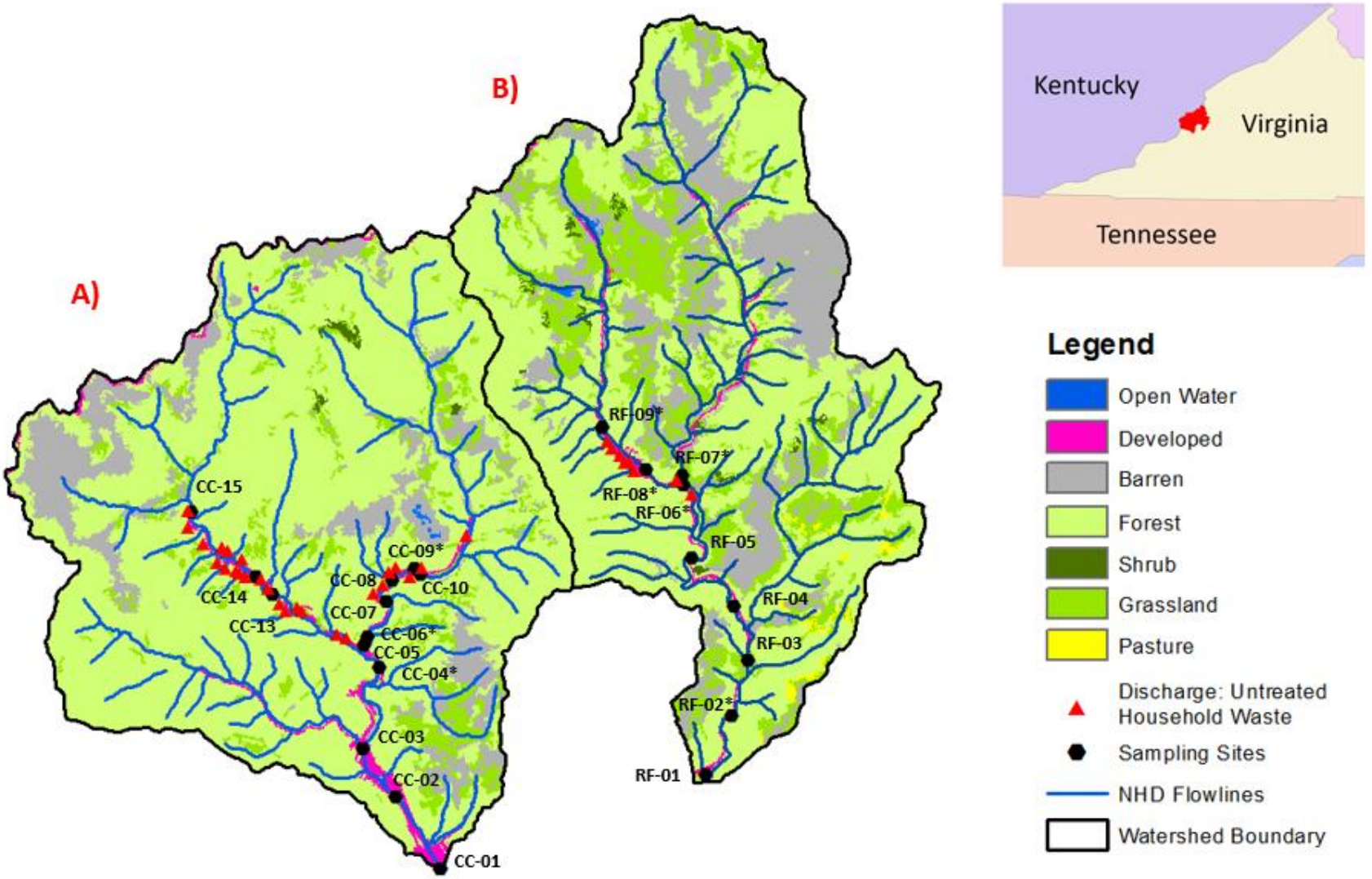


Figure 3.1 Sampling sites and known discharges for 12-digit HUC watersheds Callahan Creek (A) and Roaring Fork (B)
*Indicates the site was tested for both HF183 and E. coli

### 3.3 *Statistical Methods*

All statistical analyses were conducted in JMP 12 (SAS, Cary, NC, USA). Normality/non-normality of each dataset was determined via observation of the normal quantile plot prior to analysis.

#### 3.3.1 *Correlations between markers*

The Spearman's rank correlation test was used to determine whether a correlation exists between detected levels of *E. coli* and the HF183 marker. This is a nonparametric test, in which the points in each dataset are ranked from the highest value to the lowest, and then the difference between ranks is squared (Equation 1):

$$1 - \frac{6\Sigma D^2}{n(n^2-1)} \quad (1)$$

where $D$ is the difference in ranks between the datasets and $n$ is the number of values in each dataset. This method returns a value between -1 and 1 assessing the overall correlation between the two observed variables (Spearmen's rho), and a p-value indicating significance.

#### 3.3.2 *Geospatial statistical model*

A statistical mixed-effects model was developed to determine if levels of *E. coli* were significantly related to the number of waste discharges upstream of each site, the distance of known waste discharges upstream from each site, and the season (winter/"high flow" vs not-winter/"low flow"). The model is a multidimensional regression analysis with fixed variables (i.e., season and waste discharges), and random variables (i.e., site location and experimental error). P-values are used as a measure of significance for individual variables. An $R^2$ value measures the overall fit of the model to the existing data.

$$\mu_{ij} = \beta_0 + \beta_1 S + \beta_2 D + \alpha_i + \varepsilon_{ij} \quad (2)$$

$$\varepsilon_{ij} \sim N(0, \sigma^2)$$

$$\alpha_i \sim N(0, \sigma^2{}_\alpha)$$

Each sampling site has a value corresponding to the sum of inverse distances to all upstream discharges of untreated household waste, *D*, as measured in ArcGIS with data from the National Hydrography dataset (59). Although the location of known waste discharges included both septic tanks and straight pipes, the effects from both sources were not differentiated as it is known that many of the septic systems in the area are not adequate or no longer function for proper waste treatment. The seasonal variable, *S*, was a binary variable indicating whether the season was winter (i.e., 1 for winter, 0 for not winter). The random variable, $\alpha$, accounts for dependence of the measurements taken within sites, while the different sampling sites are considered independent. The random error variable, $\varepsilon$, represents all other possible sources of random errors, e.g. whether residents were home and discharging waste when samples were collected. These random variables were normally distributed with a mean centered on 0. Each sampling site was also given a value for its distance along the stream from the watershed outlet, as measured in ArcGIS. All of the variables were modeled in the linear regression shown above via JMP (equation 2), where $\beta_n$ represents regression constants.

### 3.4 *Culture-based microbiological analyses*

Analysis for *E. coli* concentrations occurred immediately upon return to the laboratory via the Colilert defined substrate method using Quanti-Tray/2000 trays (IDEXX, Westbrook, ME, USA). Following 24 hours incubation at 37ºC, a most probable number of bacteria could be determined based on the number of positive wells (i.e., fluorescent under UV light). All samples were stored at 4ºC for the first 24 hours following collection and re-diluted if necessary to capture bacteria within appropriate detection limit.

3.5 *Molecular analyses*

A 100 mL aliquot from each sample was vacuum filtered through a Millipore 0.4 µm filter in duplicate (Merck KGaA, Darmstadt, Germany) to preserve for future molecular analyses. Filter effluent was discarded and filters were stored in cryotubes at -80º C before DNA extraction. Within six months, DNA from the filters was extracted using a PowerWater DNA Isolation Kit (MO BIO Laboratories, Carlsbad, CA, USA). In brief, filters were placed vertically in PowerBead Tubes (MO BIO Laboratories, Carlsbad, CA, USA) and shaken with a Mixer Mill MM 400 (Retsch, Haan, Germany) prior to the addition of kit reagents and isolation of DNA via repeated centrifugation steps. DNA concentrations were quantified with a Qubit 2.0 flurometer (Thermo Fisher Scientific, Waltham, MA, USA).

The *Bacteroides* HF183 genetic marker was quantified via qPCR with a Taqman probe, as described previously by Griffith et. al., 2013 (60) (Tables 3.1 and 3.2). All samples were amplified in triplicate by a Realplex Mastercycler (Eppendorf, Hamber, Germany) in a 96-well plate (Thermo Fisher Scientific, Waltham, MA, USA). Each well contained 20 µL of Master Mix (table 3.2) and 5 µL of sample. A no-template control was tested along with the samples to indicate any sources of contamination. The Mastercycler settings were as follows: 50º C for 2 minutes, 95º C for 10 minutes, then 40 cycles of 95º C for 15 seconds and 60º C for one minute. A 2439 base plasma containing the HF183 marker (60) at known amounts (between 10 and $10^6$ copies) was measured for the quantification of samples via the creation of a standard calibration curve. Results were only considered valid if the standard curve had an $R^2$ value above 0.99 and efficiency between 0.90 and 1.10. Each sample contained 1 µL of an internal amplification control to ensure that replication inhibition could be detected during qPCR (Integrated DNA Technologies, Corallville, Iowa, USA). The limit of detection (LOD) for HF183 was determined by scaling the lowest detectable amount of HF183 in tested samples to the total amount of eluted sample from DNA extraction. The same process was repeated with the lowest value on the standard curve (10 copies/mL) to determine the limit of quantification (LOQ).

Table 3.1. Primer Probe Mix Content for HF183 Analysis

| Item | Volume |
|---|---|
| Forward Primer (HF183) (Integrated DNA Technologies, Corallville, Iowa, USA) 5'- ATCATGAGTTCACATGTCCG -3' | 10 µL |
| Reverse Primer (BacR287) (Integrated DNA Technologies, Corallville, Iowa, USA) 5'- CTTCCTCTCAGAACCCCTATCC -3' | 10 µL |
| Ultrapure DEPC-Treated Water (Thermo Fisher Scientific, Waltham, MA, USA) | 572 µL |
| TaqMan probe (BacP234MGB) (Thermo Fisher Scientific, Waltham, MA, USA) [6FAM] - 5'- CTAATGGAACGCATCCC –MGB | 4 µL |
| TaqMan probe (Bac234IAC) (Thermo Fisher Scientific, Waltham, MA, USA) [VIC]-5'- AACACGCCGTTGCTACA –MGB | 4 µL |

Table 3.2. Master Mix Content for HF183 Analysis per Sample

| Item | Volume |
|---|---|
| Ultrapure DEPC-Treated Water (Thermo Fisher Scientific, Waltham, MA, USA) | 4 µL |
| Plasmid with IAC target (60) | 1 µL |
| SsoAdvanced Universal Probes Supermix (Bio Rad, Marnes-la-Coquette, France) | 12.5 µL |
| Bovine Serum Albumin (Thermo Fisher Scientific, Waltham, MA, USA) | 2.5 µL |
| Primer Probe Mix | 3 µL |

## 4. Results and Discussion

### *4.3 Association of Pollutant Levels with Untreated Household Waste*

There is a significant, positive relationship ($p < 0.05$) between the sum of the inverse distances to all upstream sources of untreated household waste, and the level of *E. coli* for both Callahan Creek and Roaring Fork. There is also a significant, positive relationship between the levels of *E. coli* and seasonality for Callahan Creek, however this relationship was not significant for Roaring Fork. Water temperature and stream flow in the Powell River were tested in both models but do not show a significant relationship with the levels of *E. coli*. Results from the statistical models for Callahan Creek and Roaring Fork are found in table 4.1. The coefficient for each variable represents the magnitude and direction of the relationship between the given explanatory variables and the response variable (i.e., *E. coli* levels, log transformed). A similar analysis with HF183 as the dependent variable could not be completed due to insufficient data and a high level of zeros (e.g. not recoverable).

Table 4.1. Callahan Creek and Roaring Fork Regression Model Coefficients

| Variable | Callahan Creek Coefficient | Callahan Creek P Value | Roaring Fork Coefficient | Roaring Fork P Value |
|---|---|---|---|---|
| Intercept | 2.69 | < 0.0001 | 0.89 | < 0.0001 |
| Winter (N)[1] | 0.84 | < 0.0001 | N/A | > 0.05 |
| Waste Discharge Value[2] | 0.08 | < 0.0001 | 0.07 | < 0.0001 |

[1]Categorical variable indicates whether the season is not winter: N is true, Y is false

[2]Variable represents the sum of the inverse distances to all known upstream sources of untreated household waste

The statistical model successfully links the discharges of untreated household waste and seasonal changes with the high levels of fecal contamination. The significant waste discharge variable indicates that with more discharges upstream from the sampling site, the level of fecal contamination is likely to be higher. A similar trend is seen for HF183 (figure 4.1), however there was insufficient data to create a model. The limit of detection for HF183 was determined at 20 copies/mL and limit of quantification at 200 copies/mL. This trend indicates that the effect of each upstream discharge is discounted with further distance. Other possible sources of *E. coli* are likely negligible as the National Land Cover Database indicates little agricultural activities (57), and fecal contamination is much higher than the baseline level above the residential areas where wildlife is most likely the most significant source of fecal contamination. The model also confirms that levels of *E. coli* are significantly lower during the winter. Since the discharges are considered point sources and their flow rates will not change with the seasons, they will be diluted in the streams differently due to seasonal flow rates of Callahan Creek. Flow rates are generally higher during the winter which would cause more dilution for the fecal indicator bacteria.

Although the model provides a link between untreated household waste, seasonal changes and high levels of fecal contamination, it cannot provide a complete explanation for the difference in *E. coli* levels longitudinally on Callahan Creek and Roaring Fork as the $R^2$ values were relatively modest (0.29 and 0.24, respectively). There are many variables affecting the varying levels of *E. coli* in the streams. These variables were tested in this model, however did not correlate well with the high level of fecal contamination. Water quality parameters were correlated with bacterial markers via Spearmen's rho (table 4.2). A positive trend is seen between *E. coli* and temperature. Higher water temperatures are present in the non-winter months where the levels of *E. coli* were higher. There is a significant negative correlation between the flow rate, depth and *E. coli* levels in Callahan Creek. This is expected as with higher flows, the FIB will be more diluted.

