Tracking the Primary Sources of Fecal Pollution in the Recharge and Contributing Zones of Edwards Aquifer in Bexar County, TX using Molecular Tools

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EXECUTIVE SUMMARY

Identifying the sources and spatial-temporal trends of fecal pollution is critical to protecting water quality and implementing adequate pollution control and prevention strategies to mitigate pollution levels and risk to environmental and human health. Karst aquifers, such as the Edwards Aquifer in Bexar County, are known to be vulnerable to human and animal waste contamination, and presently, there is an incomplete understanding of the sources of pathogenic fecal contamination in the Edwards, a source of drinking water to more than 2 million people in south-central Texas. Conventional fecal indicator bacteria (FIB) are unreliable due to widely varying survival rates in the environment, failure to discriminate between sources, and inability to distinguish between fecal bacteria associated with recent contamination events and those adapted to secondary habitats.

The purpose of this study was to apply microbial source tracking (MST) techniques to evaluate fecal pollution inputs within the recharge and contributing zones of the Edwards Aquifer in Bexar County, Texas. MST can identify and quantify the source and species of fecal bacteria by targeting Bacteroidales 16S rRNA genes using polymerase chain reaction (PCR) techniques. Bacteroidales do not have the ambiguous survival and sourcing issues present with conventional FIBs and can provide good spatial-temporal resolution. Quantitative PCR-based methods for three general fecal markers (BacUni, *E. coli* and Entero1), five animal-associated assays (BacCow, BacCan, Chicken/Duck-Bac, Rum2Bac and GFD) and two human-associated markers (BacHum and HF183) were applied to resolve spatial-temporal patterns, seasonal variability, and to identify and quantify the sources of fecal contamination. In addition to these molecular tools, other tests including water temperature, pH, dissolved oxygen (DO) and nitrate/nitrite/ammonia were used to document water quality. The effects of stormwater on marker levels and correlations among the markers and with environmental parameters were also investigated.

Water samples were collected bi-monthly from 20 sites within the recharge and contributing zones of Edwards Aquifer from January 2018 to March 2020. The sites consisted of a combination of three wells, fourteen creek sites and three ponds/lakes. Levels for the three general markers were highest and exhibited a similar spatial and temporal distribution pattern across the sampling sites, and thus, it was determined that the sole use of these markers was not sufficient for specific fecal source identification in the Edwards. Among the animal associated markers, the highest levels were observed for Rum2Bac/BacCow and GFD, suggesting a higher percentage of fecal pollution to be coming from the bovine/ruminant and bird populations in the study area. Other individual sources that encompassed lesser proportions of fecal pollution included dog wastes and human-derived wastes. The predominant sources of fecal contamination identified in the Edwards Aquifer study area were ranked in the decreasing order of their presence: avian including gull, ducks etc. (85%), ruminant including cattle and deer (67%), dog (40%) and human-derived (17%).

The cow/ruminant marker concentrations were higher for rural creek sites due to farms and ranches in the area. Higher levels of canine-derived contamination was observed for pond sites near residential areas and urban creek sites, that could likely be attributed to poor pet waste management practices in the area. The concentrations of the GFD avian marker were relatively higher at all surface water sites (except for well sites) suggesting that bird fecal pollution is a major source of concern for Edwards Aquifer. The Chicken/Duck-Bac marker was consistently higher in the pond sites as compared to creek sites. The porcine marker (Pig-1-Bac) was rarely detected in water samples suggesting that pig waste was not a relatively significant concern for the Edwards Aquifer. Human-associated Bacteroidales markers were detected mostly at surface water sites near densely populated urban areas and/or rural areas with high septic tank density, suggesting that their presence is the result of larger human population served by septic tanks or sewer infrastructure.

The highest fecal bacterial loadings were observed for site 5 and 18. Site 5 is a pond site in the contributing zone nearby a densely populated subdivision while site 18 is located on Leon Creek in the contributing zone located next to Interstate-10. Also, there is golf course nearby in the Dominion neighborhood. The runoff resulting from these activities may be a source of fecal contamination in the creek. Additionally, concentrations of fecal markers were higher for the stormwater samples, signifying that peak fecal concentration loadings occurred during storm-related events. The lowest fecal bacterial levels were observed for well water sites (Sites 1, 2 and 10) indicating that the natural biogeochemical processes are somewhat effective in decreasing the concentrations of surface-derived microbial contaminants in the groundwater.

This study is the first to examine and compare fecal contamination in the recharge and contributing zones of the Edwards Aquifer using a molecular MST approach targeting Bacteroidales 16S rRNA gene-based assays. The Bacteroidales marker assays, when combined with land use and weather information, can allow for a better understanding of the sources and fluxes of fecal contamination, which can help devise effective mitigation measures to protect water quality. Overall, these methods were successful in providing empirical scientific evidence of the predominant sources of fecal contamination in the Edwards Aquifer. While some of these contamination sources may be difficult to manage (i.e. dogs and other animals), the microbial source tracking study also provided evidence of sources in the watershed that can be controlled or mitigated for, such as human-derived sources that can be considered a public health risk. Results of the combined studies provide the basis for continued education and public outreach regarding sources of bacterial contamination in the Edwards Aquifer. For example, the scientific knowledge gained from this study provides the opportunity for resource managers to strengthen on-site septic system management programs and ensure they are aimed at reducing bacteria levels in the watershed. While these results should not be extrapolated to other geographic regions, information gained from the overall approach, the application of these tools, and lessons learned can be applied to other watersheds.

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ACRONYMS AND ABBREVIATIONS

µg	microgram
µl	microliter
BMP	best management practice
DNA	deoxyribonucleic acid
EPA	Environmental Protection Agency
FIB	fecal indicator bacteria
ml	milliliter
MST	microbial source tracking
ng	nanogram
OSSF	on-site sewage facility
PCR	polymerase chain reaction
qPCR	quantitative polymerase chain reaction
RNA	ribonucleic acid
SSO	Sanitary Sewer Overflow

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1.0 INTRODUCTION

The Edwards Aquifer is San Antonio's primary source of water and is important to the city's economic viability. Rainfall enters the aquifer through fractures, caves, sinkholes and other features replenishing the aquifer. However, development over the aquifer's recharge and contributing zones impacts the quality and quantity of water entering the aquifer and reduces the number of recharge features needed to maintain San Antonio's primary water resource. Sewage overflows and stormwater runoff introduce high levels of fecal bacteria into waterways and are considered the primary cause of water quality impairments in urbanized areas, particularly those affected by sanitary sewer overflows (SSOs) and on-site sewage treatment facilities (septic tanks). Identifying the primary sources of fecal pollution is critical for implementing adequate pollution control and prevention strategies.

The identification of the types of sources that contribute to bacteria in water systems is key when developing strategies to reduce bacteria and other pollution level in surface and groundwater as well as evaluating their potential impact on the environment. In a karst region where sources are not easily known or understood, microbial source tracking (MST) techniques can provide an opportunity to analyze water samples in a way that identifies the source of fecal bacteria in the sample, from simply identifying whether the source is human or animal to, at times, identifying the source down to the species (e.g., cow, dog, deer). The molecular methods used for MST most commonly include the analysis of genetic material (e.g., deoxyribonucleic acid [DNA] or ribonucleic acid [RNA]) to determine the source (human or animal) that contributed to the fecal bacteria observed in the water sample. The underlying assumption of these types of MST methods is that there are genetic sequences unique to bacteria from a particular host that can be used to identify where the bacteria originated.

Between 2017 and 2021, researchers at the University of Texas at San Antonio (UTSA), Civil and Environmental Engineering Department in San Antonio, Texas conducted microbial source tracking studies in order to determine what the predominant sources of fecal bacterial pollution are in the Edwards Aquifer recharge and contributing zones in Bexar County, Texas. They were implemented through a Proposition 1 Edwards Aquifer Protection Projects within Urbanized Areas of Bexar County's Recharge and Contributing Zones Funding Agreement between the City of San Antonio, the San Antonio River Authority, and UTSA. The results of the studies are reported here.

1.1 Background

High levels of fecal bacteria are the most commonly reported contaminants affecting the quality of recreational waters and drinking water sources in the United States (Santo Domingo et al. 2007, USEPA 2016). Pathogenic bacteria of various genera from human and animal fecal waste enter environmental waters via leaking on-site septic systems, wastewater treatment plant effluents, discharge from waste lagoons and pits at livestock facilities, from leaching of manure, and waste from wildlife and domestic animals (Dowd et al. 2008, Heaney et al. 2015, Cao et al. 2013). Fecal contamination of water poses a serious risk to human and environmental health via waterborne disease outbreaks, deterioration of recreational and drinking water quality, and degradation of aquatic ecology (Santo Domingo et al. 2007). Human illness and disease can result from drinking or swimming in water that contains pathogens or from eating shellfish harvested from such waters (EPA 2006). Globally, at least 2 billion people use a drinking water source that is contaminated with human and animal waste. Contaminated water can transmit diseases such as diarrhea, cholera, dysentery, typhoid, and polio and is estimated to cause 485,000 diarrheal deaths globally each year (WHO 2019). In addition, fecal contamination outbreaks into drinking water systems not only cause gastrointestinal (GI) illness, but skin irritations, respiratory diseases, and ear, nose and eye infections (Prüss et al. 1998, Soller et al. 2010, Tseng et al. 2012).

Sewage overflows and stormwater runoff introduce high levels of fecal bacteria into waterways and are considered the primary cause of water quality impairments in urbanized areas, while rural sources include livestock manure from barnyards, pastures, rangelands, feedlots, unfenced farm animals, improper manure or sewage land application, poorly maintained manure storage, and wildlife (Reed et al. 2011, USEPA 2009). In the state of Texas, approximately 42 % of assessed rivers and streams are identified as impaired, where the water body does not meet applicable water quality standards or is threatened for one or more designated uses by one or more pollutants (TCEQ 2019, USEPA 2014).

Presently, there is an incomplete understanding of the sources of fecal contamination found in the water entering the Edwards Aquifer, a source of drinking water to more than 2 million people in south-central Texas. Karst aquifers, such as the Edwards, are known to be vulnerable to human and animal waste contamination (Pronk et al. 2007) as disease outbreaks have been traced back to contaminated water originating from these systems (O'reilly et al. 2007, Borchardt et al. 2011, Wallender et al. 2014). A close link has been reported between impaired water quality and karst topography when evaluating karst water sources in the United States (Zhang et al. 2014, Reed et al. 2011). Rapid movement of water from the land surface to the subsurface through fractures, conduits and sinkholes lessens the amount of time available for physical processes and biogeochemical reactions that would otherwise decrease the concentrations of surface-derived contaminants in the subsurface. Furthermore, transport of microbial contaminants in karst systems is facilitated by high groundwater flow velocities, particularly following heavy rainfalls or other stormwater events (Kelly et al. 2009).

Identifying the primary sources of fecal pollution is critical for protecting human and environmental health and implementing adequate pollution control and prevention strategies. In this regard, risk management practices in water resources have become critical to the public health community not only due to their importance in preventing human diseases but also due to the economic repercussions associated with treatment and reuse (Soller et al. 2010). Poor microbial water quality increases the costs of services derived from water use and reuse. Hence, accurate and reliable fecal source identification is crucial to the implementation of best management practices (BMPs) that can accurately and cost effectively prevent, control, and remediate fecal pollution events and maintain water quality.

1.2 Microbial Source Tracking

Presently, there is no single biological, chemical or molecular fecal indicator used to monitor water quality. Nevertheless, microbial surrogates of fecal origin, such as members of the coliform and fecal streptococci bacterial groups, remain the most widely used indicators of fecal contamination because they exist in the intestinal tract of humans and other animals in large numbers. Commonly used fecal indicator bacteria (FIB) include *E. coli*, a species within the fecal coliform subset, and enterococci, a subgroup within the fecal streptococcus group. Although not generally harmful themselves, FIBs are used to indicate the potential presence of pathogenic microorganisms such as bacteria, viruses, and protozoans that also live in human and animal digestive systems, as it is difficult, time-consuming, and expensive to test directly for the presence of a large variety of pathogens (EPA 2012). As a water quality monitoring tool, they provide a baseline for establishing public health risks (Hooper et al. 2001, Anderson et al. 2005), however, these conventional FIBs are unreliable for fecal source tracking (FST) applications due to widely varying survival rates in environment, failure to discriminate between human and animal sources, and their inability to distinguish between fecal bacteria associated with recent contamination events and those adapted to secondary habitats (Anderson et al. 2005, Whitman et al. 2003). FST techniques can provide an opportunity to analyze water samples in a way that identifies the source, human or animal, and species, cow, dog, pig, etc., of fecal bacteria. Identifying the sources of fecal pollution that contribute to bacteria in environmental waters is paramount when developing strategies to reduce bacteria and other pollution levels in surface and groundwater as well as evaluating their potential impact on the environment.

Fecal source tracking can be categorized into culture-based library-dependent, culture-based library-independent, culture-independent library-dependent and culture-independent library-independent methods. For library-dependent methods, comparisons between test patterns or fingerprints from a library database and environmental samples are required for fecal source identification. Library-independent, culture-independent methods include molecular methods of microbial source tracking (MST), which are based on the use of genetic markers assayed directly from DNA extracted from a water sample (Field et al. 2007). MST includes three methodologies used to determine sources of fecal bacteria: biochemical, chemical, and molecular (Meays et al. 2004). Early microbial source tracking methods relied on fecal coliform/fecal streptococci (FC/FS) ratios to assess general sources of nonpoint fecal pollution. In this method, FC/FS ratios less than 0.1 indicate a wild animal source, between 0.1 and 0.6 domestic animals, and greater than 4.0 indicate humans as the sources of fecal bacteria (Geldreich et al. 1976). More recent studies have found difficulties using FC/FS ratio for agricultural settings (Howell et al. 1996) and the use of FC/FS ratios for fecal source tracking is no longer recommended by the American Public Health Association (APHA 2005).

Recent advances in molecular biology have led to the development and advancement of molecular methods of MST using anaerobic bacterial genes to identify and quantify microorganisms in many environments. In comparison to culture-based and other MST methods, molecular MST methods have the advantage of sampling an entire population present in a water sample, are not solely limited to culturable microbes, require less time to perform, and do not require a library database (Field et al. 2007). Molecular MST most commonly includes the analysis of genetic material (e.g., DNA or RNA) to determine bacterial sources in an environmental water sample. The underlying assumption of this MST method is that there are genetic sequences unique to bacteria from a particular host that can be used to identify bacterial origin. The majority of bacterial cells in feces include fecal anaerobes, such as *Bacteroides*, which are present at much higher densities than coliforms and enterococci (Savage 2001). Molecular MST relies on polymerase chain reaction (PCR), a method of amplifying DNA by making billions of copies of an original template strand by cycling through different temperature gradients. Because a large number of copies can be made from a single target DNA segment, and bacterial cells need not be alive to have intact nucleic acids, the method is extremely sensitive for detecting the presence of microorganisms. Within a few hours, this method can provide detection results for a target gene through agarose gel electrophoresis (Toze et al. 1999). The ability of PCR to produce a large number of copies of a specific nucleic acid target makes it well suited for a rapid, sensitive, and specific detection of emerging pathogens or their more representative surrogate in water samples. However, this method can only identify the occurrence of the targeted gene and cannot calculate its concentration; therefore, it is used solely for qualitative analysis (Schuelke et al. 2000). Emerging real-time quantitative PCR (qPCR) can measure the fluorescence of a reporting dye and monitor the amplification through each temperature cycle, which is then compared with a known standard to determine the concentration of the target gene. Moreover, qPCR can provide greater sensitivity and can enable discrimination of gene numbers across a wider dynamic range than otherwise found with endpoint PCR, because the reaction is monitored throughout the amplification process as opposed to a single analysis after completion of the final PCR cycle (Smith et al. 2009).

Of the fecal bacteria, the *Bacteroidales* group is an advantageous qPCR target because they are obligate anaerobes and survive for only short periods of time after release from their hosts into oxygenated surface water. Walters et al. (2009) found that *Bacteroidales* DNA markers survive and persist for approximately 4 to 10 days in freshwater. As a result, these bacteria do not have the ambiguous survival and sourcing issues that can be present with *E. coli* and *Enterococcus* spp., and as such, can be linked with good spatio-temporal resolution to a sampling site (Lu et al. 2012, Kapoor et al. 2013, Kapoor et al. 2015). The most common qPCR markers target the *Bacteroidales* 16S rRNA genes. These genetic markers may also detect viable but nonculturable (VBNC) cells that are not detected by conventional cultivation approaches but may still pose a public health risk (Kapoor et al. 2018). Kildare et al. (2007) used *Bacteroidales* 16S rRNA gene sequences for quantitative detection of universal, human, bovine, and canine-associated fecal contamination. Recent studies have developed additional qPCR-based assays for human-associated (Shanks et al.

2010, Seurinck et al. 2005), chicken and duck (Kobayashi et al. 2012), and swine (Heaney et al. 2015, Lamendella et al. 2009) *Bacteroidales* markers. In a karst region where sources of fecal contamination are not easily known or understood, molecular methods used for MST provide a way to determine the sources of fecal contamination. This study applied qPCR-based molecular MST methods to identify human and animal sources of fecal *Bacteroidales* in the Edwards Aquifer and to gain a better understanding of the spatial and temporal patterns of these fecal indicator organisms.

The EPA has validated qPCR-based methods, such as Method 1609.1 and Method 1611.1, to identify fecal indicator bacteria; however, until very recently there was no validated method for microbial source tracking to characterize fecal pollution in recreational waters originating from a specific host. In March 2019, the EPA validated Method 1696 to characterize human sources of fecal pollution using the HF183 qPCR assay (EPA 2019). While this method has been approved, no regulatory water quality standards have been created for the HF183 assay or any of the *Bacteroidales* qPCR-based assays. As such, the qPCR markers used in this study identify trends in fecal contamination and provide a relative comparison of fecal contamination levels between pollution sources but do not elucidate a risk to human or environmental health. Concentrations of FIBs in recreational waters are linked to adverse health outcomes in humans through epidemiology studies, however, few recreational water epidemiology studies have measured qPCR markers in conjunction with conventional FIB and attempted to correlate them with GI illness in humans (Boehm et al. 2015). Studies using quantitative microbial risk assessment (QMRA) have shown that health risk increases as the concentration of human qPCR markers increases (Boehm et al. 2015, Sinigalliano et al. 2010, Staley et al. 2012). The need for additional human health risk assessment modeling and the adoption of regulations for water quality standards related to qPCR-based assays presents a research gap in the use of molecular microbial source tracking for the protection of human and environmental health.