Table 4.2. Spearmen's rho correlation analysis between microbial indicators and water quality parameters

| | Temp | Cond | DO | pH | Depth | Flow Rate[1] |
|---|---|---|---|---|---|---|
| **Callahan Creek** | | | | | | |
| HF183 | 0.09 | **0.58**[2] | -0.15 | 0.09 | 0.01 | 0.11 |
| E. coli | **0.24** | **0.2** | **-0.25** | **0.22** | **-0.23** | **-0.2** |
| **Roaring Fork** | | | | | | |
| E. coli | **0.26** | 0.03 | **-0.22** | -0.13 | 0.13 | -0.03 |

[1]Flow rate data from USGS gauge station 03529500 in the Powell River

[2]**Bold** indicates a significant value with $p < 0.05$

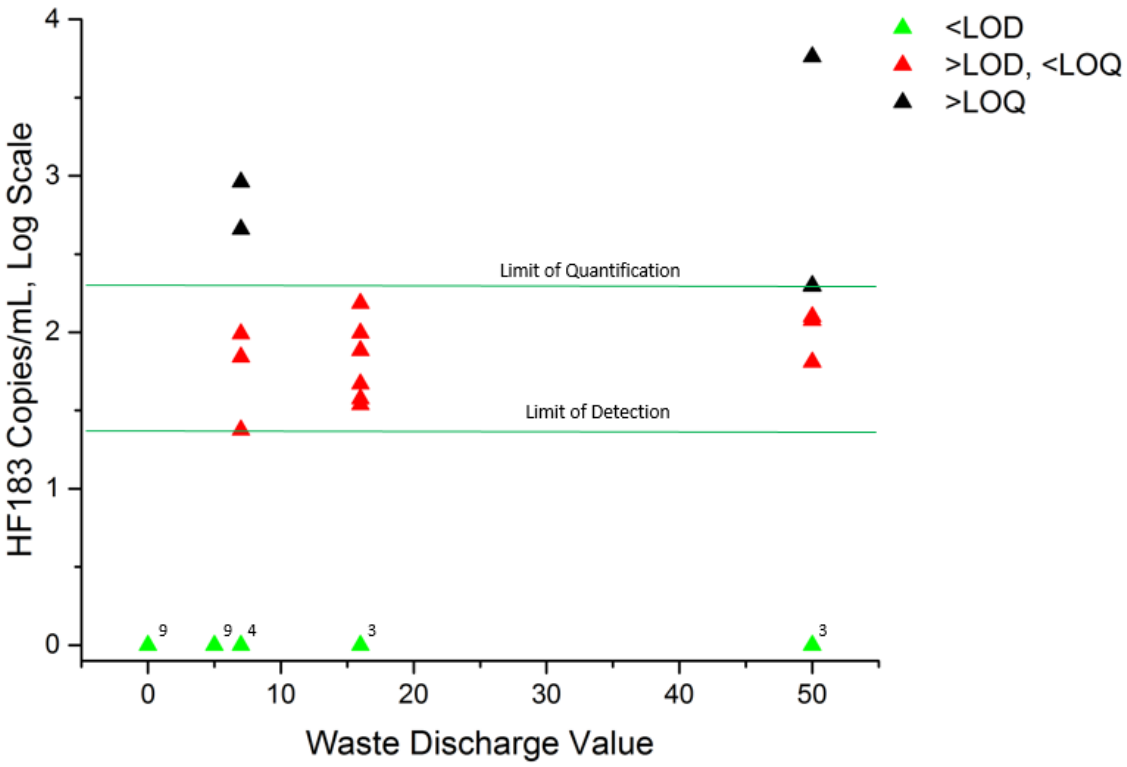


Figure 4.1. HF183 vs. Sum of Inverse Distances to Waste Discharges in Callahan Creek. The data points are extrapolated below the limit of quantification, and the number of samples below the limit of detection is indicated by n.

*4.2. HF183 Presence*

The genetic marker HF183 was found in varying levels in samples collected from Callahan Creek but was not found in any samples collected from Roaring Fork (Figure 4.2). Water quality data for both Callahan Creek and Roaring Fork are summarized in tables 4.3 and 4.4 respectively. HF183 is known to be present at varying levels in different individuals (54); the levels of HF183 observed in Callahan Creek may well be related to the number of "carriers" versus "non-carriers" contributing to the fecal contamination at the time of the sampling. Though both watersheds comprise only a few scattered communities, Callahan Creek does have roughly double the population of Roaring Fork (61). Perhaps due to the low population density, there may not be enough HF183 carriers in Roaring Fork to produce measurable levels of contamination. Additionally, residents may have had high water usage prior to sampling, diluting the waste containing the genetic marker (50).

Table 4.3. Water Quality Data for Callahan Creek

| Site ID | | E. coli (MPN/100 mL) | HF183 (Copies/mL) | Temperature (°C) | Conductivity (µS/cm) | Dissolved Oxygen (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|---|
| | Max | 3405 | 9 | 19.7 | 731 | 15.80 | 8.52 | 33 |
| CC-10 | Median | 0 | 0 | 10.1 | 398 | 9.60 | 8.08 | 23 |
| | Min | 0 | 0 | 2.7 | 211 | 4.90 | 7.40 | 7 |
| | Max | 13327 | 5770 | 19.7 | 1918 | 15.80 | 8.80 | 64 |
| CC-09 | Median | 632 | 118 | 11.3 | 858 | 8.75 | 8.25 | 28 |
| | Min | 0 | 3 | 3.1 | 213 | 6.00 | 7.80 | 4 |
| | Max | 16071 | 913 | 19.4 | 910 | 15.30 | 8.71 | 43 |
| CC-06 | Median | 738 | 23 | 10.2 | 570 | 9.70 | 8.14 | 20 |
| | Min | 0 | 8 | 3.9 | 254 | 6.30 | 7.64 | 10 |
| | Max | 3277 | 152 | 18.9 | 903 | 14.60 | 8.72 | 51 |
| CC-04 | Median | 304 | 37 | 10.4 | 608 | 9.25 | 8.10 | 25 |
| | Min | 96 | 10 | 4.2 | 340 | 5.80 | 7.65 | 8 |
| | Max | 2847 | 17 | 19.0 | 833 | 14.70 | 8.72 | 58 |
| CC-01 | Median | 115 | 1 | 10.2 | 552 | 10.30 | 8.13 | 41 |
| | Min | 0 | 0 | 2.7 | 328 | 4.70 | 7.57 | 13 |

Table 4.4. Water Quality Data for Roaring Fork

| Site ID | | E. coli (MPN/100 mL) | Temperature (°C) | Conductivity (µS/cm) | Dissolved Oxygen (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| | Max | 457 | 23.8 | 1285 | 14.30 | 8.61 | 53 |
| RF-09 | Median | 1 | 11.0 | 805 | 9.63 | 8.11 | 33 |
| | Min | 0 | 3.6 | 416 | 5.30 | 7.82 | 6 |
| | Max | 20459 | 21.6 | 1136 | 14.80 | 8.57 | 43 |
| RF-08 | Median | 413 | 10.8 | 678.5 | 9.88 | 8.10 | 23 |
| | Min | 9 | 3.9 | 360 | 5.40 | 7.83 | 9 |
| | Max | 228 | 22.3 | 1331 | 15.60 | 8.61 | 53 |
| RF-07 | Median | 0 | 10.9 | 959.5 | 9.70 | 8.02 | 23 |
| | Min | 0 | 5.5 | 679 | 5.20 | 7.77 | 8 |
| | Max | 7712 | 19.3 | 1120 | 13.70 | 8.50 | 66 |
| RF-06 | Median | 202 | 11.2 | 827.5 | 9.25 | 7.88 | 33 |
| | Min | 0 | 5.4 | 410 | 5.30 | 7.36 | 9 |
| | Max | 3451 | 20.3 | 1170 | 15.90 | 8.55 | 64 |
| RF-02 | Median | 100 | 11.0 | 827 | 9.95 | 8.01 | 38 |
| | Min | 0 | 3.3 | 531 | 1.60 | 7.61 | 12.2 |

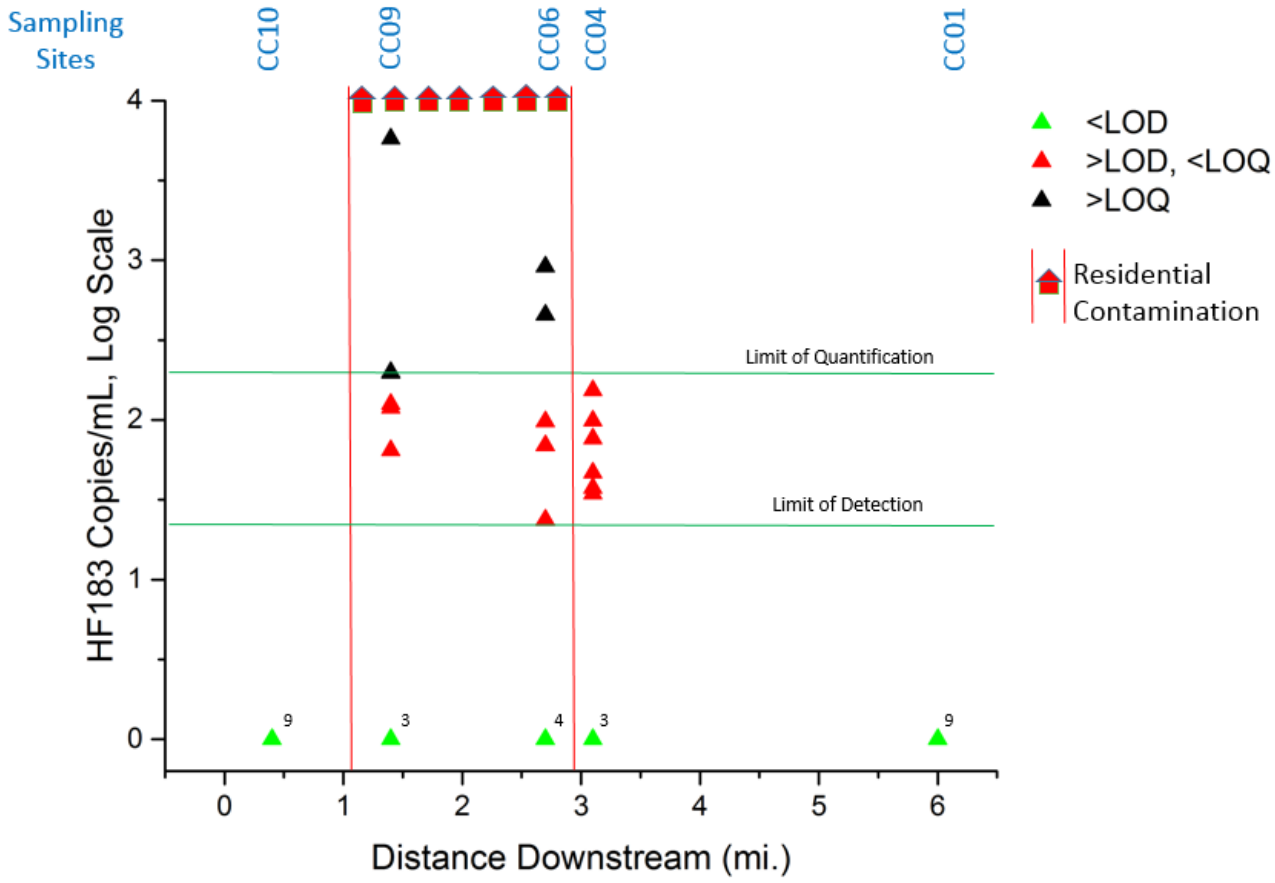


Figure 4.2. Longitudinal Levels of HF183 in Callahan Creek. Distance is measured along the stream starting from the watershed outlet. The data points are extrapolated below the limit of quantification. The number of samples below the limit of detection is indicated by n.

Previous studies have measured HF183 in raw sewage at up to $10^9$ and $10^{10}$ copies per liter (53). Samples taken directly from the household waste effluent ranged from detected but no quantifiable to 59,400 copies per mL. As illustrated in Figure 4.2, HF183 levels were negligible at the most upstream point, which would be expected, given that this sampling site is above the known discharge points of untreated household waste. The next sampling site is directly below a residential community, and shows a spike in the levels of HF183. The genetic marker remains at these levels until the furthest downstream sampling site, where it returns to its baseline levels (i.e. below the limit of detection). The low levels found in this experiment are likely due both to dilution by Callahan Creek and the low number of contributing residences. However detection of the HF183 marker provides a strong indication of human contamination, as it is not generally found in any other animal (14).

Levels of HF183 observed in Callahan Creek do show month-to-month variability (e.g. undetectable to 5,770 copies per 100 mL), as summarized in table 4.5. There are two notable high values in June 2014, where the level of HF183 is relatively high in consecutive sampling sites. This may be due to the timing of sample collection (i.e., when the samples were taken relative to when the HF183-bearing waste was discharging in to the environment).

Figure 4.5. Month to Month Variation of HF183 in Callahan Creek

| Date | CC10 | CC09 | CC06 | CC04 | CC01 |
|---|---|---|---|---|---|
| 4/14 | -[1] | - | - | - | - |
| 5/14 | - | 200[3] | +[2] | + | - |
| 6/14 | - | 5770 | 913 | - | - |
| 7/14 | - | + | + | + | - |
| 8/14 | - | - | - | - | - |
| 1/16 | - | + | - | + | - |
| 2/16 | - | + | 454 | + | - |
| 3/16 | - | - | + | + | - |
| 4/16 | - | 200 | - | + | - |

[1]Negative sample: < 20 copies/mL
[2]Positive sample: >20 copies/mL, <200 copies/mL
[3]Quantifiable sample : >200 copies/mL

It would be expected that levels of HF183 would be affected by seasonal environmental factors such as dilution from differing flow rates of the tributary streams or die-off from UV radiation; both factors would be expected to have negative correlations with the level of HF183 measured. While these may provide some anecdotal explanation of variability in HF183, they are probably less influential than the sample collection timing, and therefore do not show a strong correlation. A Spearmen's rho correlation analysis between the microbial indicators and other water quality parameters can be found in table 4.3. HF183 and conductivity show a moderate correlation (Table 4.2, Spearmen's rho = 0.58). A positive correlation is likely as both water quality parameters originate from the same source: untreated household waste.

*4.3 Co-occurrence of HF183 and E. coli*

Levels of *E. coli* versus HF183 for each sampling site are displayed in figure 4.3. HF183 and *E. coli* do not show a significant positive correlation in Callahan Creek due to insufficient HF183 data above the level of quantification. However a positive trend can be seen when looking at data extrapolated below the LOQ. *E. coli* and *Bacteroides* both originate from human fecal contamination, and therefore a positive correlation between both biological markers is expected. However, imperfections in their co-occurrence may occur for number of reasons. Due to the variability of HF183 in the human population, individual discharges may not always contain HF183 while they contain *E. coli*, and different discharges may have been contributing to the tributaries in question at the time of the sampling. In 20 out of the 45 samples, HF183 was below the detection limit while *E. coli* was detected. These samples are mainly at the sampling sites least influenced by untreated household waste (i.e. at the bottom of the watershed, before the residential areas). Therefore in polluted areas, when searching for HF183, it is more critical to sample close to the source of contamination.

*E. coli* may also originate from other sources such as wildlife and domestic animals, while HF183 generally does not which may explain cases where *E. coli* is present while HF183 is not. However, downstream levels of *E. coli* remains higher than the baseline level before the residential areas (figure 4.4), and therefore it is more likely that discrepancies between the two markers is caused by differences in longitudinal persistence. In Roaring Fork, *E. coli* shows a similar trend of remaining elevated at the watershed outlet (figure 4.5), however it cannot be compared with HF183 as HF183 was not detected in this watershed. *Bacteroides* and *E. coli* are both anaerobic bacteria acclimated to the mammalian gut, and therefore do not survive well the aerobic environment. In most cases both bacteria will experience die-off, however die-off may be occurring at differing rates. Tambalo et. al. (2012) found a decay rate in the human associated *Bacteroides* marker BacH that was quicker than *E. coli.* This study concluded that the exposure to sunlight and predation may be causing a quicker die-off rate in *Bacteroides* (48). Dick et. al. showed that human associated markers show a faster decay than cultured *E. coli* in conditions with reduced predation and exposure to sunlight (62). It is possible that these conditions are present, and contributing to a shorter decay time of HF183 than *E. coli* in Callahan Creek, which is likely causing some variability in their co-occurrence. However since the stream reach has fast

moving waters, die-off may not have enough time to become significant, and may be overshadowed by differences in abundance and dilution. Another contributing factor toward differing longitudinal persistence could arise from analytical techniques: HF183, quantified by qPCR, measures 16S-rRNA from viable and non-viable *Bacteroides*, while the IDEXX procedures measures viable *E. coli* growth in a nutrient rich medium. Non-viable genetic matter may take longer to completely decay in the environment than viable organisms. Therefore persistence of viable *Bacteroides* is likely shorter than HF183, and would show a larger discrepancy with levels of *E. coli*.