1.3 The Edwards Aquifer

The Edwards Aquifer is located on the eastern edge of the Edwards Plateau in south-central Texas and extends through parts of Kinney, Uvalde, Zavala, Medina, Frio, Atascosa, Comal, Guadalupe, Hays, and Bexar counties, covering an area of approximately 7,200 square miles. It is comprised of a group of porous limestones between 300-700 feet thick, of which the main geologic unit is the

Edwards Limestone, and is characterized as a karst aquifer due to its high permeability. The Edwards can be divided into three main zones based on the ability of water to infiltrate into the Aquifer from the surface: the recharge zone, contributing zone and artesian zone. The contributing zone, also known as the drainage or catchment area, is a relatively impermeable area where streams and runoff from rainfall flow to the recharge zone. It covers approximately 5,400 square miles. Surface water from streams originating in the contributing zone or from direct rainfall infiltrates the Aquifer through the highly faulted and fractured limestone outcrop in the recharge zone. Approximately 1,250 square miles of Edwards Limestone is exposed at the ground surface and composes the recharge zone. In the artesian zone, water is confined underground by pressure from layers of impermeable rock which overlie the Edwards limestone (Eckhardt 2019, EAA 2019).

The Edwards Aquifer serves as the principal source of water for the region’s agricultural and industrial needs and is characterized by many springs which provide flow for endangered species habitat and for recreational purposes (EAA 2019). The economic benefit of the study lies in the supply and demand of the drinking water industry, which is adversely impacted by the presence of fecal pollution in drinking water resources. This is especially important for karst aquifers, such as the Edwards, which also serves as the primary source of drinking water to more than 2 million people, including the City of San Antonio and its surrounding communities, as disease outbreaks have been traced back to contaminated drinking water originating from karst systems (O'reilly et al. 2007, Borchardt et al. 2011, Wallender et al. 2014). In the largest documented case of waterborne disease outbreak in Bexar County, TX, approximately 2,000 people became ill after a well in Braun Station, a suburb of San Antonio, was contaminated with sewage (Solo-Gabriele et al. 1996). Presently, there is an incomplete understanding of the sources of potentially pathogenic fecal contamination found in Edwards Aquifer water.

1.4 Project Objectives and Approach

The primary goal of this project was to design and implement an efficient fecal source tracking and evaluation program for the Recharge and Contributing Zones of Edwards Aquifer in Bexar County, TX. We seek to identify potential sources of fecal bacteria such as (1) municipal waste/runoff including on-site sewage facilities and sanitary sewer overflows, and (2) animal waste (livestock and domesticated animals) as well as other contributing factors (water temperature, nutrients, and available organic material). The spatiotemporal fecal input, including source identification, potential load allocations and factors that contribute to seasonal variability of microbial concentrations have been evaluated in this project. The pilot project has been designed to support the San Antonio River Authority (SARA) in evaluating fecal microbial input into Aquifer water from the contributing and recharge zones and assessing its potential impact on water quality as well as the health of humans and the environment. Specific objectives for the study included the following:

- Determine the presence or absence of human and animal sources of fecal contamination in the Edwards Aquifer.
- Determine the presence or absence of pathogenic organisms (*E. coli* O157:H7) and their locations.
- Determine the predominant sources of fecal contamination, if present, at specific key sites and examine the differences between sites.
- Determine if and/or what temporal trends may exist based on the time period sampled.
- Determine what sources may be controllable.
- Use results from this study for public outreach, mitigation where practical, and improvement of the design of future water-quality monitoring projects.

The potential impact of microbial influx from the contributing and recharge zones into groundwater was assessed on the presence of both fecal bacteria and human pathogens like *E. coli* O157:H7. qPCR of host-associated Bacteroidales 16S rRNA genes (human, bovine, canine, swine, avian etc.) and tests for human pathogens were used to identify the origin of fecal contamination in the region. In addition to these molecular tools, other tests including water temperature, pH, dissolved oxygen (DO) and nitrate/nitrite/ammonia have also been used to document water quality. Altogether, the molecular tools applied in this study along with spatial and temporal sampling will provide information on several fecal contaminants, their locations and relative abundance. The information from this study will assist the City and SARA with pollution source location and the development of BMPs for mitigating bacterial contributions to the water entering the aquifer.

A prospective 1-year study was also undertaken to examine differences in fecal pollution sources and levels between urban and rural areas based on proximal land use features of specific key sites. The purpose of this sub-study was to apply MST techniques in conjunction with land-use information to evaluate fecal contamination along developed and undeveloped lands within the recharge and contributing zones of the Edwards Aquifer in Bexar County, Texas. To examine fecal pollution sources, Cibolo Creek Watershed and Leon Creek Watershed were chosen as the study areas as both are located within the contributing and recharge zones of the Edwards Aquifer and streams in both watersheds have been identified as impaired due to pathogenic bacteria (EPA 2014). Additionally, it was found that the sources of fecal pollution differ between areas of rural and urban land use (Kelsey et al. 2004, Lee et al. 2014). As such, determination of site locations within the watersheds was also based on an analysis and characterization of land use. Sample sites were selected along two creeks: Balcones and Leon. Balcones Creek is located within the Cibolo Creek Watershed and forms the border between Bexar and Kendall counties. Furthermore, it runs outside of any major city, while Leon Creek is located within the northwestern city limits of San Antonio, the second most populous city in the state of Texas. This study will be the first to examine and compare fecal contamination at different land use areas in the portion of the Edwards Aquifer recharge and contributing zones within Bexar County using a molecular MST approach for source identification and quantification.

2.0 STUDY DESIGN AND METHODOLOGY

2.1 Sampling Sites

The present study was conducted at twenty sites located within the Edwards Aquifer recharge and contributing zones in Bexar County, Texas, USA (Figure 1). The sites consisted a combination of wells, creek sites and ponds. Sanitary sewer overflows (SSOs), on-site sewage treatment facilities (septic tanks) and livestock/animals are assumed to be the primary sources of fecal pollution, and twenty sampling sites (Site 1 to 20) were chosen based on the presence and proximity to pollution sources within the recharge and contributing zones of the Edwards Aquifer within Bexar County (Table 1). Site 20 was later excluded from the study due to access issues and a new site 21 was added instead. Thus, in this report, we used twenty sites (site 1-19, and site 21) for the analysis purposes. The pictures of the sampling sites are shown in Appendix A.

2.2 Sample collection and DNA extraction

Water samples were collected by grab sampling in sterile 1-liter Nalgene (Rochester, NY) bottles from each site bi-monthly over a two-year period from January 2018 to March 2020, for a total of 56 sample collection events. All water samples were transported on ice to the laboratory at UTSA (San Antonio, Texas), where the samples were filtered within 6 hours of collection. Water samples (300mL) were filtered in duplicate on a vacuum manifold through 0.45-μm-pore-size, 47 mm diameter polycarbonate membranes (Pall Corporation, Ann Arbor, Michigan) and immediately stored at -80 °C until DNA extraction. Sterile de-ionized water controls were filtered with each sample event to check for cross contamination during sample processing. After filter membranes were allowed to thaw to room temperature, DNA was extracted using the DNeasy PowerLyzer PowerSoil Kit (Qiagen, Hilden, Germany) according to the manufacture's protocol. A Nanodrop One^{C} Spectrophotometer (Thermo Scientific, Wilmington, DE) was used to determine DNA purity and concentration (ng/μL) and DNA extracts were stored at -20 °C until used in qPCR assays.

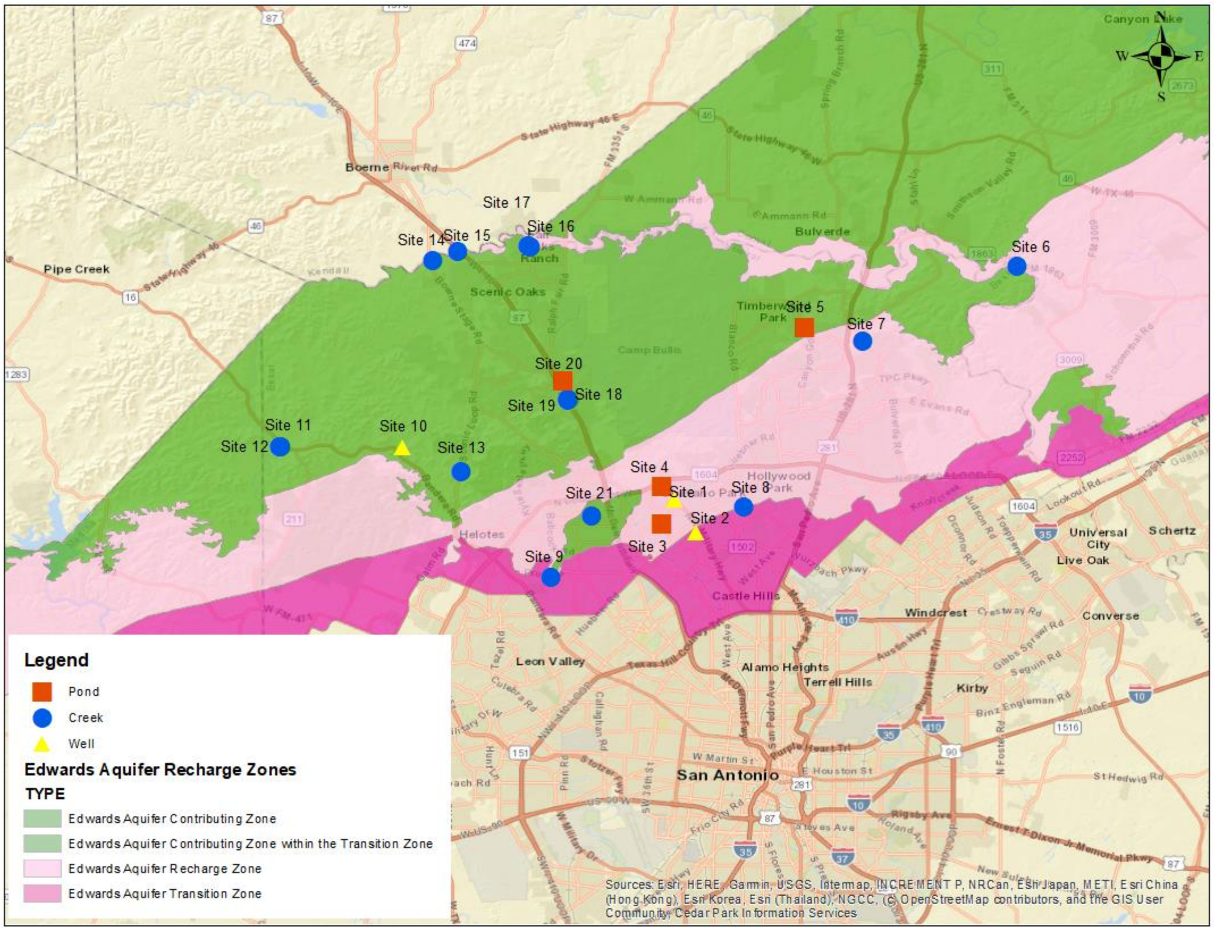

Figure. 1. Map of sampling sites located on the Edwards Aquifer recharge and contributing zones in Bexar County, Texas, USA.

Table 1. Description of sampling sites in this study.

Site No.	Type	Zone	Latitude (DD)	Longitude (DD)	Site description
1	Groundwater public service well	Recharge	29.5884	-98.5578	Located in Shavano Park; about 435 feet depth
2	Groundwater public service well	Recharge	29.5685	-98.5447	Located in Shavano Park; about 510 feet depth
3	Pond	Recharge	29.5740	-98.5649	Three outfalls located to the west and NW, approximately 100 feet.
4	Pond	Recharge	29.5971	-98.5650	One outfall located approximately 550 feet to the NW
5	Pond	Contributing	29.6926	-98.4787	Bordered on East side by densely populated subdivision
6	Creek	Recharge	29.7296	-98.3509	Located in Cibolo Creek; Farthest location in the NE part of Bexar County in recharge zone.
7	Creek	Recharge	29.6847	-98.4439	Located in Elm Waterhole Creek; Densely populated subdivision on West side; about 600 feet west of an outfall.
8	Creek	Transition	29.5845	-98.5154	Located in Panther Springs Creek; Surrounded by neighborhood on all sides; about 225 feet South and 300 feet NW of two outfalls
9	Creek	Recharge	29.5422	-98.6320	Located in Leon Creek; About 375 feet NW of an outfall.
10	Private groundwater well	Contributing	29.6200	-98.7220	Located in Helotes, about 60 feet depth

11	Creek	Contributing	29.6207	-98.7946	Located in San Geronimo Creek; east of Bandera Road
12	Creek	Contributing	29.6208	-98.7956	Located in San Geronimo Creek; west of Bandera Road
13	Creek	Contributing	29.6061	-98.6859	Located in Helotes Creek in rural area
14	Creek	Contributing	29.7331	-98.7035	Located in Balcones Creek in rural area in the drainage zone on the northern boundary of Bexar County; About 5,500 feet west of site 15.
15	Creek	Contributing	29.7385	-98.6880	Located in Balcones Creek in rural area in the drainage zone on the northern boundary of Bexar County; About 5,500 feet east of site 14.
16	Creek	Recharge	29.7411	-98.6446	Located in Balcones Creek in rural area in the recharge zone on the northern boundary of Bexar County; About 2,250 feet SE of site 17.
17	Creek	Recharge	29.7420	-98.6456	Located in Balcones Creek in rural area in the recharge zone on the northern boundary of Bexar County; About 2,250 feet NW of site 16.
18	Creek	Contributing	29.6489	-98.6216	Located in Leon Creek; One outfall in about 20 feet to the North; about 400 feet downstream of site 19.
19	Creek	Contributing	29.6494	-98.6219	Located in Leon Creek; One outfall in about 20 feet to the South; about 400 feet upstream of site 18.
20	Pond	Contributing	29.6605	-98.6247	About 550 feet NE of outfall
21	Creek	Contributing Zone within Transition Zone	29.5793	-98.6074	Located in Leon Creek in the contributing zone within the transition zone; about 650 feet SW of apartment complex and 200 feet NE of an outfall

Concentrations of fecal bacteria can change dramatically over short periods of time particularly during precipitation events. During wet weather events, more frequent sampling is required, since fecal bacterial concentrations can vary more than 30-fold over a 24-hour period (Meays et al., 2004). There can also be considerable variation between wet and dry weather bacterial concentrations, which is most likely due to storm runoff and high SSO flows causing a dramatic input of fecal matter and resuspension of bacteria. High fecal concentrations after storms may also be due to inputs of waste material from overland flows or newly inundated areas. In addition to routine bi-monthly sampling, a total of 13 storm event-related samples were collected at sites 3, 4, 5, 7, 9, 13, 16, 19, and 21 using Nalgene Storm Water Sampler and Mounting Kit (Thermo Fisher Scientific) (Table 2). Storm event-related sampling included multi-day samples for before, during, and after storm events to determine peak concentration loadings. Routine and storm event-related sampling should permit identification of possible source areas, transport processes, and source/host organisms for the fecal waste inputs into the Edwards Aquifer.

Table 2. Storm-event related sample collection events.

Collection Event No.	Collection Dates	Site Number	Rainfall in previous 7 days (inches)
1	8/31/18 - 9/5/18	19	9.58
2	10/12/18 - 10/16/18	19	2.14
3	10/12/18 - 10/16/18	21	2.14
4	4/5/19 - 4/8/19	4	1.04
5	4/22/19 - 4/26/19	13	0.23
6	5/31/19 - 6/7/19	5	2.1
7	10/24/19 - 10/25/19	3	0.91
8	10/24/19 - 10/25/19	7	3.26
9	1/10/2020 - 1/11/2020	16	0.57
10	5/23/2020 - 5/25/2020	9	1.03
11	5/23/2020 - 5/25/2020	21	2.14
12	6/23/2020 - 6/27/2020	19	0.31
13	6/23/2020 - 6/27/2020	21	0.31

2.3 qPCR analyses

The occurrence and relative abundance of eleven different MST markers in surface water samples was measured using qPCR assays with DNA extracts as the templates (Table 3). The fecal bacterial markers included six primer sets targeting Bacteroidales 16S rRNA genes: Universal Bacteriodales (BacUni assay), human-associated Bacteroidales (HF183 and BacHum assays), Chicken/Duck Bacteriodales (Chicken/Duck-Bac assay), Dog Bacteriodales (BacCan assay), Pig Bacteroidales (Pig-1-Bac) and Cow Bacteriodales (BacCow assay) (Kildare et al. 2007, Haugland et al. 2010, Kobayashi et al. 2013). In addition, two conventional fecal bacterial groups were also tested using qPCR based assays, E. coli (EC23S857 assay) (Chern et al. 2011) and Enterococcus spp. (Entero1 assay) (Ludwig & Schleifer, 2000). In the second year of the sampling, we discontinued two markers (Pig Bacteroidales and *E. coli* O157:H7) and added two new markers targeting ruminant Bacteroidales (Rum2Bac) and avian associated marker (GFD). For the purposes of this study, the qPCR targets are grouped into three classifications: general indicators, which include the BacUni, EC23S857, and Entero1 assays; animal-associated markers including the BacCan, BacCow, Chicken/Duck-Bac, Pig-1-Bac, Rum2Bac and GFD assays; and the human-associated markers including HF183 and BacHum.