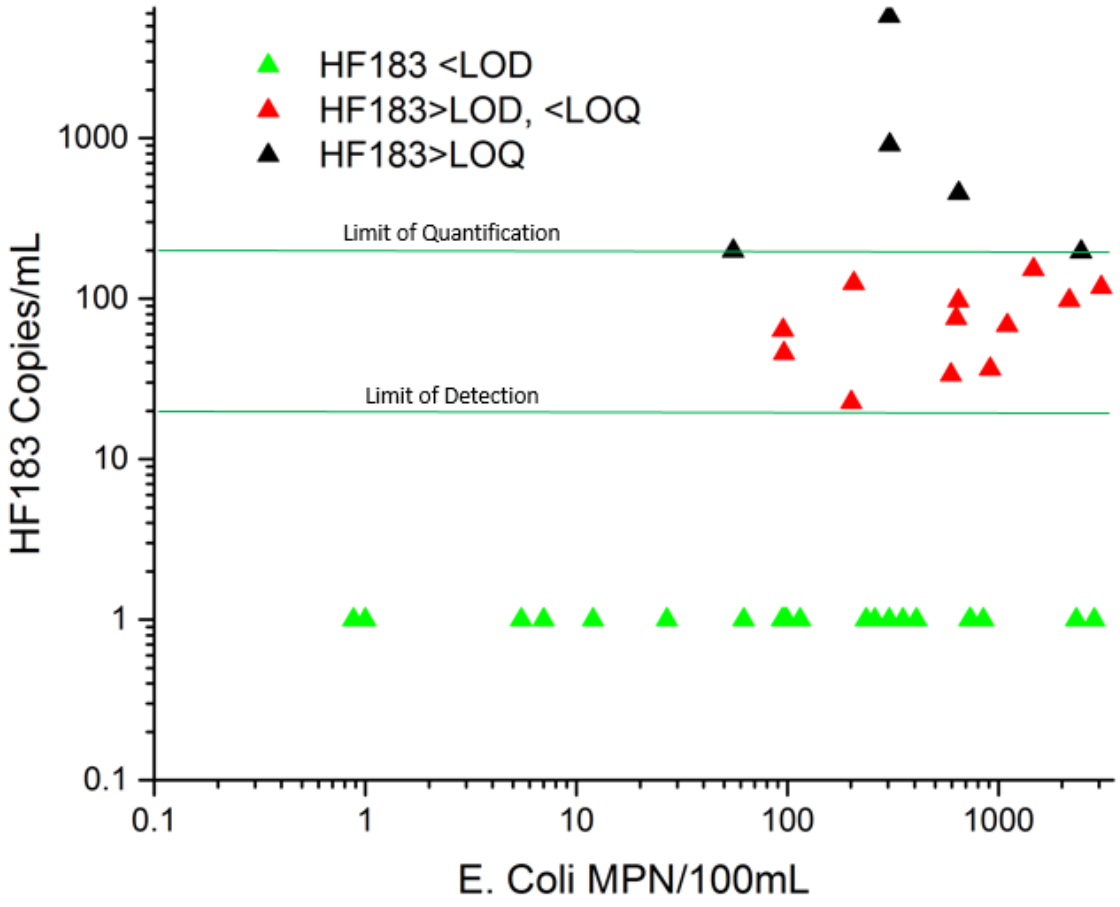


Figure 4.3. HF183 vs. *E. coli* in Callahan Creek. Data is extrapolated below the limit of quantification. The data below the limit of detection is considered not detected.

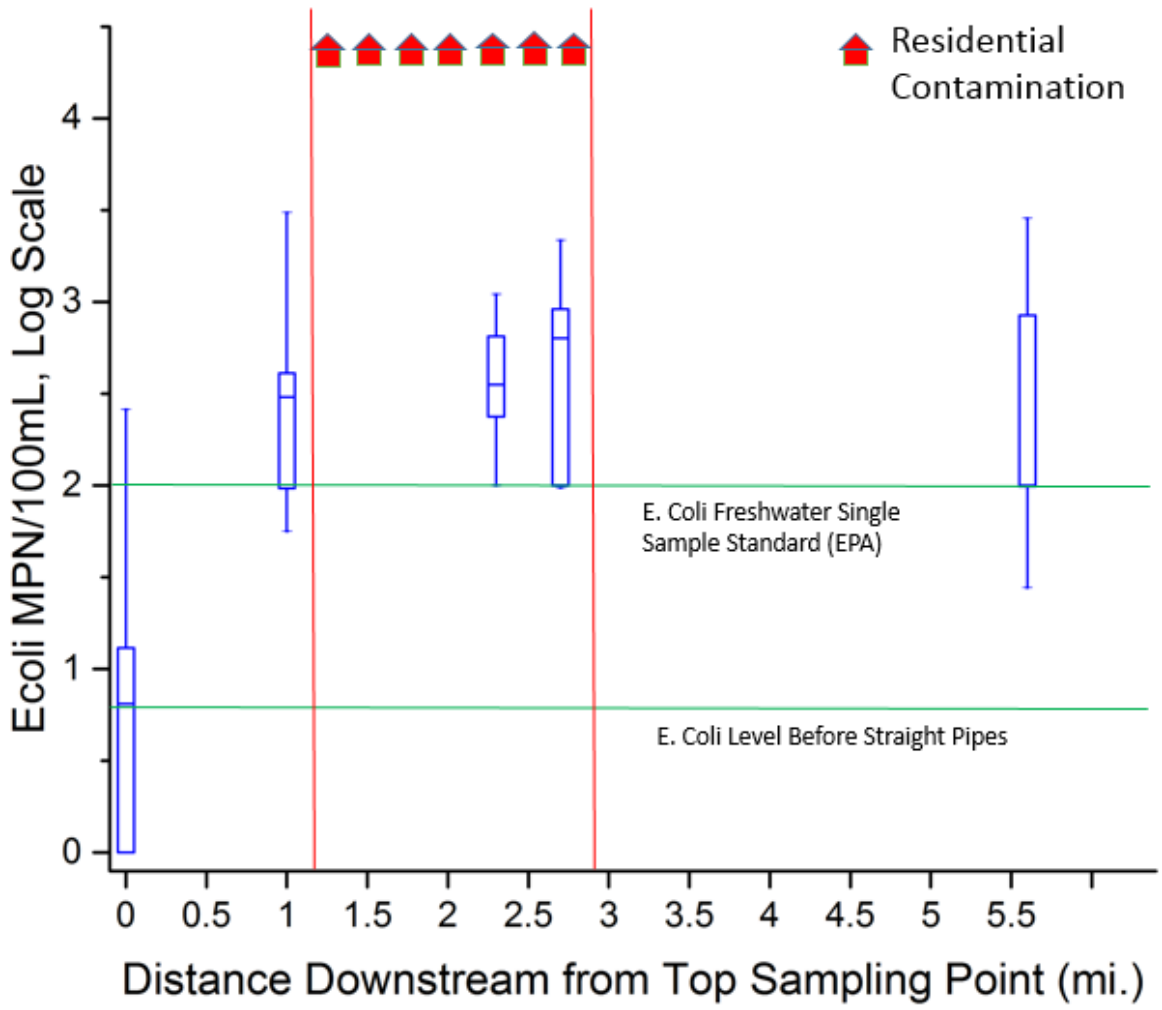


Figure 4.4. Longitudinal *E. coli* Levels in Callahan Creek. The box and whisker plots display the minimum and maximum values, 25$^{th}$ and 75$^{th}$ quartiles as well as the median values.

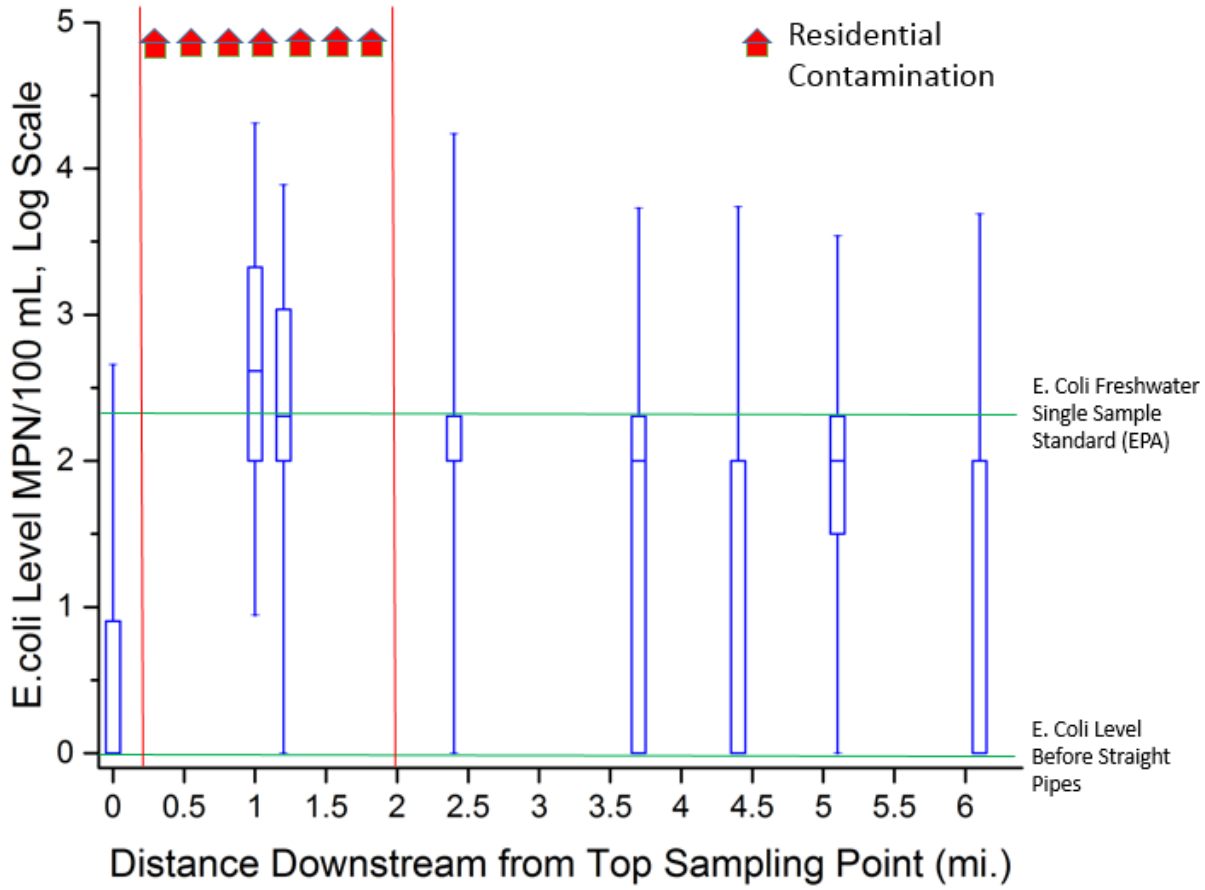


Figure 4.5. Longitudinal *E. coli* Levels in Roaring Fork. The box and whisker plots display the minimum and maximum values, 25$^{th}$ and 75$^{th}$ quartiles as well as the median values.

## 5. Conclusions

Although the source tracking marker, HF183, has routinely been detected downstream from populated areas contaminated by human sewage (14, 33, 53), there has been little previous demonstration of its usefulness in rural areas. This study demonstrated that HF183 can be found in rural watersheds with known upstream direct discharges of untreated household waste (e.g. via "straight-pipes"). However, the levels of HF183 detected were generally low, and the marker was only found in one of two study watersheds; the watersheds have similar sanitation problems, but relatively different population sizes. It is known that HF183 is present at varying levels in different individuals (54), and the study results imply that the presence of HF183 in receiving waters may depend on the population size of "carriers" of this genetic marker. In addition, due to the pulse-nature of straight-pipe discharges, sample collection timing may significantly impact the ability to capture HF183. Seasonal temperature and hydrologic fluctuations may be significant factors as well.

When comparing HF183 and *E. coli* in the study watershed where HF183 was consistently detected, there was not enough quantifiable data for a correlation between HF183 and E. coli, however a positive trend could be seen with HF183 data extrapolated below the level of quantification. Previous studies have found weak, positive correlations between the two markers (48, 55). Imperfections in the co-occurrence between the two markers are likely explained by many different factors, including the varying ratio of HF183 and *E. coli* in each individual discharge, and differences in fate and transport between the two organisms. It appears that HF183 returns to the baseline levels more quickly than *E. coli,* which is consistent with previous work also showing lower persistence rates of *Bacteroides* (48). Differences in persistence may be due to the different effect of UV radiation and predation on the two organisms (62). Although the two indicators are related, it may be hard to use HF183 in widespread monitoring as it is not always detected when human fecal contamination and a public health hazard is clearly present. However if human contamination is suspected, HF183 and *E. coli* used in conjunction may provide strong evidence of *human* fecal contamination in addition to showing elevated levels of fecal contamination. This is consistent with Sidhu et. al. (2013) and Harwood et. al. (2009), who

have suggested using a strategically selected set of markers for the most informed water quality assessment.

In addition to detecting HF183, *E. coli* levels were statistically associated with discharges of untreated household waste via a multiple linear regression model. The model indicates that the level of *E. coli* depends on the number of discharges as well as the distances of discharges from the sampling site and confirmed that levels of *E. coli* are lower during the winter months. The multiple linear regression model worked well in these watersheds to link the high levels of fecal contamination to the discharges of untreated household waste. This may be as there were no other significant sources of fecal contamination (e.g., agriculture, wildlife). However more variables such as UV radiation and stream flow may need to be included to create a model with more predictive abilities.

This multi-disciplinary and multi-step approach may lead to effective targeting of contamination sources, and reduce overall exposure to pathogens in impoverished rural communities. These findings may have applications beyond the scope of Appalachia, as there are other areas in the United States affected by poor sanitation infrastructure and lacking resources for infrastructure investment, including farmland on the US-Canada and US-Mexico border, as well as areas in the semiarid plains, and in states in the south and southeast (1, 2). Mitigation priorities set by source tracking could lead to more effective measures to reduce fecal contamination in the environment and thereby lower the risk of waterborne disease outbreaks in rural communities.

## 6. Future Research

HF183 was successfully recovered in Callahan Creek. The river may be the point of exposure to waterborne disease for those using the waters for recreational purposes, however it may be useful to determine if the hazard persists to private drinking water. Studies could be undertaken to determine whether the marker and waterborne pathogens can move through soil. It may also be useful to determine if HF183 is present in the private wells and spring water used for household purposes. Additionally, there is little information on the incidence of waterborne disease in the area. Surveys could be conducted to determine if local residents are affected at all by waterborne disease, and what toll it has on the local economy. With this information, more informed decisions can be made on the scale of investment in wastewater infrastructure.

Although HF183 was detected in this study, it was only recovered from one watershed. There are many areas in Appalachia with poor sanitation, and it may be useful to determine the conditions necessary for successful detection of HF183. Based on the findings of this study, it may be wise for regulators to use the genetic marker only in areas that meet certain requirements as part of their water quality assessment "toolbox". An important factor would therefore be to determine the necessary population density for the marker to appear. Tambalo et. al. (2012) found a *Bacteroides* associated marker in a rural watershed and stated that future research could be done in the area to gain insight on the population density and household waste treatment systems. A study could be undertaken in adjacent (or as close as possible) watersheds polluted with known sources of human fecal contamination but with varying population sizes. The levels of HF183 could be compared and determine whether there is a relationship between the population size and the level of HF183 recovered. If regulators limit the use of this marker only to areas where the population density is large enough for it to be recovered, it may save them money and effort in refraining to search for the marker where it cannot be found.