All qPCR assays were performed on the CFX96 Touch Real-Time PCR Detection System (Bio-Rad, Hercules, CA) using either iTaq™ Universal Probes Supermix (Bio-Rad) or SsoAdvanced Universal SYBR Green Supermix (Bio-Rad). The reaction mixtures (25 μL) contained 12.5μL of supermix, 300 nM of each primer (forward and reverse), 100 nM of probe, and 2 μL of DNA template. The qPCR data were analyzed using Bio-Rad's CFX Manager Software (version 3.1). Standard curves were run in duplicate for each qPCR plate by using serially diluted plasmid standards purchased from Integrated DNA Technologies (IDT, Skokie, IL) containing the sequences for each of the targeted genes. Each standard curve was generated from at least six 10-fold plasmid dilutions in duplicate. The percent amplification efficiencies were calculated by the instrument manufacturer's instructions (Bio-Rad). Extraction controls and no-template controls (three per plate) were used to check for cross contamination, and 10-fold dilutions of selected DNA extracts were used to test for PCR inhibition as described in previous study (Pitkänen et al. 2013).

Table 3. Primers/probes for the PCR assays used in the study.

Assay	Primer/probe Sequence (5'-3')	Reference
Universal Bacteroidales (BacUni)	BacUni-520f: CGTTATCCGGATTTATTGGGTTTA BacUni-690r: CAATCGGAGTTCTTCGTGATATCTA BacUni-656p: 6-FAM-TGGTGTAGCGGTGAAA-TAMRA-MGB	Kildare et al., 2007
Human Bacteroidales (BacHum)	BacHum-160f : TGAGTTCACATGTCCGCATGA BacHum-241r: CGTTACCCCGCCTACTATCTAATG BacHum-193p: 6-FAM-TCCGGTAGACGATGGGGATGCGTT-TAMRA	Kildare et al., 2007
Human-specific Bacteroidales (HF183)	HF183-1: ATCATGAGTTCACATGTCCG BthetR1: CGTAGGAGTTTGGACCGTGT BthetP1: 6FAM-CTGAGAGGAAGGTCCCCCACATTGGA-TAMRA	Haugland et al., 2010
Cow Bacteroidales (BacCow)	BacCow-CF128f: CCAACCTTCCCGATACTC BacCow-305r: GGACCGTGTCTCAGTTCCAGTG BacCow-257p: 6-FAM-TAGGGGTTCTGAGAGGAAGGTCCCCC-TAMRA	Kildare et al., 2007
Dog Bacteroidales (BacCan)	BacCan- 545f: GGAGCGCAGACGGGTTTT BacUni-690r: CAATCGGAGTTCTTCGTGATATCTA BacUni-656p: 6-FAM-TGGTGTAGCGGTGAAA-TAMRA-MGB	Kildare et al., 2007
Pig Bacteroidales (Pig-1-Bac)	Pig-1-Bac32Fm: AACGCTAGCTACAGGCTTAAC Pig-1-Bac108R: CGGGCTATTCCTGACTATGGG Pig-1-Bac44P: FAM-ATCGAAGCTTGCTTTGATAGATGGCG-BHQ-1	Mieszkin et al., 2009
Chicken-Duck Bac (Bird-specific)	qCD362F-HU: AATATTGGTCAATGGGCGAGAG qcD464R-HU: CACGTAGTGTCCGTTATTCCCTTA qBac394 MGB-HU: FAM-TCCTTCACGCTACTTGG-MGB	Kobayashi et al., 2013
General *Enterococcus* (Entero1)	ECST748F: AGAAATTCCAAACGAACTTG ENC854R: CAGTGCTCTACCTCCATCATT GPL813TQ: 6FAM-TGGTTCTCTCCGAAATAGCTTTAGGGCTA-TAMRA	Ludwig & Schleifer, 2000
***E. coli* (EC23S857)**	F: GGTAGAGCACTGTTTTGGCA R: TGTCTCCCGTGATAACTTTCTC P: 6FAM-TCATCCCGACTTACCAACCCG-TAMRA	Chern et al., 2011
Ruminant Bacteroidales (Rum2Bac)	BacB2-590F: ACAGCCCGCGATTGATACTGGTAA Bac708Rm: CAATCGGAGTTCTTCGTGAT BacB2-626P: FAM-ATGAGGTGGATGGAATTCGTGGTGT TAMRA	Mauffret et al., 2012
Avian associated marker (GFD)	F: TCGGCTGAGCACTCTAGGG R: GCGTCTCTTTGTACATCCCA	Green et al., 2012

2.4 PCR for *E. coli* O157:H7

We used a multiplex PCR protocol by using primer sets that directly detect genes that are involved in biosynthesis of O157 and H7 antigens and primer pairs that identify intimin, the major known virulence trait of *E. coli* O157:H7, which mediates the intimate adherence of the organism to host cells (Hu et al., 1999). The DNA extracted for each water sample was used as a template in the multiplex PCR reaction. The multiplex PCR product was analyzed using gel electrophoresis. The simultaneous amplification of the 3 DNA products corresponding to genes for O157, H7 and intimin is used as a standard for positive identification of a pathogenic sample. The pathogenic *E. coli* was not detected in any of the samples in the first year of sampling, and was later discontinued in consultation with the City of San Antonio.

2.5 Water quality parameters and rainfall data

Water quality parameters were measured for all sites and sample collection events throughout the course of the study to monitor water quality and examine spatial and temporal variability. Measured water quality parameters included surface water temperature, dissolved oxygen (DO), pH and dissolved nitrogen species (nitrate/nitrite/ammonia). DO and temperature were measured on-site using an Intellical™ LDO101 Field Luminescent/Optical probe and HQ40d portable multi-meter (HACH, Loveland, CO). pH was measured on-site using an Intellical™ PHC101 probe and HQ40d portable multi-meter (HACH, Loveland, CO). Ammonia (NH_3-N), nitrite (NO_2^--N) and nitrate (NO_3^--N) concentrations were measured in the UTSA laboratory using Salicylate Method 10205 (HACH TNTplus 830 ultra-low range kit), USEPA Diazotization Method 10207 (HACH TNTplus 839 low range kit) and Dimethylphenol Method 10206 (HACH TNTplus 835 low range kit), respectively, and a HACH DR 2800 spectrophotometer. Values were reported as below detection limit (BDL) if measured below the TNTplus kit limit of detection. For statistical analysis, BDL and non-detect (ND) data points were given a value of zero. Rainfall data within 24 hrs and seven days prior to sample collection was obtained from the USGS National Water Information System (https://waterdata.usgs.gov/tx/nwis/). USGS stations, 08178593, 08183978, 08180941, 08180586, 08183900 and 08178585 were chosen based on their proximity to sampling sites.

2.6 Land-use characterization study

The present study was conducted along the reaches of Balcones Creek and Leon Creek located within the Edwards Aquifer recharge and contributing zones in Bexar County, Texas, USA (Figure 2). Water samples collected over a one-year period from January 2018 to February 2019 were used for this study, for a total of 26 sample collection events. Balcones and Leon creeks are located within the Cibolo and Leon watersheds, respectively and flows across limestone bedrock. The entire length of the Balcones Creek is in the Edwards Aquifer contributing zone while approximately one-third of the Leon Creek is in the contributing zone. The source of Balcones Creek is located in Bandera County, approximately one mile southwest of the Bexar, Kendall and Bandera County line junction. The creek transects a rural area, flowing east for approximately 15 km and forming the boundary for Bexar and Kendall counties until convergence with the Upper Cibolo Creek at the junction of Bexar, Kendall and Comal counties. According to the 2014 Texas Water Quality Assessment Report, the Cibolo Creek has been identified as impaired both upstream and downstream of convergence with Balcones (USEPA 2014, Tanvir Pasha et al. 2020). The Leon Creek source is located in northwestern Bexar County and flows in a generally southern direction approximately 65 km through the western portion of the city of San Antonio, TX, until it's convergence with the Medina River south of San Antonio. Approximately 42 km (65%) of the Leon Creek have been identified as impaired (USEPA 2014).

Sanitary sewer overflows (SSOs), on-site sewage treatment facilities (septic tanks) and livestock/animals are assumed to be the primary sources of fecal pollution, and eight sampling sites were chosen based on the presence and proximity to pollution sources within the recharge and contributing zones of the Edwards Aquifer within Bexar County. Sites 14, 15, 16 and 17 are located along Balcones Creek (Figure 2), and were re-named as B1, B2, B3 and B4, respectively for this sub-study. Sites B-1 and B-2 are located furthest upstream in the Edwards Aquifer contributing zone. Sites B-3 and B-4 are located near the City of Fair Oaks Ranch in the Edwards Aquifer recharge zone, with site B-4 as the most downstream site. Approximately 45% of the residential properties in Fair Oaks Ranch use septic systems and are not part of the city's wastewater collection (City of Fair Oaks Ranch, 2019). Sites 9, 18, 19, and 21 are located along Leon Creek within the City of San Antonio and are predominantly urban land-use sites, and were re-named as L1, L2, L3 and L4, respectively for this sub-study. Sites L-1 and L-2 are located upstream near the Dominion neighborhood and the Dominion Country Club and golf course in the Edwards Aquifer contributing zone. Site L-3 is located east of the University of Texas at San Antonio (UTSA) and next to a student housing apartment complex in the Edwards Aquifer contributing zone within the transition zone. Site 4 is located furthest downstream in a residential area in the Edward's Aquifer recharge zone.