Studies on sampling timing could also optimize the most effective HF183 recovery. It would be useful to conduct studies to determine what time of the year is optimal for discovering the marker. Additionally, studies could determine daily discharge trends of untreated household

waste. This could improve the recovery of HF183, particularly in watersheds that have a borderline population density for marker recovery.

A better understanding of the fate and transport of HF183 may allow for better interpretation of data results. More can be done to understand the fate and transport of the source tracking maker, particularly its die-off patterns and ability to live in soil. After successful detection of BacH in Austria, Reischer et. al. (2007) suggested tests to understand its ability to live in soil and sediments (49). It may also be useful to find a genetic marker on *E. coli* and compare that with HF183. Comparing two genetic markers as opposed to a genetic marker and viable bacteria may improve the correlation between *E. coli* and *Bacteroides*.

Similarly these tests could be repeated in other areas in the United States with infrastructure problems; this study may serve as precedent in testing for HF183 in rural areas. It has been shown that HF183 can be recovered below individual discharges of untreated household waste in Appalachia. Areas such as farmland on the US-Canada and US-Mexico border, as well as areas in the semiarid plains, and in states in the south and southeast may also find HF183 useful to justify the investment of sewage infrastructure.

The statistical model in this study drew a link between untreated household waste and elevated levels of fecal contamination without the use of another genetic marker. It raises the question on the necessity of HF183 or other source tracking markers in this area. It may be useful to determine if enough statistical evidence, using only indicators currently in water quality assessment, could be assembled to link the high levels of fecal contamination with the waste discharges, and justify the investment in sewage infrastructure. It would also be useful to explore whether a similar statistical model could be used in watersheds where other sources of fecal contamination are present (e.g., agriculture, wildlife).

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## Appendix A. HF183 Data

Table A.1. HF183 in Callahan Creek Water Samples

| Site ID | Date | HF183 (Copies/mL) | Site ID | Date | HF183 (Copies/mL) |
|---|---|---|---|---|---|
| CC-01 | 4/11/2014 | 0 | CC-06 | 8/14/2014 | 8 |
| CC-01 | 5/15/2014 | 7 | CC-06 | 1/15/2016 | 9 |
| CC-01 | 6/17/2014 | 1 | CC-06 | 2/12/2016 | 454 |
| CC-01 | 7/21/2014 | 17 | CC-06 | 3/18/2016 | 97 |
| CC-01 | 8/14/2014 | 8 | CC-06 | 4/13/2016 | 12 |
| CC-01 | 1/15/2016 | 7 | CC-09 | 4/11/2014 | 16 |
| CC-01 | 2/12/2016 | 0 | CC-09 | 5/15/2014 | 200 |
| CC-01 | 3/18/2016 | 0 | CC-09 | 6/17/2014 | 5770 |
| CC-01 | 4/13/2016 | 0 | CC-09 | 7/21/2014 | 118 |
| CC-04 | 4/11/2014 | 14 | CC-09 | 8/14/2014 | 5 |
| CC-04 | 5/15/2014 | 75 | CC-09 | 1/15/2016 | 125 |
| CC-04 | 6/17/2014 | 10 | CC-09 | 2/12/2016 | 64 |
| CC-04 | 7/21/2014 | 152 | CC-09 | 3/18/2016 | 3 |
| CC-04 | 8/14/2014 | 12 | CC-09 | 4/13/2016 | 200 |
| CC-04 | 1/15/2016 | 46 | CC9SP | 2/12/2016 | 0 |
| CC-04 | 2/12/2016 | 98 | CC-10 | 4/11/2014 | 0 |
| CC-04 | 3/18/2016 | 37 | CC-10 | 5/15/2014 | 0 |
| CC-04 | 4/13/2016 | 34 | CC-10 | 6/17/2014 | 5 |
| CC5SP | 2/12/2016 | 813 | CC-10 | 7/21/2014 | 9 |
| CC5SP | 3/18/2016 | 171 | CC-10 | 8/14/2014 | 0 |
| CC5SP | 4/13/2016 | 59400 | CC-10 | 1/15/2016 | 0 |
| CC-06 | 4/11/2014 | 16 | CC-10 | 2/12/2016 | 0 |
| CC-06 | 5/15/2014 | 68 | CC-10 | 3/18/2016 | 0 |
| CC-06 | 6/17/2014 | 913 | CC-10 | 4/13/2016 | 0 |
| CC-06 | 41841 | 23 | | | |

## Appendix B. Water Quality Data Callahan Creek

Table B.1. Water Quality Data for Callahan Creek Water Samples, CC-01

| Site ID | Date | E. coli (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-01 | 8/20/2012 | 202 | 15.56 | 770 | 10.2 | 8.55 | ND |
| CC-01 | 9/20/2012 | 516 | 14 | 660 | 10.4 | 8.42 | ND |
| CC-01 | 10/18/2012 | 100 | 10.94 | 663 | 12 | 8.56 | ND |
| CC-01 | 11/15/2012 | 100 | 5.67 | 509 | 14.5 | 8.72 | ND |
| CC-01 | 12/13/2012 | 100 | 3.72 | 431 | 8.1 | 7.94 | ND |
| CC-01 | 1/11/2013 | 521 | 9.56 | 542 | 4.7 | 8.24 | ND |
| CC-01 | 2/14/2013 | 745 | 6.61 | 490 | 12 | 8.64 | ND |
| CC-01 | 3/21/2013 | 202 | 3.44 | 376 | 14.1 | 8.2 | ND |
| CC-01 | 4/25/2013 | 0 | 9.8 | 484 | 10.9 | 8.72 | ND |
| CC-01 | 5/29/2013 | 100 | 16.2 | 722 | 7.7 | 8.12 | ND |
| CC-01 | 6/24/2013 | 1480 | 17.3 | 711 | 9.8 | 8.12 | ND |
| CC-01 | 7/22/2013 | 99 | 19 | 689 | 6.9 | 8.1 | ND |
| CC-01 | 8/19/2013 | 202 | 18.3 | 832 | 5 | 8.14 | ND |
| CC-01 | 9/25/2013 | 626 | 15.9 | 833 | 6.4 | 8.19 | 25 |
| CC-01 | 10/23/2013 | 306 | 10.2 | 700 | 6.3 | 8.18 | 13 |
| CC-01 | 11/13/2013 | 100 | 2.7 | 627 | 6.6 | 8.3 | 26 |
| CC-01 | 12/12/2013 | 100 | 5.9 | 442 | 14.7 | 7.61 | 48 |
| CC-01 | 1/16/2014 | 738 | 4.5 | 409 | 13.8 | 7.66 | 53 |
| CC-01 | 2/21/2014 | 201 | 8.4 | 328 | 10.6 | 7.57 | 58 |
| CC-01 | 3/28/2014 | 100 | 9 | 550 | 10.5 | 7.93 | 41 |
| CC-01 | 4/11/2014 | 100 | 14.5 | 564 | 10.6 | 8.46 | 46 |
| CC-01 | 5/15/2014 | 2847 | 17.1 | 509 | 8.2 | 7.87 | 48 |
| CC-01 | 6/17/2014 | 100 | 18.9 | 817 | 8.6 | 8.08 | 30 |
| CC-01 | 7/21/2014 | 99 | 17.4 | 553 | 9.1 | 7.86 | 48 |
| CC-01 | 8/12/2014 | 2334 | 19 | 471 | 8.2 | 8.06 | 51 |
| CC-01 | 1/15/2016 | 115 | 3.7 | 524 | 12.4 | 7.9 | 14 |
| CC-01 | 2/12/2016 | 27 | 3.7 | ND | ND | ND | ND |
| CC-01 | 3/18/2016 | 96 | 8.9 | 512 | 11.4 | 8.1 | 17 |
| CC-01 | 4/13/2016 | 846 | 14.5 | 564 | 10.6 | 8.4 | 18 |

Table B.2. Water Quality Data for Callahan Creek Water Samples, CC-02

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-02 | 8/20/2012 | 100 | 15.56 | 770 | 10.3 | 8.52 | ND |
| CC-02 | 9/20/2012 | 413 | 14 | 670 | 10.5 | 8.33 | ND |
| CC-02 | 10/18/2012 | 202 | 11 | 675 | 11.7 | 8.51 | ND |
| CC-02 | 11/15/2012 | 0 | 5.61 | 510 | 14.3 | 8.56 | ND |
| CC-02 | 12/13/2012 | 0 | 3.94 | 442 | 11.4 | 8.17 | ND |
| CC-02 | 1/11/2013 | 306 | 9.61 | 544 | 9 | 8.31 | ND |
| CC-02 | 2/14/2013 | 202 | 7 | 502 | 12.2 | 8.6 | ND |
| CC-02 | 3/21/2013 | 100 | 3.72 | 383 | 13.8 | 8.31 | ND |
| CC-02 | 4/25/2013 | 202 | 9.8 | 490 | 9.8 | 8.65 | ND |
| CC-02 | 5/29/2013 | 100 | 15.9 | 731 | 7.7 | 8.13 | ND |
| CC-02 | 6/24/2013 | 3451 | 17.3 | 719 | 7.5 | 8.09 | ND |
| CC-02 | 7/22/2013 | 413 | 18.9 | 705 | 6.9 | 8.09 | ND |
| CC-02 | 8/19/2013 | 100 | 18 | 840 | 5.1 | 8.13 | ND |
| CC-02 | 9/25/2013 | 738 | 15.7 | 844 | 6.5 | 8.15 | 38 |
| CC-02 | 10/23/2013 | 201 | 10.3 | 799 | 6.4 | 8.04 | 53 |
| CC-02 | 11/13/2013 | 0 | 3.6 | 649 | 6.5 | 8.23 | 76 |
| CC-02 | 12/12/2013 | 0 | 5.9 | 447 | 14.7 | 7.59 | 46 |
| CC-02 | 1/16/2014 | 0 | 4.6 | 415 | 14.3 | 7.66 | 39 |
| CC-02 | 2/21/2014 | 100 | 8.4 | 323 | 10.9 | 7.58 | 53 |
| CC-02 | 3/28/2014 | 306 | 9 | 554 | 10.4 | 7.96 | 25 |
| CC-02 | 4/11/2014 | 100 | 14 | 570 | 9.9 | 8.19 | 28 |
| CC-02 | 5/15/2014 | 1223 | 16.9 | 504 | 8.5 | 7.82 | 33 |
| CC-02 | 6/17/2014 | 516 | 18.5 | 747 | 9 | 8.14 | 13 |
| CC-02 | 7/21/2014 | 413 | 17.2 | 560 | 9.3 | 7.96 | 28 |
| CC-02 | 8/12/2014 | 1323 | 19.1 | 527 | 8.2 | 8.04 | 38 |

Table B.3. Water Quality Data for Callahan Creek Water Samples, CC-03

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-03 | 8/20/2012 | 409 | 15.17 | 800 | 10.2 | 8.53 | ND |
| CC-03 | 9/20/2012 | 202 | 13.89 | 710 | 10.3 | 8.43 | ND |
| CC-03 | 10/18/2012 | 100 | 10.5 | 713 | 11.2 | 8.52 | ND |
| CC-03 | 11/15/2012 | 306 | 5.72 | 536 | 13.2 | 8.51 | ND |
| CC-03 | 12/13/2012 | 413 | 4.33 | 450 | 11.7 | 8.27 | ND |
| CC-03 | 1/11/2013 | 100 | 9.72 | 593 | 9.1 | 8.35 | ND |
| CC-03 | 2/14/2013 | 201 | 7 | 548 | 12.2 | 8.64 | ND |
| CC-03 | 3/21/2013 | 306 | 3.83 | 421 | 11.9 | 8.34 | ND |
| CC-03 | 4/25/2013 | 626 | 9.7 | 531 | 10.7 | 8.66 | ND |
| CC-03 | 5/29/2013 | 0 | 15.4 | 726 | 9.4 | 8.11 | ND |
| CC-03 | 6/24/2013 | 6437 | 17 | 755 | 7.4 | 8.08 | ND |
| CC-03 | 7/22/2013 | 202 | 18.4 | 765 | 7.3 | 8.09 | ND |
| CC-03 | 8/19/2013 | 409 | 17.7 | 886 | 5.7 | 8.13 | ND |
| CC-03 | 9/25/2013 | 202 | 15.7 | 887 | 6.6 | 8.15 | 15 |
| CC-03 | 10/23/2013 | 409 | 10.3 | 826 | 6.6 | 8.11 | 17 |
| CC-03 | 11/13/2013 | 306 | 3.4 | 658 | 6.6 | 8.23 | 20 |
| CC-03 | 12/12/2013 | 413 | 6 | 484 | 13.4 | 7.66 | 33 |
| CC-03 | 1/16/2014 | 80 | 4.9 | 461 | 14.2 | 7.79 | 34 |
| CC-03 | 2/21/2014 | 202 | 8.4 | 344 | 10.6 | 7.68 | 56 |
| CC-03 | 3/28/2014 | 405 | 9.2 | 598 | 10.4 | 7.97 | 28 |
| CC-03 | 4/11/2014 | 100 | 13.9 | 617 | 9.8 | 8.16 | 20 |
| CC-03 | 5/15/2014 | 632 | 16.4 | 539 | 8.4 | 7.82 | 28 |
| CC-03 | 6/17/2014 | 201 | 17.7 | 767 | 9 | 8.21 | 18 |
| CC-03 | 7/21/2014 | 202 | 16.9 | 588 | 9.1 | 8 | 28 |
| CC-03 | 8/12/2014 | 1967 | 19 | 562 | 8.3 | 8.08 | 25 |

Table B.4. Water Quality Data for Callahan Creek Water Samples, CC-04

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-04 | 8/20/2012 | 521 | 15.72 | 820 | 10.3 | 8.51 | ND |
| CC-04 | 9/20/2012 | 201 | 14.06 | 720 | 10.2 | 8.44 | ND |
| CC-04 | 10/18/2012 | 306 | 10.72 | 723 | 11.2 | 8.51 | ND |
| CC-04 | 11/15/2012 | 304 | 6.17 | 535 | 13.6 | 8.58 | ND |
| CC-04 | 12/13/2012 | 100 | 4.56 | 490 | 12.8 | 8.32 | ND |
| CC-04 | 1/11/2013 | 100 | 9.78 | 590 | 9.2 | 8.37 | ND |
| CC-04 | 2/14/2013 | 413 | 7.28 | 553 | 12.3 | 8.66 | ND |
| CC-04 | 3/21/2013 | 304 | 4.17 | 422 | 7.5 | 8.43 | ND |
| CC-04 | 4/25/2013 | 202 | 10.3 | 537 | 7.5 | 8.72 | ND |
| CC-04 | 5/29/2013 | 201 | 16 | 775 | 7.5 | 8.1 | ND |
| CC-04 | 6/24/2013 | 3277 | 17.2 | 759 | 7.2 | 8.07 | ND |
| CC-04 | 7/22/2013 | 1464 | 18.5 | 779 | 6.7 | 8.07 | ND |
| CC-04 | 8/19/2013 | 521 | 17.6 | 898 | 5.8 | 8.09 | ND |
| CC-04 | 9/25/2013 | 202 | 15.8 | 903 | 6.7 | 8.12 | 22 |
| CC-04 | 10/23/2013 | 413 | 10.4 | 830 | 7.8 | 8.08 | 15 |
| CC-04 | 11/13/2013 | 304 | 4.4 | 706 | 6.3 | 8.27 | 18 |
| CC-04 | 12/12/2013 | 100 | 6.2 | 487 | 13.8 | 7.67 | 30 |
| CC-04 | 1/16/2014 | 101 | 5 | 460 | 14.6 | 7.86 | 33 |
| CC-04 | 2/21/2014 | 96 | 8.4 | 340 | 10.6 | 7.65 | 51 |
| CC-04 | 3/28/2014 | 1089 | 9.3 | 599 | 10.5 | 8.04 | 28 |
| CC-04 | 4/11/2014 | 100 | 14.9 | 625 | 9.3 | 8.2 | 25 |
| CC-04 | 5/15/2014 | 632 | 16.2 | 560 | 8.5 | 7.82 | 30 |
| CC-04 | 6/17/2014 | 100 | 17.8 | 849 | 8.7 | 8.08 | 23 |
| CC-04 | 7/21/2014 | 1464 | 16.9 | 580 | 9.1 | 8.02 | 25 |
| CC-04 | 8/12/2014 | 731 | 18.9 | 532 | 8.3 | 8.05 | 30 |
| CC-04 | 8/20/2012 | 96 | 4.7 | 591 | 12.1 | 8.1 | 9 |
| CC-04 | 9/20/2012 | 2162 | 4.7 | ND | ND | ND | ND |
| CC-04 | 10/18/2012 | 912 | 9.4 | 617 | 9.8 | 8.1 | 8 |
| CC-04 | 11/15/2012 | 596 | 14.9 | 625 | 9.3 | 8.2 | 10 |

Table B.5. Water Quality Data for Callahan Creek Water Samples, CC-05

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-05 | 8/20/2012 | 979 | 14.72 | 810 | 9.8 | 8.54 | ND |
| CC-05 | 9/20/2012 | 2341 | 13.5 | 750 | 9.9 | 8.49 | ND |
| CC-05 | 10/18/2012 | 2590 | 10.17 | 706 | 11.6 | 8.56 | ND |
| CC-05 | 11/15/2012 | 100 | 5.67 | 508 | 13.5 | 8.66 | ND |
| CC-05 | 12/13/2012 | 0 | 4.44 | 449 | 12.6 | 8.34 | ND |
| CC-05 | 1/11/2013 | 745 | 9.89 | 556 | 9.3 | 8.39 | ND |
| CC-05 | 2/14/2013 | 516 | 7.61 | 511 | 11.2 | 8.61 | ND |
| CC-05 | 3/21/2013 | 18501 | 4.06 | 365 | 9.8 | 8.39 | ND |
| CC-05 | 4/25/2013 | 3986 | 10.1 | 520 | 8.6 | 8.72 | ND |
| CC-05 | 5/29/2013 | 11602 | 15.4 | 750 | 7.3 | 8.13 | ND |
| CC-05 | 6/24/2013 | 7173 | 17.1 | 661 | 7 | 8.11 | ND |
| CC-05 | 7/22/2013 | 632 | 18.1 | 756 | 6.6 | 8.11 | ND |
| CC-05 | 8/19/2013 | 979 | 17.1 | 907 | 6.1 | 8.16 | ND |
| CC-05 | 9/25/2013 | 620 | 15.2 | 876 | 6.9 | 8.16 | 15 |
| CC-05 | 10/23/2013 | 3051 | 10.4 | 808 | 6.6 | 8.13 | 17 |
| CC-05 | 11/13/2013 | 100 | 4.3 | 683 | 6.6 | 8.31 | 18 |
| CC-05 | 12/12/2013 | 0 | 5.7 | 437 | 12.5 | 7.65 | 18 |
| CC-05 | 1/16/2014 | 201 | 4.5 | 413 | 15.6 | 7.83 | 34 |
| CC-05 | 2/21/2014 | 72 | 7.8 | 259 | 10.7 | 7.69 | 33 |
| CC-05 | 3/28/2014 | 3310 | 9.8 | 578 | 10.2 | 8.11 | 15 |
| CC-05 | 4/11/2014 | 413 | 14 | 506 | 9.7 | 8.19 | 20 |
| CC-05 | 5/15/2014 | 2133 | 16.4 | 493 | 8.4 | 7.86 | 18 |
| CC-05 | 6/17/2014 | 620 | 17.1 | 810 | 8.7 | 8.13 | 20 |
| CC-05 | 7/21/2014 | 632 | 16.6 | 481 | 9 | 8.03 | 30 |
| CC-05 | 8/12/2014 | 304 | 19.5 | 473 | 8.2 | 8.06 | 36 |

Table B.6. Water Quality Data for Callahan Creek Water Samples, CC-06

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-06 | 41141 | 960 | 14.5 | 820 | 10.3 | 8.57 | ND |
| CC-06 | 41172 | 2745 | 13.5 | 730 | 10.3 | 8.49 | ND |
| CC-06 | 41200 | 4434 | 10.22 | 702 | 11.2 | 8.56 | ND |
| CC-06 | 41228 | 16071 | 5.67 | 508 | 13.2 | 8.61 | ND |
| CC-06 | 41256 | 0 | 4.39 | 414 | 12.5 | 8.39 | ND |
| CC-06 | 41285 | 1089 | 9.89 | 577 | 9.2 | 8.44 | ND |
| CC-06 | 41319 | 1078 | 7.28 | 563 | 11.9 | 8.7 | ND |
| CC-06 | 41354 | 413 | 3.94 | 380 | 10 | 8.45 | ND |
| CC-06 | 41389 | 3451 | 10.2 | 522 | 8.7 | 8.71 | ND |
| CC-06 | 41423 | 8329 | 15.2 | 753 | 7.4 | 8.11 | ND |
| CC-06 | 41449 | 6198 | 17.1 | 727 | 7.1 | 8.1 | ND |
| CC-06 | 41477 | 202 | 18.1 | 763 | 6.7 | 8.1 | ND |
| CC-06 | 41505 | 960 | 17.2 | 910 | 6.3 | 8.15 | ND |
| CC-06 | 41542 | 632 | 15.2 | 877 | 6.9 | 8.15 | 17 |
| CC-06 | 41570 | 745 | 10.4 | 784 | 6.7 | 8.13 | 13 |
| CC-06 | 41591 | 16071 | 4.1 | 672 | 6.8 | 8.31 | 17 |
| CC-06 | 41620 | 0 | 5.6 | 441 | 12.2 | 7.68 | 28 |
| CC-06 | 41655 | 306 | 4.5 | 414 | 15.3 | 7.84 | 24 |
| CC-06 | 41691 | 33 | 7.8 | 254 | 10.8 | 7.64 | 43 |
| CC-06 | 41726 | 3546 | 9.8 | 578 | 10.3 | 8.1 | 20 |
| CC-06 | 41740 | 100 | 14 | 544 | 9.7 | 8.16 | 25 |
| CC-06 | 41774 | 1100 | 16.3 | 487 | 8.5 | 7.86 | 18 |
| CC-06 | 41807 | 306 | 16.9 | 828 | 8.6 | 8.08 | 20 |
| CC-06 | 41841 | 202 | 16.6 | 482 | 9 | 7.94 | 23 |
| CC-06 | 41863 | 738 | 19.4 | 471 | 8.1 | 8.03 | 28 |
| CC-06 | 42384 | 236 | 5.2 | 619 | 12 | 8.2 | 12 |
| CC-06 | 42412 | 647 | 5.2 | ND | ND | ND | ND |
| CC-06 | 42447 | 645 | 9.4 | 518 | 11 | 8.1 | 11 |
| CC-06 | 42473 | 352 | 14 | 544 | 9.7 | 8.1 | 10 |

Table B.7. Water Quality Data for Callahan Creek Water Samples, CC-07

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-07 | 8/20/2012 | 10426 | 15.56 | 700 | 10.2 | 8.65 | ND |
| CC-07 | 9/20/2012 | 3089 | 14 | 670 | 10.3 | 8.48 | ND |
| CC-07 | 10/18/2012 | 2686 | 10.06 | 616 | 11.4 | 8.6 | ND |
| CC-07 | 11/15/2012 | 10394 | 5.11 | 434 | 13.8 | 8.68 | ND |
| CC-07 | 12/13/2012 | 413 | 2.89 | 335 | 13.5 | 8.38 | ND |
| CC-07 | 1/11/2013 | 4711 | 9.06 | 444 | 9.7 | 8.36 | ND |
| CC-07 | 2/14/2013 | 2157 | 5.67 | 410 | 12.3 | 8.62 | ND |
| CC-07 | 3/21/2013 | 100 | 2.5 | 294 | 10.3 | 8.41 | ND |
| CC-07 | 4/25/2013 | 3310 | 9.9 | 414 | 8.8 | 8.7 | ND |
| CC-07 | 5/29/2013 | 405 | 16.3 | 451 | 7.2 | 8.15 | ND |
| CC-07 | 6/24/2013 | 61314 | 18.2 | 666 | 6.8 | 8.11 | ND |
| CC-07 | 7/22/2013 | 1579 | 19.6 | 667 | 5 | 8.12 | ND |
| CC-07 | 8/19/2013 | 10426 | 18.6 | 828 | 5.9 | 8.2 | ND |
| CC-07 | 9/25/2013 | 1449 | 15.8 | 755 | 6.7 | 8.17 | 14 |
| CC-07 | 10/23/2013 | 7976 | 10 | 675 | 8.8 | 8.15 | 15 |
| CC-07 | 11/13/2013 | 10394 | 2.9 | 560 | 8.8 | 8.35 | 16 |
| CC-07 | 12/12/2013 | 413 | 5 | 414 | 13.4 | 7.6 | 28 |
| CC-07 | 1/16/2014 | 521 | 3.1 | 314 | 16.6 | 7.68 | 20 |
| CC-07 | 2/21/2014 | 46 | 7.6 | 222 | 10.8 | 7.67 | 43 |
| CC-07 | 3/28/2014 | 1089 | 8.3 | 389 | 11.3 | 8.35 | 20 |
| CC-07 | 4/11/2014 | 1199 | 14.1 | 397 | 9.7 | 8.25 | 17 |
| CC-07 | 5/15/2014 | 860 | 16.7 | 405 | 8.2 | 7.92 | 20 |
| CC-07 | 6/17/2014 | 2086 | 18.4 | 719 | 8.4 | 8.24 | 18 |
| CC-07 | 7/21/2014 | 1579 | 17.2 | 408 | 9 | 8.06 | 28 |
| CC-07 | 8/12/2014 | 507 | 19.7 | 410 | 8.2 | 8.05 | 28 |

Table B.8. Water Quality Data for Callahan Creek Water Samples, CC-08

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-08 | 8/20/2012 | 6496 | 16.39 | 740 | 10.5 | 8.64 | ND |
| CC-08 | 9/20/2012 | 5555 | 14.39 | 700 | 9.9 | 8.42 | ND |
| CC-08 | 10/18/2012 | 8162 | 10.61 | 645 | 11.2 | 8.54 | ND |
| CC-08 | 11/15/2012 | 4959 | 5.5 | 447 | 14.1 | 8.61 | ND |
| CC-08 | 12/13/2012 | 100 | 3.17 | 350 | 13.4 | 8.28 | ND |
| CC-08 | 1/11/2013 | 1323 | 8.33 | 256 | 11.1 | 8.15 | ND |
| CC-08 | 2/14/2013 | 521 | 5.94 | 315 | 13.1 | 8.49 | ND |
| CC-08 | 3/21/2013 | 409 | 2.5 | 274 | 13.7 | 8.32 | ND |
| CC-08 | 4/25/2013 | 2133 | 10.2 | 440 | 10.5 | 8.54 | ND |
| CC-08 | 5/29/2013 | 632 | 17 | 655 | 7.3 | 8.1 | ND |
| CC-08 | 6/24/2013 | 16071 | 18.3 | 646 | 6.5 | 7.96 | ND |
| CC-08 | 7/22/2013 | 409 | 19.6 | 604 | 6 | 7.96 | ND |
| CC-08 | 8/19/2013 | 6496 | 18.7 | 885 | 7.4 | 8.15 | ND |
| CC-08 | 9/25/2013 | 2281 | 15.8 | 754 | 7.1 | 8.08 | 5 |
| CC-08 | 10/23/2013 | 3893 | 10.1 | 678 | 4.5 | 8.08 | 8 |
| CC-08 | 11/13/2013 | 4959 | 3.2 | 568 | 7.6 | 8.27 | 10 |
| CC-08 | 12/12/2013 | 100 | 5.1 | 362 | 17.3 | 7.51 | 33 |
| CC-08 | 1/16/2014 | 202 | 3.3 | 276 | 13.1 | 7.65 | 17 |
| CC-08 | 2/21/2014 | 201 | 7.8 | 194 | 10.7 | 7.68 | 46 |
| CC-08 | 3/28/2014 | 738 | 8.4 | 420 | 11.1 | 8.25 | 14 |
| CC-08 | 4/11/2014 | 2882 | 14.7 | 398 | 9.3 | 8.4 | 13 |
| CC-08 | 5/15/2014 | 1211 | 16.7 | 407 | 8.4 | 7.88 | 20 |
| CC-08 | 6/17/2014 | 521 | 19.2 | 781 | 8.7 | 8.14 | 10 |
| CC-08 | 7/21/2014 | 409 | 17.3 | 400 | 8.9 | 7.98 | 20 |
| CC-08 | 8/12/2014 | 1613 | 19.8 | 420 | 8.2 | 8.07 | 23 |

Table B.9. Water Quality Data for Callahan Creek Water Samples, CC-09

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-09 | 8/20/2012 | 1596 | 17.89 | 1510 | 8.6 | 8.69 | ND |
| CC-09 | 9/20/2012 | 2433 | 15.5 | 1280 | 8.7 | 8.64 | ND |
| CC-09 | 10/18/2012 | 852 | 11.67 | 1081 | 9.6 | 8.67 | ND |
| CC-09 | 11/15/2012 | 1563 | 6.94 | 953 | 12.4 | 8.78 | ND |
| CC-09 | 12/13/2012 | 100 | 5 | 898 | 12 | 8.62 | ND |
| CC-09 | 1/11/2013 | 5041 | 9.94 | 862 | 10.2 | 8.69 | ND |
| CC-09 | 2/14/2013 | 632 | 8.11 | 955 | 11.6 | 8.8 | ND |
| CC-09 | 3/21/2013 | 0 | 4.06 | 477 | 15.8 | 8.59 | ND |
| CC-09 | 4/25/2013 | 626 | 11.3 | 853 | 10.9 | 8.74 | ND |
| CC-09 | 5/29/2013 | 1869 | 18.1 | 1608 | 6 | 8.27 | ND |
| CC-09 | 6/24/2013 | 13327 | 18.5 | 1172 | 6.5 | 8.17 | ND |
| CC-09 | 7/22/2013 | 3069 | 19.7 | 1101 | 6.2 | 8.2 | ND |
| CC-09 | 8/19/2013 | 1596 | 18.5 | 1299 | 6.3 | 8.21 | ND |
| CC-09 | 9/25/2013 | 1890 | 15.8 | 811 | 6.7 | 8.06 | 20 |
| CC-09 | 10/23/2013 | 2462 | 10.3 | 689 | 7.7 | 8.07 | 22 |
| CC-09 | 11/13/2013 | 1563 | 3.6 | 566 | 7.8 | 8.21 | 19 |
| CC-09 | 12/12/2013 | 100 | 6.7 | 901 | 12.4 | 8.1 | 33 |
| CC-09 | 1/16/2014 | 201 | 3.9 | 829 | 14.6 | 8.03 | 38 |
| CC-09 | 2/21/2014 | 14 | 7.7 | 213 | 10.7 | 7.8 | 64 |
| CC-09 | 3/28/2014 | 202 | 8.5 | 435 | 10.9 | 8.51 | 30 |
| CC-09 | 4/11/2014 | 304 | 14.9 | 762 | 8.7 | 8.44 | 36 |
| CC-09 | 5/15/2014 | 2462 | 17.1 | 933 | 7.8 | 8.16 | 25 |
| CC-09 | 6/17/2014 | 304 | 19.7 | 1918 | 7.8 | 8.23 | 28 |
| CC-09 | 7/21/2014 | 3069 | 17.5 | 514 | 8.8 | 8.34 | 36 |
| CC-09 | 8/12/2014 | 409 | 19.7 | 415 | 8.1 | 8.16 | 33 |
| CC-09 | 1/15/2016 | 206 | 3.1 | 470 | 12.7 | 8.1 | 4 |
| CC-09 | 2/12/2016 | 95 | 3.1 | ND | ND | ND | ND |
| CC-09 | 3/18/2016 | 94 | 9.5 | 418 | 10.5 | 8 | 8 |
| CC-09 | 4/13/2016 | 55 | 14.9 | 762 | 8.7 | 8.4 | 14 |