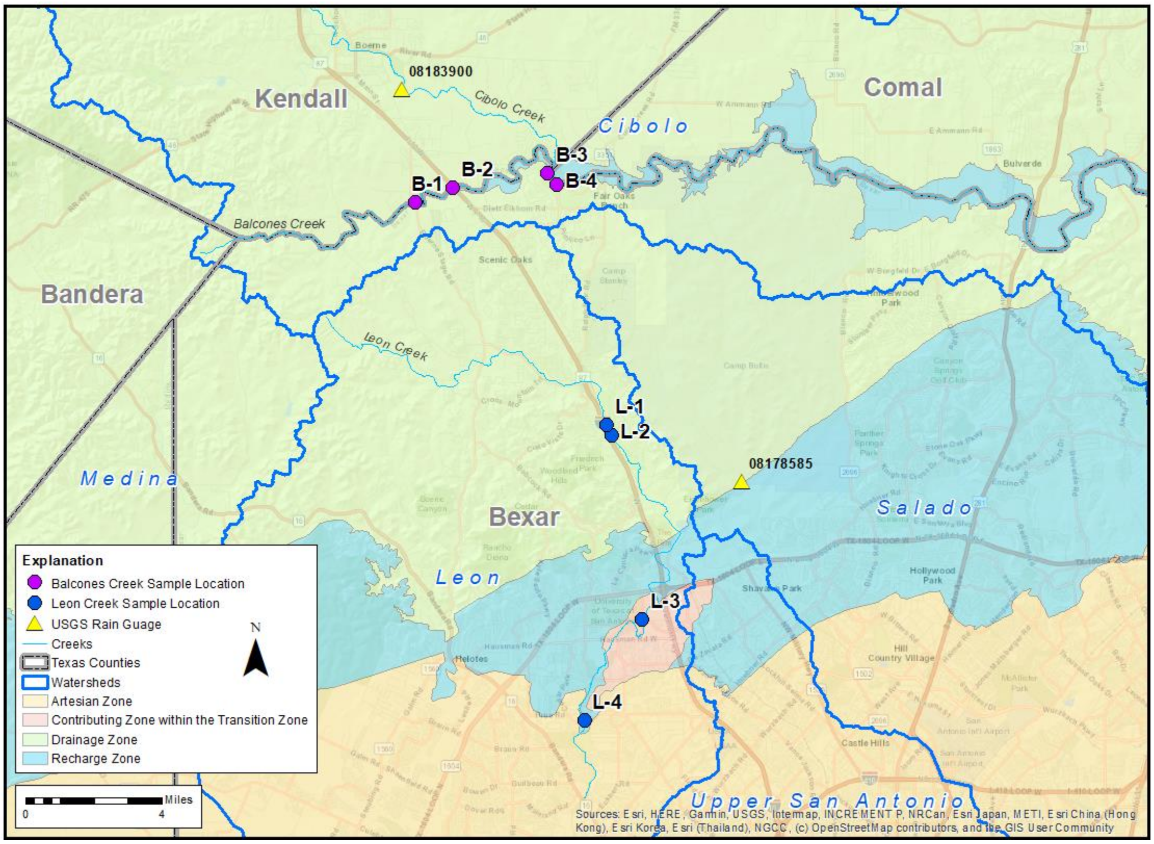

Figure 2. The land-use study area with sampling site locations. Sites B-1 through B-4 are located on Balcones Creek and sites L-1 through L-4 are located on Leon Creek.

An a priori assessment of study sites was conducted based on proximal land-use information obtained from Bexar County (Figure 3) and a detailed analysis for each creek was performed as factors influencing fecal contamination sources and levels differ between urban and rural areas (Ridley et al. 2014). Land use for each creek was characterized based on four features of interest: on-site sewage facilities (OSSFs), wastewater collection infrastructure, average human population, and percentage of developed vs. undeveloped land. Land use features of interest were investigated at a 1-km spatial scale to support detection of recent contamination from nearby inputs. ArcMap 10.5.1 (Environmental Systems Research Institute, Redlands, CA) was used to create 1-km buffer zones encompassing sampling sites along the length of Leon and Balcones creek and used to perform all calculations for features of interest. The total area analyzed for Leon and Balcones creek buffer zones was approximately 30.1 and 15.5 km^2, respectively. The average human population for each zone was calculated based on census tract data obtained from Bexar County and the Texas Demographic Center. The average number of septic systems per person and wastewater collection infrastructure density were calculated for each zone using OSSF data obtained from Bexar County Public Works, residential address data from Kendall County and the City of Fair Oaks Ranch, and underground wastewater collection infrastructure data from San Antonio Water Systems (SAWS). The percentage of developed vs. undeveloped land for Leon Creek was obtained from the City of San Antonio and approximated based visualizations of impervious cover within the Balcones Creek buffer zone from the World Imagery Basemap in ArcMap. Additionally, land use parameters were also estimated for each individual site using the above approach and correlation coefficients were calculated between the marker occurrence and land use variables.

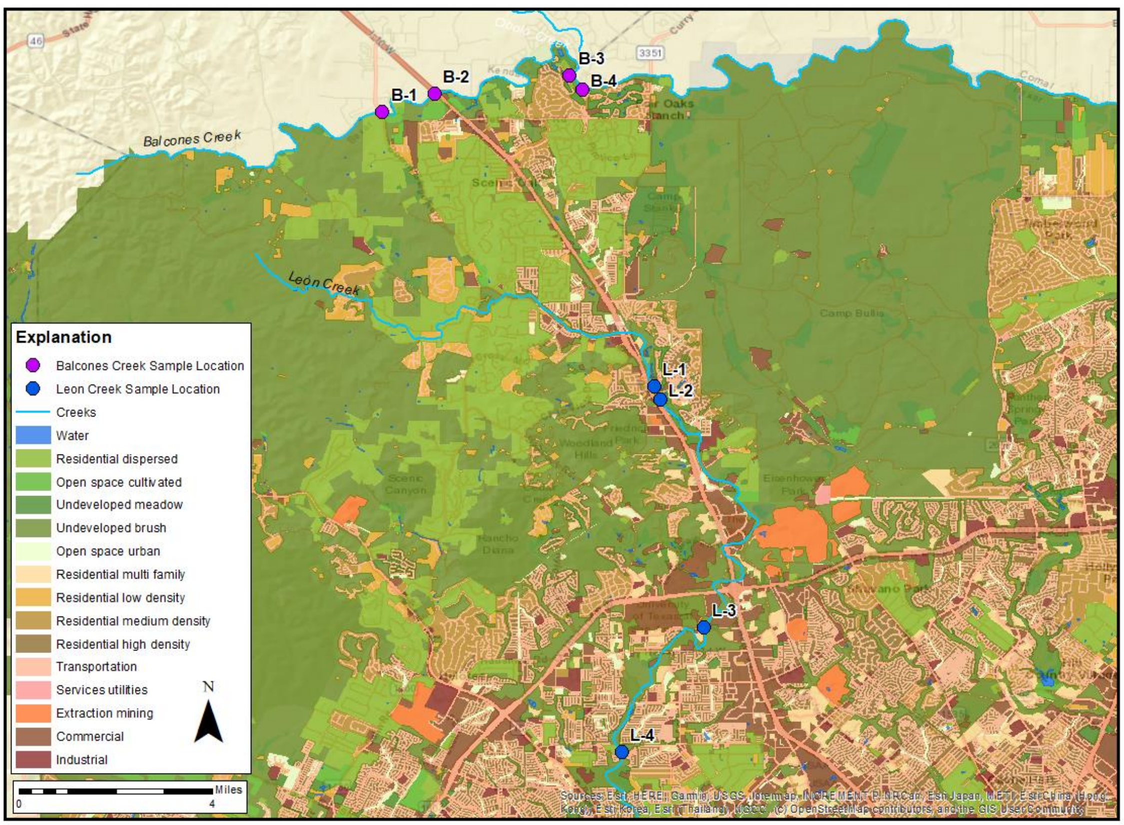

Figure 3. Map showing land use and impervious cover in 2017 for Bexar County, Texas.

2.7 Data analyses

Based on qPCR standard curves, the targeted marker copy number per 100 mL of water was calculated for all samples with values above the limit of quantification for each assay, and all raw data were $\log_{10}$ transformed before statistical analysis. Results below detection limit were assigned a value of one before log transformation. Statistical tests showed that microbiological and environmental data were not distributed normally. Nonparametric Wilcoxon signed rank test was used to determine if differences between marker concentrations among sampling sites were statistically significant. Using Microsoft Excel, box plots were generated for each qPCR marker across all the sites. Horizontal lines within the boxes represent the median (2nd quartile) and the lower and upper ends of the boxes represent the 1st and 3rd quartiles, respectively. The mean is represented by an "x". Whisker caps and open circles represent the maximum/minimum marker concentrations and outliers, respectively. Spearman's rank correlation coefficients (r) between marker concentrations and water quality parameters were also calculated. For comparison purposes, coefficients are characterized by a previously published scale (Stachler et al. 2018): 0.2–0.39 (weak correlation), 0.4–0.59 (moderate correlation), 0.6–0.79 (strong correlation), and 0.8–1 (very strong correlation). Differences and correlations were considered statistically significant when $p < 0.05$.