Table B.10. Water Quality Data for Callahan Creek Water Samples, CC-10

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-10 | 8/20/2012 | 0 | 16.06 | 560 | 9.3 | 8.32 | ND |
| CC-10 | 9/20/2012 | 0 | 14.89 | 610 | 9.6 | 8.17 | ND |
| CC-10 | 10/18/2012 | 3405 | 10.28 | 588 | 11.1 | 8.35 | ND |
| CC-10 | 11/15/2012 | 0 | 5.61 | 408 | 12.8 | 8.42 | ND |
| CC-10 | 12/13/2012 | 0 | 3.44 | 312 | 15.1 | 8.24 | ND |
| CC-10 | 1/11/2013 | 0 | 9.11 | 412 | 10.1 | 8.06 | ND |
| CC-10 | 2/14/2013 | 0 | 5.89 | 360 | 12.5 | 8.36 | ND |
| CC-10 | 3/21/2013 | 0 | 2.67 | 275 | 13.7 | 8.25 | ND |
| CC-10 | 4/25/2013 | 0 | 10.1 | 389 | 10.3 | 8.4 | ND |
| CC-10 | 5/29/2013 | 0 | 16.3 | 507 | 6.9 | 7.91 | ND |
| CC-10 | 6/24/2013 | 306 | 17.9 | 597 | 6.8 | 7.78 | ND |
| CC-10 | 7/22/2013 | 13 | 19.3 | 579 | 6.3 | 7.81 | ND |
| CC-10 | 8/19/2013 | 0 | 18.3 | 731 | 6.5 | 7.81 | ND |
| CC-10 | 9/25/2013 | 202 | 15.6 | 708 | 9.6 | 7.89 | 24 |
| CC-10 | 10/23/2013 | 0 | 10.1 | 652 | 4.9 | 7.93 | 22 |
| CC-10 | 11/13/2013 | 0 | 2.8 | 524 | 7.7 | 8 | 20 |
| CC-10 | 12/12/2013 | 0 | 4.9 | 370 | 10.4 | 7.4 | 30 |
| CC-10 | 1/16/2014 | 99 | 3.2 | 291 | 15.8 | 7.48 | 30 |
| CC-10 | 2/21/2014 | 10 | 7.7 | 211 | 10.8 | 8.52 | 28 |
| CC-10 | 3/28/2014 | 8 | 8.2 | 345 | 10.8 | 8.21 | 23 |
| CC-10 | 4/11/2014 | 1 | 14 | 352 | 9.3 | 8.19 | 23 |
| CC-10 | 5/15/2014 | 63 | 16.6 | 352 | 8.4 | 7.92 | 25 |
| CC-10 | 6/17/2014 | 8 | 18.1 | 527 | 8.1 | 8 | 18 |
| CC-10 | 7/21/2014 | 13 | 17.1 | 327 | 8.8 | 7.97 | 30 |
| CC-10 | 8/12/2014 | 260 | 19.7 | 398 | 8 | 7.93 | 33 |
| CC-10 | 1/15/2016 | 0 | 2.7 | 398 | 12.6 | 8.1 | 7 |
| CC-10 | 2/12/2016 | 5 | 2.7 | ND | ND | ND | ND |
| CC-10 | 3/18/2016 | 1 | 9.2 | 393 | 10.8 | 8.1 | 8 |
| CC-10 | 4/13/2016 | 0 | 14 | 352 | 9.3 | 8.1 | 9 |

Table B.11. Water Quality Data for Callahan Creek Water Samples, CC-12

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-12 | 8/20/2012 | 626 | 15.56 | 830 | 10.4 | 8.59 | ND |
| CC-12 | 9/20/2012 | 2135 | 13.67 | 750 | 10.2 | 8.5 | ND |
| CC-12 | 10/18/2012 | 0 | 10.72 | 711 | 11.1 | 8.57 | ND |
| CC-12 | 11/15/2012 | 2011 | 6.06 | 517 | 13.6 | 8.75 | ND |
| CC-12 | 12/13/2012 | 201 | 4.44 | 413 | 13.2 | 8.56 | ND |
| CC-12 | 1/11/2013 | 2590 | 10.06 | 569 | 9.9 | 8.46 | ND |
| CC-12 | 2/14/2013 | 409 | 8.11 | 567 | 11.9 | 8.66 | ND |
| CC-12 | 3/21/2013 | 521 | 4.39 | 375 | 13.1 | 8.55 | ND |
| CC-12 | 4/25/2013 | 1211 | 11 | 514 | 10.2 | 8.73 | ND |
| CC-12 | 5/29/2013 | 2157 | 16.6 | 760 | 7.2 | 8.15 | ND |
| CC-12 | 6/24/2013 | 5291 | 17.4 | 732 | 11.4 | 8.08 | ND |
| CC-12 | 7/22/2013 | 521 | 18.3 | 758 | 6.8 | 8.1 | ND |
| CC-12 | 8/19/2013 | 626 | 17.5 | 917 | 6.7 | 8.19 | ND |
| CC-12 | 9/25/2013 | 413 | 15.4 | 872 | 6.9 | 8.13 | 14 |
| CC-12 | 10/23/2013 | 4711 | 10.5 | 806 | 8.4 | 8.09 | 10 |
| CC-12 | 11/13/2013 | 2011 | 5.1 | 691 | 8 | 8.29 | 17 |
| CC-12 | 12/12/2013 | 201 | 6 | 438 | 14.8 | 7.65 | 20 |
| CC-12 | 1/16/2014 | 99 | 5 | 417 | 15.1 | 7.92 | 20 |
| CC-12 | 2/21/2014 | 100 | 8.2 | 261 | 10.8 | 7.82 | 23 |
| CC-12 | 3/28/2014 | 1211 | 10 | 506 | 10.2 | 8.23 | 25 |
| CC-12 | 4/11/2014 | 413 | 14.8 | 511 | 9.2 | 8.05 | 20 |
| CC-12 | 5/15/2014 | 10758 | 16.2 | 492 | 8.4 | 8.21 | 15 |
| CC-12 | 6/17/2014 | 413 | 18.2 | 842 | 8.8 | 8.42 | 15 |
| CC-12 | 7/21/2014 | 521 | 17.2 | 495 | 8.9 | 8.16 | 20 |
| CC-12 | 8/12/2014 | 413 | 19.6 | 476 | 8.1 | 8 | 20 |

Table B.12. Water Quality Data for Callahan Creek Water Samples, CC-13

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-13 | 8/20/2012 | 852 | 18.11 | 880 | 9.7 | 8.66 | ND |
| CC-13 | 9/20/2012 | 738 | 15.33 | 750 | 10.1 | 8.69 | ND |
| CC-13 | 10/18/2012 | 413 | 12.06 | 787 | 11.4 | 8.76 | ND |
| CC-13 | 11/15/2012 | 202 | 7.67 | 600 | 13 | 8.79 | ND |
| CC-13 | 12/13/2012 | 0 | 5.11 | 555 | 12.7 | 8.55 | ND |
| CC-13 | 1/11/2013 | 304 | 9.83 | 629 | 10.5 | 8.56 | ND |
| CC-13 | 2/14/2013 | 100 | 8.56 | 601 | 11.9 | 8.75 | ND |
| CC-13 | 3/21/2013 | 1731 | 5.28 | 489 | 13.1 | 8.56 | ND |
| CC-13 | 4/25/2013 | 100 | 11.8 | 604 | 10.1 | 8.81 | ND |
| CC-13 | 5/29/2013 | 0 | 17.2 | 818 | 7.1 | 8.18 | ND |
| CC-13 | 6/24/2013 | 1869 | 17.3 | 799 | 7.2 | 8.13 | ND |
| CC-13 | 7/22/2013 | 100 | 19.5 | 842 | 6.4 | 8.2 | ND |
| CC-13 | 8/19/2013 | 852 | 18.4 | 924 | 8.9 | 8.18 | ND |
| CC-13 | 9/25/2013 | 23593 | 16.8 | 934 | 6.8 | 8.19 | 22 |
| CC-13 | 10/23/2013 | 1613 | 10.7 | 845 | 8.5 | 8.23 | 22 |
| CC-13 | 11/13/2013 | 202 | 4.5 | 702 | 7.6 | 8.3 | 20 |
| CC-13 | 12/12/2013 | 0 | 6.9 | 552 | 15.1 | 7.81 | 30 |
| CC-13 | 1/16/2014 | 78 | 6.1 | 523 | 15.5 | 7.82 | 25 |
| CC-13 | 2/21/2014 | 28 | 9.3 | 398 | 10.4 | 7.69 | 48 |
| CC-13 | 3/28/2014 | 516 | 9.7 | 634 | 10.6 | 8.22 | 23 |
| CC-13 | 4/11/2014 | 100 | 14.9 | 681 | 9.5 | 8.19 | 20 |
| CC-13 | 5/15/2014 | 738 | 15.7 | 620 | 8.7 | 8.02 | 23 |
| CC-13 | 6/17/2014 | 100 | 19.7 | 891 | 8.5 | 8.21 | 15 |
| CC-13 | 7/21/2014 | 100 | 18.1 | 680 | 8.9 | 8.11 | 23 |
| CC-13 | 8/12/2014 | 1596 | 18.7 | 548 | 8.4 | 8.03 | 28 |

Table B.13. Water Quality Data for Callahan Creek Water Samples, CC-13

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-14 | 8/20/2012 | 960 | 18.11 | 890 | 10.4 | 8.68 | ND |
| CC-14 | 9/20/2012 | 1890 | 15.83 | 780 | 10.1 | 8.64 | ND |
| CC-14 | 10/18/2012 | 9338 | 12.5 | 819 | 12.3 | 8.79 | ND |
| CC-14 | 11/15/2012 | 3225 | 8.06 | 615 | 12.7 | 8.81 | ND |
| CC-14 | 12/13/2012 | 100 | 5.61 | 581 | 12.7 | 8.6 | ND |
| CC-14 | 1/11/2013 | 852 | 10.06 | 646 | 10.7 | 8.59 | ND |
| CC-14 | 2/14/2013 | 0 | 8.94 | 624 | 11.7 | 8.74 | ND |
| CC-14 | 3/21/2013 | 3405 | 5.56 | 510 | 12.8 | 8.57 | ND |
| CC-14 | 4/25/2013 | 0 | 11.9 | 617 | 10.5 | 8.8 | ND |
| CC-14 | 5/29/2013 | 306 | 17.1 | 831 | 8.1 | 8.16 | ND |
| CC-14 | 6/24/2013 | 3361 | 17.4 | 809 | 7.2 | 8.11 | ND |
| CC-14 | 7/22/2013 | 202 | 19.4 | 845 | 6.5 | 8.2 | ND |
| CC-14 | 8/19/2013 | 960 | 18.1 | 935 | 6.8 | 8.18 | ND |
| CC-14 | 9/25/2013 | 201 | 17 | 955 | 6.7 | 8.19 | 38 |
| CC-14 | 10/23/2013 | 969 | 11 | 859 | 8.4 | 8.27 | 29 |
| CC-14 | 11/13/2013 | 3225 | 6 | 755 | 7.1 | 8.26 | 23 |
| CC-14 | 12/12/2013 | 100 | 7.2 | 576 | 12.7 | 7.77 | 51 |
| CC-14 | 1/16/2014 | 409 | 6.5 | 551 | 15.1 | 7.93 | 40 |
| CC-14 | 2/21/2014 | 9 | 9.4 | 429 | 10.4 | 7.7 | 61 |
| CC-14 | 3/28/2014 | 852 | 9.9 | 650 | 10.5 | 8.22 | 38 |
| CC-14 | 4/11/2014 | 202 | 14.8 | 687 | 9.6 | 8.24 | 33 |
| CC-14 | 5/15/2014 | 202 | 15.6 | 627 | 8.7 | 8 | 41 |
| CC-14 | 6/17/2014 | 100 | 19.5 | 827 | 8.5 | 8.2 | 61 |
| CC-14 | 7/21/2014 | 202 | 18.1 | 713 | 8.9 | 8.08 | 43 |
| CC-14 | 8/12/2014 | 1211 | 18.5 | 588 | 8.5 | 7.98 | 43 |

Table B.14. Water Quality Data for Callahan Creek Water Samples, CC-15

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| CC-15 | 8/20/2012 | 0 | 17.89 | 930 | 9.4 | 8.33 | ND |
| CC-15 | 9/20/2012 | 0 | 15.94 | 820 | 9.6 | 8.33 | ND |
| CC-15 | 10/18/2012 | 100 | 13.06 | 883 | 10.4 | 8.43 | ND |
| CC-15 | 11/15/2012 | 0 | 8.5 | 647 | 12.3 | 8.53 | ND |
| CC-15 | 12/13/2012 | 0 | 6.94 | 640 | 11.9 | 8.45 | ND |
| CC-15 | 1/11/2013 | 0 | 10.11 | 681 | 10.9 | 8.43 | ND |
| CC-15 | 2/14/2013 | 0 | 8.94 | 658 | 11.4 | 8.54 | ND |
| CC-15 | 3/21/2013 | 0 | 6.39 | 545 | 13.6 | 8.41 | ND |
| CC-15 | 4/25/2013 | 0 | 12.2 | 653 | 10.3 | 8.53 | ND |
| CC-15 | 5/29/2013 | 0 | 17 | 854 | 6.9 | 7.93 | ND |
| CC-15 | 6/24/2013 | 100 | 17.3 | 843 | 6.9 | 7.82 | ND |
| CC-15 | 7/22/2013 | 12 | 19.2 | 895 | 6.3 | 7.92 | ND |
| CC-15 | 8/19/2013 | 0 | 17.9 | 968 | 6.6 | 7.91 | ND |
| CC-15 | 9/25/2013 | 0 | 17.2 | 981 | 6.6 | 8.01 | 23 |
| CC-15 | 10/23/2013 | 0 | 11.8 | 907 | 7.3 | 8.03 | 8 |
| CC-15 | 11/13/2013 | 0 | 8.1 | 824 | 6.5 | 8.14 | 15 |
| CC-15 | 12/12/2013 | 0 | 7.6 | 604 | 14.7 | 7.6 | 28 |
| CC-15 | 1/16/2014 | 1 | 7.2 | 592 | 14.8 | 7.79 | 30 |
| CC-15 | 2/21/2014 | 8 | 9.5 | 454 | 10.3 | 7.73 | 51 |
| CC-15 | 3/28/2014 | 5 | 10 | 685 | 10 | 8.15 | 18 |
| CC-15 | 4/11/2014 | 0 | 14.5 | 728 | 9.1 | 8.07 | 18 |
| CC-15 | 5/15/2014 | 156 | 15.5 | 643 | 8.8 | 7.91 | 48 |
| CC-15 | 6/17/2014 | 4 | 19.5 | 961 | 8 | 8.08 | 13 |
| CC-15 | 7/21/2014 | 12 | 18.3 | 782 | 8.8 | 7.98 | 43 |
| CC-15 | 8/12/2014 | 210 | 18.8 | 688 | 8.1 | 7.93 | 36 |