2.8 Quality Assurance/Quality Control

All environmental samples were collected and handled according to procedures outlined in the EPA field sample collection protocol - EPA Method 1669 (USEPA, 1996) including labeling of containers and logging of sample information on field logs. All samples were transported on ice and were accepted in good condition, according to protocol. Samples were logged in upon arrival at the laboratory and given a unique sample number for identification purposes. The following quality control activities were conducted during the PCR laboratory analysis: filtration controls, positive controls, no template controls, method accuracy, and specificity.

3.0 RESULTS AND DISCUSSION

3.1 Performance of qPCR assays

Each qPCR well plate included duplicate standard curves generated from serial dilution of known target copies per reaction which were used to determine the amplification efficiencies and linear ranges of the qPCR assays. The linear range of quantification for all qPCR assays was between 10^1 and 10^6 copies per reaction. The qPCR amplification efficiencies for all the assays ranged from 95 to 120 % and R^2 values were all greater than 0.948 (Table 3). PCR inhibition tests were done on the first three set of samples for each site (12% of total samples). PCR inhibition tests resulted in a Ct value proportional to a 10-fold dilution relative to the undiluted DNA templates, suggesting that PCR inhibition did not interfere with the amplification efficiency. DNA extraction controls and no template controls (three per qPCR plate) were run to check cross-contamination and the absence of contamination in the qPCR experiments was confirmed.

Table 3. Average standard curve qPCR amplification efficiencies and R^2 values obtained through BioRad CFX Manager 3.1 software.

Assay	Amplification Efficiency (%)	R^2
Universal *Bacteroidales* (BacUni)	95.8	0.990
Human-associated *Bacteroidales* (BacHum)	119.8	0.963
Human-associated *Bacteroidales* (HF183)	119.7	0.948
Cow *Bacteroidales* (BacCow)	101.8	0.995
Dog *Bacteroidales* (BacCan)	110.2	0.951
Chicken/Duck *Bacteroidales* (Chicken/Duck-Bac)	120.1	0.945
General *Enterococcus* (Entero1)	97.6	0.996
E. coli (EC23S857)	107.4	0.991

3.2 Fecal pollution trends in the Edwards Aquifer study area

Fecal source identification qPCR methods for general (*E. coli*, enterococci and BacUni), ruminant (Rum2Bac), cattle (BacCow), canine (BacCan), avian (Chicken/Duck-Bac and GFD), and human

(BacHum and HF183) markers were used to characterize fecal pollution trends in water samples collected from the recharge and contribution zones of the Edwards Aquifer. A total of 1120 water samples were processed in this study. The targeted fecal bacterial groups were frequently detected in water samples (Table 4); however, the swine marker (Pig-1-Bac) was detected in less than 5% of samples in the first year of sampling, and was later discontinued in consultation with the City of San Antonio.

Table 4. Detection of markers used in this study in different sites within the aquifer.

Site	% of tested water samples positive for marker										No. of samples tested
	BacUni	*E. coli*	Entero1	BacCow	BacCan	Chicken/Duck-Bac	Rum2Bac	GFD	HF183	BacHum	
1	40	83	90	19	17	6	67	29	2	0	48
2	38	87	94	15	23	2	68	13	2	6	53
3	96	93	100	66	55	59	66	91	9	14	56
4	93	95	96	71	58	64	67	95	2	13	55
5	95	96	96	87	78	80	69	91	7	7	55
6	93	94	98	50	33	19	67	91	9	9	54
7	98	95	100	66	38	11	73	98	18	23	56
8	93	93	96	61	48	33	61	83	30	39	54
9	94	96	98	56	39	17	69	93	19	19	54
10	96	86	100	20	22	4	64	86	2	4	50
11	98	98	100	58	13	9	66	96	8	15	53
12	94	98	100	67	42	33	65	94	12	15	52
13	98	96	100	65	36	20	67	95	29	44	55
14	94	100	98	50	30	15	69	94	4	9	54
15	96	100	100	57	36	15	62	98	11	19	53
16	81	84	89	42	35	35	60	82	9	9	57
17	94	98	98	78	43	33	74	94	22	24	54
18	100	100	100	70	56	56	72	96	13	20	54
19	93	96	96	63	52	36	70	91	25	27	56
21	100	100	100	46	41	11	63	94	11	17	54

The three general markers, *E.coli* (94%), enterococci (98%) and BacUni (89%) were detected in most of the samples. These assays cover a wide diversity of bacteria, including numerically dominant fecal bacteria groups, and therefore, the results suggest that the DNA was of good quality and that PCR inhibitors were removed in most samples. Among the animal-associated markers, study sites showed the highest detections for BacCan, BacCow and GFD, suggesting a larger percentage of fecal pollution to be coming from canine, cow and avian sources in the study area. Site 17 had the highest occurrence of BacCow (78%), while site 5 had the highest detection for BacCan (78%) among all the study sites. Chicken/Duck-Bac detection in water samples also varied significantly across study sites, but overall, it was detected with much less frequency than the BacCan or BacCow marker. Detection for Chicken/Duck Bac marker ranged from 2 to 80%, with highest occurrence for sites 5 and 4. The water samples yielded significantly different amplification results across study sites with the human-associated qPCR assays (Kruskal-Wallis test, $p < 0.05$). The two human-associated markers displayed similar levels, but were slightly higher for the BacHum marker at all sites. For example, detection across study sites ranged from 2 to 30% for the HF183 assay. Sites 1, 2, 4 and 10 exhibited the lowest frequency of amplification (4%), while sites 8 and 13 exhibited the highest detection frequencies (30% and 29%, respectively) for the HF183 assay. Sites 8 and 13 also exhibited the highest detection frequencies for the BacHum assay (39% and 44%, respectively). Detailed findings for each marker are presented and discussed below organized by general, ruminant, canine, avian, and human fecal pollution trends. The concentration of each marker for all sampling sites is given in Appendix B.

General markers trends. The three general indicators, *E. coli* (EC23S857), *Enterococci* (Entero1) and Universal *Bacteroidales* (BacUni) showed the highest concentrations among the qPCR markers, with detection frequency >90 % for most of the samples, and exhibited a similar spatial distribution pattern across the sampling sites (Figure 4). Higher levels of Entero1 and EC23S857 are expected as these markers include all *Enterococcus* and *E. coli* bacteria found in the gut communities of mammals and birds and are not associated with a specific host (Field & Samadpour 2007, Kapoor et al. 2018). Additionally, both bacteria may survive and grow in a wide variety of environmental habitats, such as soil and aquatic environments, with little or no input from human or animal fecal sources, provided resource availability and key abiotic conditions are propitious (Anderson et al. 2005, Whitman et al. 2003, Byappanahalli et al. 2012, Van Elsas et al. 2010). BacUni has been developed and identified as a “universal” marker sequence for the quantitative detection of all fecal *Bacteroidales*, and as such, higher levels of this marker are also expected (Kildare et al. 2007). Therefore, the sole use of these general markers for fecal source tracking in the Edwards Aquifer recharge and contributing zones is not sufficient for specific fecal source identification. The highest fecal bacterial levels based on general markers were observed for site 5 and 18. Site 5 is a pond site in the contributing zone nearby a densely populated subdivision while site 18 is on Leon Creek in the contributing zone located next to Interstate-10. Also, there is golf course nearby in the Dominion neighborhood. The runoff resulting from these activities may be a source of fecal contamination in the creek. Notably, the lowest levels of general markers were observed for well water sites (Sites 1, 2 and 10) indicating that the natural biogeochemical processes are somewhat effective in decreasing the concentrations of surface-derived microbial contaminants in the groundwater. While *E. coli* testing confirms the presence of unsafe levels of fecal pollution at several surface water sites in the Edwards Aquifer, these measurements do not specify pollution sources making it difficult to plan cost-effective remediation efforts.

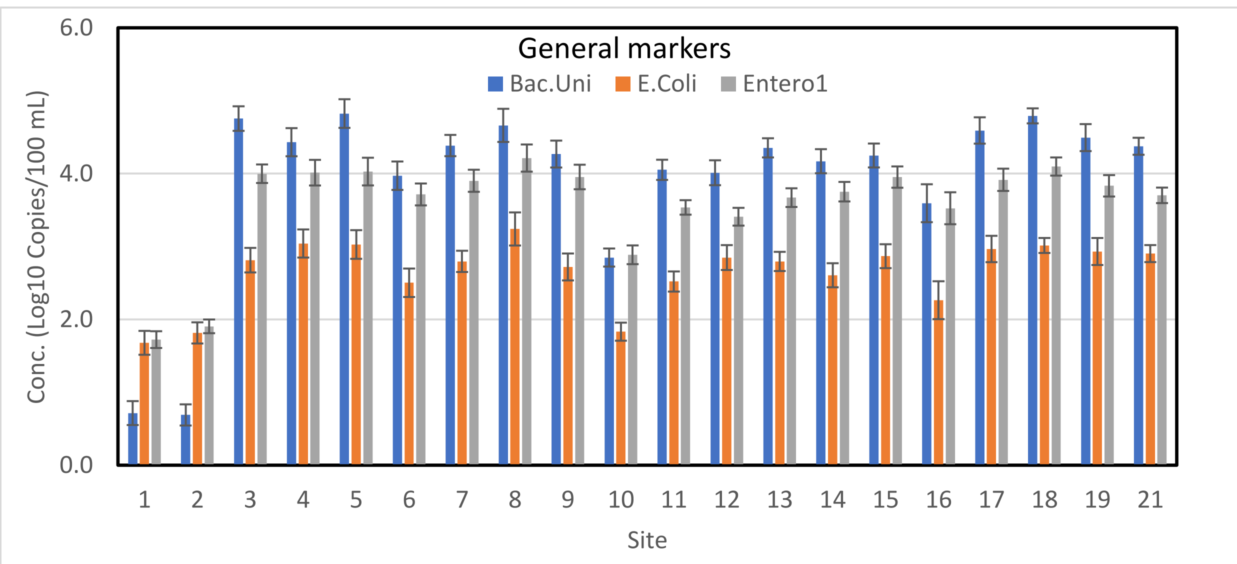

Figure 4. Spatial variation of general markers across the study sites. Bar graph represents average for each site and error bars represent standard error.