## Appendix C. Water Quality Data Roaring Fork

Table C.1. Water Quality Data for Roaring Fork Water Samples, RF-01

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-01 | 8/20/2012 | 100 | 17.39 | 1020 | 10 | 8.58 | ND |
| RF-01 | 9/20/2012 | 100 | 14.78 | 890 | 10.4 | 8.51 | ND |
| RF-01 | 10/18/2012 | 100 | 11.61 | 874 | 11.5 | 8.67 | ND |
| RF-01 | 11/15/2012 | 0 | 6 | 740 | 12.4 | 8.44 | ND |
| RF-01 | 12/13/2012 | 0 | 6.17 | 636 | 15.8 | 8.61 | ND |
| RF-01 | 1/11/2013 | 100 | 10.22 | 779 | 8.9 | 8.62 | ND |
| RF-01 | 2/14/2013 | 100 | 6.83 | 715 | 10.2 | 8.6 | ND |
| RF-01 | 3/21/2013 | 99 | 4.94 | 587 | 13.8 | 8.6 | ND |
| RF-01 | 4/25/2013 | 202 | 10.2 | 713 | 12.5 | 7.93 | ND |
| RF-01 | 5/29/2013 | 0 | 18.3 | 1096 | 11.2 | 8.17 | ND |
| RF-01 | 6/24/2013 | 4874 | 17.4 | 1045 | 7.3 | 7.95 | ND |
| RF-01 | 7/22/2013 | 100 | 18.9 | 1047 | 4.5 | 8.11 | ND |
| RF-01 | 8/19/2013 | 100 | 19 | 1151 | 2.9 | 8.2 | ND |
| RF-01 | 9/25/2013 | 0 | 15.7 | 1124 | 6.8 | 8.15 | 14 |
| RF-01 | 10/23/2013 | 745 | 10.7 | 1054 | 6 | 8.22 | 13 |
| RF-01 | 11/13/2013 | 0 | 4.6 | 885 | 7.3 | 8.33 | 10 |
| RF-01 | 12/12/2013 | 0 | 0 | 662 | 14.8 | 7.85 | 28 |
| RF-01 | 1/16/2014 | 23 | 5.3 | 647 | 13.2 | 7.95 | 24 |
| RF-01 | 2/21/2014 | 28 | 9.5 | 518 | 10.9 | 7.99 | 71 |
| RF-01 | 3/28/2014 | 100 | 10.1 | 786 | 10.2 | 8.29 | 64 |
| RF-01 | 4/11/2014 | 100 | 11.5 | 801 | 10.5 | 8.19 | 56 |
| RF-01 | 5/15/2014 | 100 | 17.3 | 905 | 8.2 | 7.8 | 41 |
| RF-01 | 6/17/2014 | 0 | 21.1 | 1137 | 8.2 | 8.03 | 18 |
| RF-01 | 7/21/2014 | 100 | 18.5 | 891 | 9 | 8.05 | 56 |
| RF-01 | 8/12/2014 | 3267 | 19.4 | 655 | 8 | 8.01 | 15 |

Table C.2. Water Quality Data for Roaring Fork Water Samples, RF-02

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-02 | 8/20/2012 | 306 | 17.33 | 1040 | 10 | 8.43 | ND |
| RF-02 | 9/20/2012 | 100 | 14.72 | 900 | 10.1 | 8.4 | ND |
| RF-02 | 10/18/2012 | 100 | 11.39 | 888 | 10.4 | 8.34 | ND |
| RF-02 | 11/15/2012 | 0 | 6.17 | 749 | 12.3 | 8.42 | ND |
| RF-02 | 12/13/2012 | 100 | 6.56 | 651 | 12.9 | 8.49 | ND |
| RF-02 | 1/11/2013 | 304 | 10.17 | 676 | 9.4 | 8.45 | ND |
| RF-02 | 2/14/2013 | 202 | 6.72 | 710 | 9.9 | 8.55 | ND |
| RF-02 | 3/21/2013 | 0 | 3.28 | 597 | 13.8 | 8.46 | ND |
| RF-02 | 4/25/2013 | 0 | 10.2 | 725 | 10.5 | 8.44 | ND |
| RF-02 | 5/29/2013 | 0 | 18.2 | 1105 | 7.1 | 8.08 | ND |
| RF-02 | 6/24/2013 | 3451 | 17.1 | 1046 | 7.2 | 7.97 | ND |
| RF-02 | 7/22/2013 | 100 | 18.7 | 1058 | 6.6 | 8.01 | ND |
| RF-02 | 8/19/2013 | 306 | 18.9 | 1170 | 4.3 | 8.09 | ND |
| RF-02 | 9/25/2013 | 100 | 15.6 | 1130 | 1.6 | 7.8 | 29 |
| RF-02 | 10/23/2013 | 1579 | 10.7 | 1067 | 6.6 | 8.06 | 38 |
| RF-02 | 11/13/2013 | 0 | 5.1 | 903 | 6.1 | 8.12 | 46 |
| RF-02 | 12/12/2013 | 100 | 6.2 | 675 | 15.9 | 7.61 | 51 |
| RF-02 | 1/16/2014 | 55 | 5.6 | 659 | 15.1 | 7.9 | 53 |
| RF-02 | 2/21/2014 | 36 | 9.6 | 531 | 10.3 | 7.79 | 64 |
| RF-02 | 3/28/2014 | 202 | 10.1 | 828 | 9.6 | 8.04 | 36 |
| RF-02 | 4/11/2014 | 100 | 11.7 | 816 | 10.3 | 8.08 | 43 |
| RF-02 | 5/15/2014 | 100 | 17.2 | 877 | 8 | 7.93 | 38 |
| RF-02 | 6/17/2014 | 100 | 20.3 | 1110 | 8.4 | 8.11 | 36 |
| RF-02 | 7/21/2014 | 100 | 18.5 | 908 | 8.7 | 8.08 | 43 |
| RF-02 | 8/12/2014 | 413 | 19.3 | 664 | 8 | 8.06 | 56 |
| RF-02 | 1/15/2016 | 31.68 | 4.8 | 794 | 12.2 | 8.1 | 12.2 |
| RF-02 | 2/12/2016 | 9.436 | ND | ND | ND | ND | ND |
| RF-02 | 3/18/2016 | 21.4 | 10.4 | 826 | 10.5 | 8.1 | 17 |
| RF-02 | 4/13/2016 | 63.17 | 11.7 | 816 | 10.3 | 8 | 25 |

Table C.3. Water Quality Data for Roaring Fork Water Samples, RF-03

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-03 | 8/20/2012 | 202 | 16.83 | 1020 | 9.9 | 8.41 | ND |
| RF-03 | 9/20/2012 | 99 | 15 | 870 | 9.9 | 8.41 | ND |
| RF-03 | 10/18/2012 | 306 | 11.33 | 861 | 10.3 | 8.33 | ND |
| RF-03 | 11/15/2012 | 0 | 6 | 734 | 12.6 | 8.5 | ND |
| RF-03 | 12/13/2012 | 0 | 6.17 | 594 | 12.8 | 8.43 | ND |
| RF-03 | 1/11/2013 | 516 | 10.06 | 777 | 9.6 | 8.46 | ND |
| RF-03 | 2/14/2013 | 100 | 7.06 | 722 | 9.8 | 8.51 | ND |
| RF-03 | 3/21/2013 | 99 | 1.17 | 590 | 13 | 8.41 | ND |
| RF-03 | 4/25/2013 | 0 | 10.3 | 722 | 10.1 | 8.45 | ND |
| RF-03 | 5/29/2013 | 0 | 18.1 | 1099 | 7.1 | 8.05 | ND |
| RF-03 | 6/24/2013 | 5461 | 17.1 | 1050 | 7.2 | 7.97 | ND |
| RF-03 | 7/22/2013 | 100 | 18.9 | 1074 | 6.5 | 7.99 | ND |
| RF-03 | 8/19/2013 | 202 | 18.9 | 1182 | 4.7 | 8.07 | ND |
| RF-03 | 9/25/2013 | 0 | 15.7 | 1147 | 4.3 | 7.99 | 42 |
| RF-03 | 10/23/2013 | 521 | 10.4 | 1064 | 7.2 | 8.04 | 46 |
| RF-03 | 11/13/2013 | 0 | 5.1 | 904 | 6.1 | 8.12 | 20 |
| RF-03 | 12/12/2013 | 0 | 6.2 | 682 | 11.9 | 7.69 | 56 |
| RF-03 | 1/16/2014 | 33 | 5.5 | 664 | 14.3 | 7.73 | 58 |
| RF-03 | 2/21/2014 | 100 | 9.6 | 535 | 10.2 | 7.8 | 61 |
| RF-03 | 3/28/2014 | 100 | 10 | 818 | 9.7 | 7.91 | 66 |
| RF-03 | 4/11/2014 | 100 | 12 | 776 | 9.9 | 8.05 | 64 |
| RF-03 | 5/15/2014 | 100 | 17.3 | 897 | 8 | 7.94 | 61 |
| RF-03 | 6/17/2014 | 100 | 20.1 | 1070 | 8.2 | 8.12 | 61 |
| RF-03 | 7/21/2014 | 100 | 18.4 | 916 | 8.6 | 8.07 | 76 |
| RF-03 | 8/12/2014 | 860 | 19.2 | 712 | 8 | 8.02 | 84 |

Table C.4. Water Quality Data for Roaring Fork Water Samples, RF-04

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-04 | 8/20/2012 | 304 | 16.22 | 1040 | 9.9 | 8.46 | ND |
| RF-04 | 9/20/2012 | 100 | 14.83 | 970 | 10.2 | 8.46 | ND |
| RF-04 | 10/18/2012 | 304 | 11.33 | 922 | 10.3 | 8.38 | ND |
| RF-04 | 11/15/2012 | 0 | 6.17 | 777 | 12.4 | 8.43 | ND |
| RF-04 | 12/13/2012 | 100 | 5.94 | 678 | 12.6 | 8.48 | ND |
| RF-04 | 1/11/2013 | 100 | 10.22 | 807 | 9.6 | 8.48 | ND |
| RF-04 | 2/14/2013 | 100 | 7.28 | 740 | 11.3 | 8.52 | ND |
| RF-04 | 3/21/2013 | 100 | 5.17 | 618 | 12.7 | 8.42 | ND |
| RF-04 | 4/25/2013 | 0 | 10.2 | 739 | 9.9 | 8.48 | ND |
| RF-04 | 5/29/2013 | 0 | 17.5 | 1107 | 7 | 8.08 | ND |
| RF-04 | 6/24/2013 | 5371 | 16.8 | 1068 | 7.3 | 7.99 | ND |
| RF-04 | 7/22/2013 | 0 | 18.4 | 1093 | 6.7 | 8 | ND |
| RF-04 | 8/19/2013 | 304 | 18.6 | 1197 | 5.8 | 8.09 | ND |
| RF-04 | 9/25/2013 | 0 | 15.6 | 1174 | 4.6 | 8.07 | 36 |
| RF-04 | 10/23/2013 | 413 | 10.2 | 1092 | 4.9 | 8.11 | 41 |
| RF-04 | 11/13/2013 | 0 | 4.4 | 916 | 7.7 | 8.11 | 39 |
| RF-04 | 12/12/2013 | 100 | 6.4 | 723 | 14.8 | 7.72 | 61 |
| RF-04 | 1/16/2014 | 32 | 5.9 | 695 | 14.2 | 7.76 | 51 |
| RF-04 | 2/21/2014 | 100 | 9.8 | 553 | 10 | 7.72 | 64 |
| RF-04 | 3/28/2014 | 100 | 10.2 | 849 | 9.7 | 7.86 | 51 |
| RF-04 | 4/11/2014 | 100 | 12 | 847 | 9.9 | 8.01 | 41 |
| RF-04 | 5/15/2014 | 306 | 16.9 | 927 | 8.2 | 7.96 | 51 |
| RF-04 | 6/17/2014 | 202 | 19.3 | 1134 | 8.2 | 8.09 | 37 |
| RF-04 | 7/21/2014 | 0 | 18 | 965 | 8.4 | 8.04 | 58 |
| RF-04 | 8/12/2014 | 852 | 18.9 | 720 | 8.2 | 7.99 | 61 |

Table C.5. Water Quality Data for Roaring Fork Water Samples, RF-05

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-05 | 8/20/2012 | 100 | 16.67 | 1070 | 9.6 | 8.37 | ND |
| RF-05 | 9/20/2012 | 99 | 14.72 | 980 | 10.1 | 8.37 | ND |
| RF-05 | 10/18/2012 | 201 | 11.33 | 936 | 10.1 | 8.3 | ND |
| RF-05 | 11/15/2012 | 100 | 6.5 | 795 | 11.9 | 8 | ND |
| RF-05 | 12/13/2012 | 100 | 7.17 | 727 | 12.2 | 8.44 | ND |
| RF-05 | 1/11/2013 | 409 | 10.39 | 828 | 10.2 | 8.34 | ND |
| RF-05 | 2/14/2013 | 100 | 7.72 | 759 | 10.9 | 8.37 | ND |
| RF-05 | 3/21/2013 | 413 | 5.72 | 636 | 17.9 | 8.32 | ND |
| RF-05 | 4/25/2013 | 0 | 10.5 | 752 | 12.4 | 8.33 | ND |
| RF-05 | 5/29/2013 | 521 | 18.5 | 1144 | 6.8 | 7.97 | ND |
| RF-05 | 6/24/2013 | 17247 | 16.7 | 1079 | 7.1 | 7.9 | ND |
| RF-05 | 7/22/2013 | 100 | 18.4 | 1111 | 6.6 | 7.9 | ND |
| RF-05 | 8/19/2013 | 100 | 18.6 | 1023 | 5.1 | 8.01 | ND |
| RF-05 | 9/25/2013 | 202 | 15.5 | 1190 | 4.9 | 8 | 19 |
| RF-05 | 10/23/2013 | ND | 10.6 | 1109 | 7.4 | 8.01 | 23 |
| RF-05 | 11/13/2013 | 100 | 4.8 | 937 | 6.6 | 7.97 | 18 |
| RF-05 | 12/12/2013 | 100 | 7.2 | 752 | 13.6 | 7.67 | 38 |
| RF-05 | 1/16/2014 | 3361 | 6.6 | 719 | 14.8 | 7.67 | 34 |
| RF-05 | 2/21/2014 | 202 | 10.3 | 567 | 10 | 7.68 | 43 |
| RF-05 | 3/28/2014 | 100 | 10.2 | 865 | 9.6 | 7.82 | 28 |
| RF-05 | 4/11/2014 | 100 | 12.2 | 862 | 9.7 | 8.04 | 41 |
| RF-05 | 5/15/2014 | 409 | 16.7 | 950 | 8.1 | 7.88 | 28 |
| RF-05 | 6/17/2014 | 100 | 20.3 | 1187 | 8 | 8.03 | 36 |
| RF-05 | 7/21/2014 | 100 | 18.6 | 989 | 8.5 | 8.01 | 51 |
| RF-05 | 8/12/2014 | 306 | 19.5 | 777 | 8.1 | 7.86 | 20 |

Table C.6. Water Quality Data for Roaring Fork Water Samples, RF-06

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-06 | 8/20/2012 | 3405 | 17.83 | 1090 | 9.5 | 8.45 | ND |
| RF-06 | 9/20/2012 | 0 | 15.67 | 1020 | 9.8 | 8.43 | ND |
| RF-06 | 10/18/2012 | 0 | 11.06 | 967 | 10.7 | 8.34 | ND |
| RF-06 | 11/15/2012 | 304 | 6.5 | 779 | 13.2 | 8.42 | ND |
| RF-06 | 12/13/2012 | 202 | 8 | 782 | 11.8 | 8.5 | ND |
| RF-06 | 1/11/2013 | 1211 | 10.22 | 845 | 11.5 | 8.42 | ND |
| RF-06 | 2/14/2013 | 100 | 8.67 | 738 | 10.6 | 8.33 | ND |
| RF-06 | 3/21/2013 | 738 | 6.67 | 583 | 12.3 | 8.13 | ND |
| RF-06 | 4/25/2013 | 0 | 11.1 | 677 | 9.3 | 8.12 | ND |
| RF-06 | 5/29/2013 | 979 | 18.4 | 1120 | 6.3 | 7.7 | ND |
| RF-06 | 6/24/2013 | 7712 | 16.7 | 979 | 6.7 | 7.72 | ND |
| RF-06 | 7/22/2013 | 100 | 18.7 | 1066 | 6.4 | 7.79 | ND |
| RF-06 | 8/19/2013 | 3405 | 17.9 | 1045 | 5.4 | 7.72 | ND |
| RF-06 | 9/25/2013 | 100 | 15.2 | 1093 | 5.3 | 7.78 | 29 |
| RF-06 | 10/23/2013 | 1310 | 11.2 | 970 | 7.4 | 7.82 | 30 |
| RF-06 | 11/13/2013 | 304 | 6.8 | 911 | 6 | 7.85 | 29 |
| RF-06 | 12/12/2013 | 202 | 7.4 | 690 | 12.9 | 7.58 | 53 |
| RF-06 | 1/16/2014 | 5731 | 8.1 | 631 | 13.7 | 7.36 | 44 |
| RF-06 | 2/21/2014 | 56 | 10.4 | 410 | 9.9 | 7.69 | 66 |
| RF-06 | 3/28/2014 | 100 | 10.8 | 685 | 8.8 | 7.76 | 38 |
| RF-06 | 4/11/2014 | 100 | 12.9 | 701 | 9.2 | 7.67 | 41 |
| RF-06 | 5/15/2014 | 2482 | 16.3 | 767 | 8 | 7.78 | 38 |
| RF-06 | 6/17/2014 | 306 | 19.3 | 871 | 8.1 | 7.92 | 33 |
| RF-06 | 7/21/2014 | 100 | 18.7 | 810 | 7.9 | 7.99 | 43 |
| RF-06 | 8/12/2014 | 202 | 19.1 | 584 | 7.9 | 7.93 | 20 |
| RF-06 | 1/15/2016 | 117 | 5.4 | 858 | 10.9 | 7.9 | 22 |
| RF-06 | 2/12/2016 | 55 | ND | ND | ND | ND | ND |
| RF-06 | 3/18/2016 | 85 | 5.4 | 858 | 10.9 | 7.9 | 22 |
| RF-06 | 4/13/2016 | 46 | 12.9 | 701 | 9.2 | 7.6 | 9 |

Table C.7. Water Quality Data for Roaring Fork Water Samples, RF-07

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-07 | 8/20/2012 | 0 | 17.78 | 1100 | 9.6 | 8.44 | ND |
| RF-07 | 9/20/2012 | 100 | 15.67 | 1010 | 9.7 | 8.43 | ND |
| RF-07 | 10/18/2012 | 0 | 11 | 968 | 10.3 | 8.35 | ND |
| RF-07 | 11/15/2012 | 0 | 6.56 | 788 | 12.1 | 8.4 | ND |
| RF-07 | 12/13/2012 | 0 | 8 | 782 | 11.8 | 8.53 | ND |
| RF-07 | 1/11/2013 | 0 | 10.17 | 856 | 10.1 | 8.47 | ND |
| RF-07 | 2/14/2013 | 0 | 7.33 | 790 | 11.5 | 8.6 | ND |
| RF-07 | 3/21/2013 | 0 | 5.94 | 679 | 12.6 | 8.49 | ND |
| RF-07 | 4/25/2013 | 202 | 10.4 | 836 | 9.6 | 8.61 | ND |
| RF-07 | 5/29/2013 | 0 | 20 | 1227 | 6.6 | 8.04 | ND |
| RF-07 | 6/24/2013 | 201 | 17.2 | 1105 | 7 | 7.99 | ND |
| RF-07 | 7/22/2013 | 12 | 19.2 | 1196 | 6.4 | 8.01 | ND |
| RF-07 | 8/19/2013 | 0 | 19.6 | 1259 | 5.2 | 8.01 | ND |
| RF-07 | 9/25/2013 | 100 | 15.9 | 1222 | 5.3 | 7.98 | 19 |
| RF-07 | 10/23/2013 | 0 | 10.6 | 1116 | 7.9 | 8.05 | 23 |
| RF-07 | 11/13/2013 | 0 | 6.2 | 804 | 6.6 | 8.02 | 24 |
| RF-07 | 12/12/2013 | 0 | 7.7 | 780 | 12.4 | 7.77 | 25 |
| RF-07 | 1/16/2014 | 32 | 6.6 | 776 | 15.6 | 7.84 | 27 |
| RF-07 | 2/21/2014 | 16 | 10.5 | 694 | 10 | 7.88 | 48 |
| RF-07 | 3/28/2014 | 0 | 9.9 | 950 | 9.7 | 7.78 | 18 |
| RF-07 | 4/11/2014 | 1 | 12.7 | 963 | 9.7 | 7.85 | 23 |
| RF-07 | 5/15/2014 | 0 | 16.6 | 1019 | 8.6 | 7.89 | 25 |
| RF-07 | 6/17/2014 | 2 | 22.3 | 1331 | 7.8 | 8 | 18 |
| RF-07 | 7/21/2014 | 12 | 19.1 | 1073 | 8.4 | 8.03 | 53 |
| RF-07 | 8/12/2014 | 228 | 19.8 | 874 | 8 | 7.95 | 46 |
| RF-07 | 1/15/2016 | 4 | 5.5 | 895 | 11.8 | 8.2 | 10 |
| RF-07 | 2/12/2016 | 0 | ND | ND | ND | ND | ND |
| RF-07 | 3/18/2016 | 3 | 10.7 | 956 | 10.5 | 8 | 8 |
| RF-07 | 4/13/2016 | 2 | 12.7 | 963 | 9.7 | 7.8 | 9 |

Table C.8. Water Quality Data for Roaring Fork Water Samples, RF-08

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-08 | 8/20/2012 | 3893 | 17.44 | 990 | 9.6 | 8.48 | ND |
| RF-08 | 9/20/2012 | 2917 | 15.94 | 570 | 10.2 | 8.37 | ND |
| RF-08 | 10/18/2012 | 413 | 10.22 | 675 | 10.4 | 8.21 | ND |
| RF-08 | 11/15/2012 | 860 | 5.44 | 738 | 12.7 | 8.38 | ND |
| RF-08 | 12/13/2012 | 100 | 7.56 | 571 | 12.2 | 8.46 | ND |
| RF-08 | 1/11/2013 | 1579 | 9.78 | 717 | 11.4 | 8.49 | ND |
| RF-08 | 2/14/2013 | 626 | 6.67 | 604 | 11.4 | 8.57 | ND |
| RF-08 | 3/21/2013 | 304 | 4.56 | 463 | 13.1 | 8.49 | ND |
| RF-08 | 4/25/2013 | 306 | 9.5 | 575 | 9.95 | 8.57 | ND |
| RF-08 | 5/29/2013 | 2109 | 19 | 979 | 6.8 | 8.12 | ND |
| RF-08 | 6/24/2013 | 20459 | 16.9 | 966 | 6.9 | 8.02 | ND |
| RF-08 | 7/22/2013 | 100 | 19.9 | 909 | 7.5 | 8.02 | ND |
| RF-08 | 8/19/2013 | 3893 | 19.6 | 1113 | 5.4 | 8.11 | ND |
| RF-08 | 9/25/2013 | 2405 | 15.8 | 1136 | 5.5 | 8.08 | 17 |
| RF-08 | 10/23/2013 | 9594 | 10 | 978 | 8.3 | 8.07 | 15 |
| RF-08 | 11/13/2013 | 860 | 6.7 | 890 | 6.5 | 8.21 | 15 |
| RF-08 | 12/12/2013 | 100 | 7.4 | 682 | 11.6 | 7.83 | 30 |
| RF-08 | 1/16/2014 | 1323 | 6.4 | 580 | 14.8 | 7.94 | 25 |
| RF-08 | 2/21/2014 | 25 | 9.9 | 360 | 10.3 | 7.91 | 43 |
| RF-08 | 3/28/2014 | 100 | 8.9 | 641 | 10.1 | 7.89 | 25 |
| RF-08 | 4/11/2014 | 201 | 12.5 | 619 | 9.8 | 8.07 | 23 |
| RF-08 | 5/15/2014 | 2420 | 16.7 | 733 | 8 | 7.97 | 23 |
| RF-08 | 6/17/2014 | 100 | 21.6 | 907 | 7.7 | 8.1 | 15 |
| RF-08 | 7/21/2014 | 100 | 19.5 | 782 | 8.1 | 8.1 | 25 |
| RF-08 | 8/12/2014 | 413 | 19.3 | 531 | 8 | 7.98 | 28 |
| RF-08 | 1/15/2016 | 55 | 3.9 | 639 | 11.9 | 8.3 | 10 |
| RF-08 | 2/12/2016 | 242 | ND | ND | ND | ND | ND |
| RF-08 | 3/18/2016 | 9 | 11.3 | 603 | 10.7 | 8 | 9 |
| RF-08 | 4/13/2016 | 239 | 12.5 | 619 | 9.8 | 8 | 16 |

Table C.9. Water Quality Data for Roaring Fork Water Samples, RF-09

| Site ID | Date | *E. coli* (MPN/100 mL) | Temp. (°C) | Cond. (µS/cm) | DO (mg/L) | pH | Stream depth (in.) |
|---|---|---|---|---|---|---|---|
| RF-09 | 8/20/2012 | 202 | 18.78 | 1150 | 9.2 | 8.51 | ND |
| RF-09 | 9/20/2012 | 0 | 15.5 | 790 | 9.7 | 8.37 | ND |
| RF-09 | 10/18/2012 | 0 | 9.94 | 1041 | 10.8 | 8.34 | ND |
| RF-09 | 11/15/2012 | 0 | 5.28 | 869 | 12.8 | 8.46 | ND |
| RF-09 | 12/13/2012 | 0 | 6.61 | 651 | 12.6 | 8.47 | ND |
| RF-09 | 1/11/2013 | 0 | 9.72 | 820 | 10.3 | 8.54 | ND |
| RF-09 | 2/14/2013 | 0 | 6.22 | 717 | 11.6 | 8.54 | ND |
| RF-09 | 3/21/2013 | 0 | 4.56 | 552 | 13 | 8.5 | ND |
| RF-09 | 4/25/2013 | 100 | 9.8 | 709 | 9.65 | 8.61 | ND |
| RF-09 | 5/29/2013 | 100 | 20.8 | 1148 | 6.3 | 8.12 | ND |
| RF-09 | 6/24/2013 | 201 | 17.2 | 1079 | 7 | 8.04 | ND |
| RF-09 | 7/22/2013 | 6 | 20.1 | 1119 | 6.2 | 8.08 | ND |
| RF-09 | 8/19/2013 | 202 | 20.3 | 1285 | 5.3 | 8.11 | ND |
| RF-09 | 9/25/2013 | 0 | 16.2 | 1184 | 5.7 | 8.11 | 43 |
| RF-09 | 10/23/2013 | 0 | 9.8 | 1086 | 10 | 8.12 | 28 |
| RF-09 | 11/13/2013 | 0 | 5.3 | 959 | 7.1 | 8.1 | 33 |
| RF-09 | 12/12/2013 | 0 | 7.5 | 769 | 12.3 | 7.84 | 38 |
| RF-09 | 1/16/2014 | 0 | 6.4 | 686 | 14.3 | 7.97 | 38 |
| RF-09 | 2/21/2014 | 8 | 10 | 416 | 10.2 | 7.82 | 53 |
| RF-09 | 3/28/2014 | 2 | 8.9 | 784 | 10.1 | 7.84 | 30 |
| RF-09 | 4/11/2014 | 3 | 12.6 | 762 | 9.6 | 8.07 | 41 |
| RF-09 | 5/15/2014 | 457 | 17.6 | 824 | 8.2 | 7.99 | 46 |
| RF-09 | 6/17/2014 | 5 | 23.8 | 1145 | 7.3 | 7.98 | 33 |
| RF-09 | 7/21/2014 | 6 | 20.4 | 929 | 8.1 | 8.06 | 43 |
| RF-09 | 8/12/2014 | 80 | 19.7 | 623 | 8.1 | 7.98 | 28 |
| RF-09 | 1/15/2016 | 0 | 3.6 | 741 | 8.3 | 8.3 | 6 |
| RF-09 | 2/12/2016 | 1 | ND | ND | ND | ND | ND |
| RF-09 | 3/18/2016 | 2 | 12 | 738 | 10.5 | 8.1 | 8 |
| RF-09 | 4/13/2016 | 0 | 12.6 | 762 | 9.6 | 8 | 7 |

## Appendix D Flow Rate in Powell River

Table D.1. Flow Rate in Powell River

| Date | Powell River Stream Flow (cfs) |
|---|---|
| 8/20/2012 | 152 |
| 9/20/2012 | 194 |
| 10/18/2012 | 68 |
| 11/15/2012 | 129 |
| 12/13/2012 | 151 |
| 1/11/2013 | 146 |
| 2/14/2013 | 246 |
| 3/21/2013 | 363 |
| 4/25/2013 | 302 |
| 5/29/2013 | 119 |
| 6/24/2013 | 208 |
| 7/22/2013 | 200 |
| 8/19/2013 | 100 |
| 9/25/2013 | 65 |
| 10/23/2013 | 38 |
| 11/13/2013 | 35 |
| 12/12/2013 | 359 |
| 1/16/2014 | 328 |
| 2/21/2014 | 768 |
| 3/28/2014 | 141 |
| 4/11/2014 | 185 |
| 5/15/2014 | 155 |
| 6/17/2014 | 68 |
| 7/21/2014 | 246 |
| 8/12/2014 | 141 |
| 1/15/2016 | 128 |
| 2/12/2016 | 201 |
| 3/18/2016 | 193 |
| 4/13/2016 | 95 |

[1]Flow Rate is measured by the USGS gauge station 03529500

## Appendix E Stream Site Location

Table E.1. Location of Stream Sites Upstream from Watershed Outlet in Callahan Creek

| Site ID | Location Upstream (mi.) |
|---|---|
| CC-01 | 0 |
| CC-02 | 0.976 |
| CC-03 | 1.7 |
| CC-04 | 2.85 |
| CC-05 | 3.328 |
| CC-06 | 3.346 |
| CC-07 | 3.927 |
| CC-08 | 4.282 |
| CC-09 | 4.604 |
| CC-10 | 4.7 |
| CC-12 | 3.149 |
| CC-13 | 4.122 |
| CC-14 | 4.422 |
| CC-15 | 5.6 |

Table E.2. Location of Stream Sites Upstream from Watershed Outlet in Roaring Fork

| Site ID | Location Upstream (mi.) |
|---|---|
| RF-01 | 0.03 |
| RF-02 | 0.96 |
| RF-03 | 1.65 |
| RF-04 | 2.37 |
| RF-05 | 3.7 |
| RF-06 | 4.88 |
| RF-07 | 4.98 |
| RF-08 | 5.12 |
| RF-09 | 6.11 |