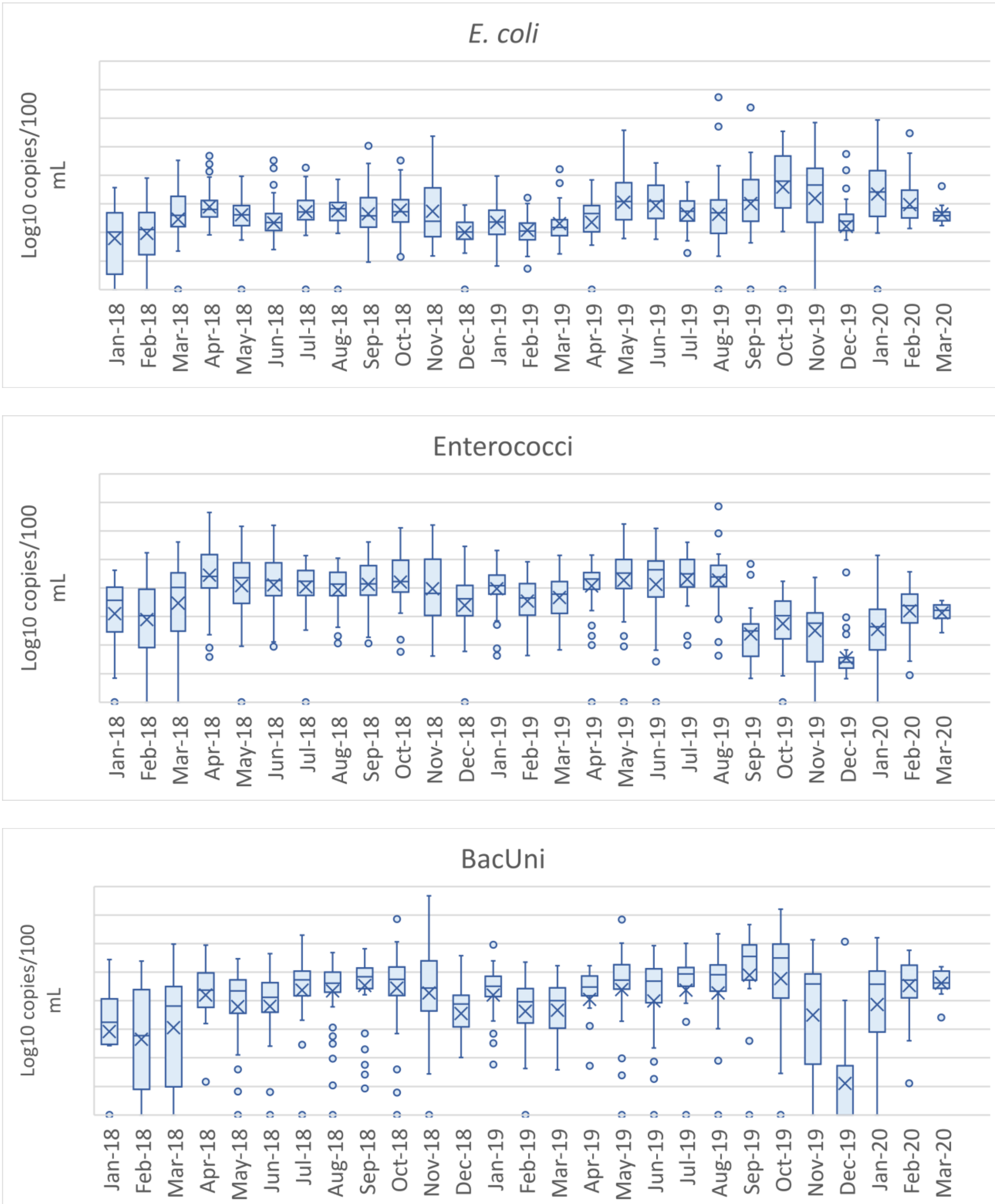

Figure 5. Temporal variation of general markers across the study period for all sites.

Ruminant fecal pollution trends. The Edwards Aquifer contributing zone within Bexar County has large number of ranches with dairy cattle population and is home to ruminant wildlife such as deers and elks. The ruminant-associated marker, Rum2Bac, was the most prevalent animal-associated genetic marker found at measurable concentrations suggesting that ruminant wildlife (cattle, deer, elk etc.) in the study area likely have a strong influence on water quality conditions (Figure 6). While Rum2Bac does not discriminate between deer, elk and cattle, BacCow genetic marker do and can therefore confirm the presence of cattle fecal pollution in a water sample. However, the concentration of BacCow marker was relatively low at well sites suggesting that the predominant source of Rum2Bac markers in well samples were deer and elks.

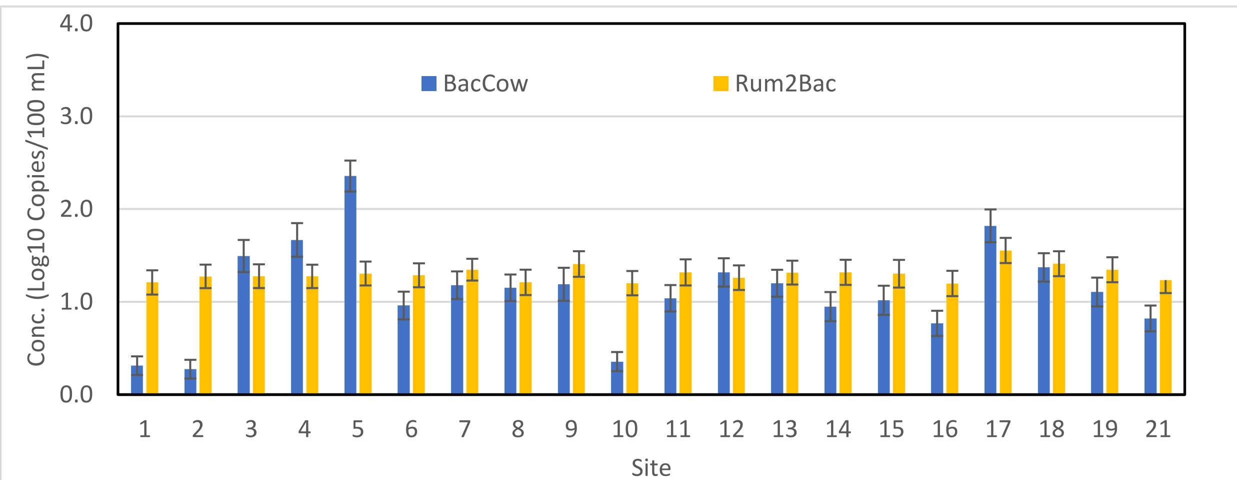

Figure 6. Spatial variation of ruminant markers across the study sites. Bar graph represents average for each site and error bars represent standard error.

The highest average Rum2Bac genetic marker concentration was found at site 17 (log_{10} 1.55 copies/100 mL). This location also had the second highest average concentration of cow-associated BacCow (log_{10} 1.82 copies/100 mL) genetic marker affirming the presence of cattle fecal pollution at this site. The site 17 catchment area includes Balcones Creek transecting a rural area having large agricultural practices. On average, higher levels of BacCow and Rum2Bac markers within the surface water sites corresponds well with the higher number of farms and ranches devoted to pasture in the study area (TDA 2019, USDA 2012). The typical ranch in Bexar County is engaged in cow/calf operation; however, other types of livestock operations found in the county include horses, sheep, goats, dairy cows and stocker calves (Bexar CAD 2019). Additionally, high BacCow marker levels could have also resulted from a lack of host specificity. A study evaluating the performance of the BacCow marker showed there is an 84% probability that a detection of the marker in a water sample is due to actual contamination from the respective host, and 38% of horse fecal extracts tested positive for the BacCow marker (Somnark et al. 2018, Kildare et al. 2007). Therefore, high detections for BacCow observed in the study area could be attributed to the large number of animal ranchlands surrounding the Balcones Creek sites and detections for BacCow observed at the developed sites could be attributed to false-positive results from non-cow fecal sources due to a lack of host specificity.

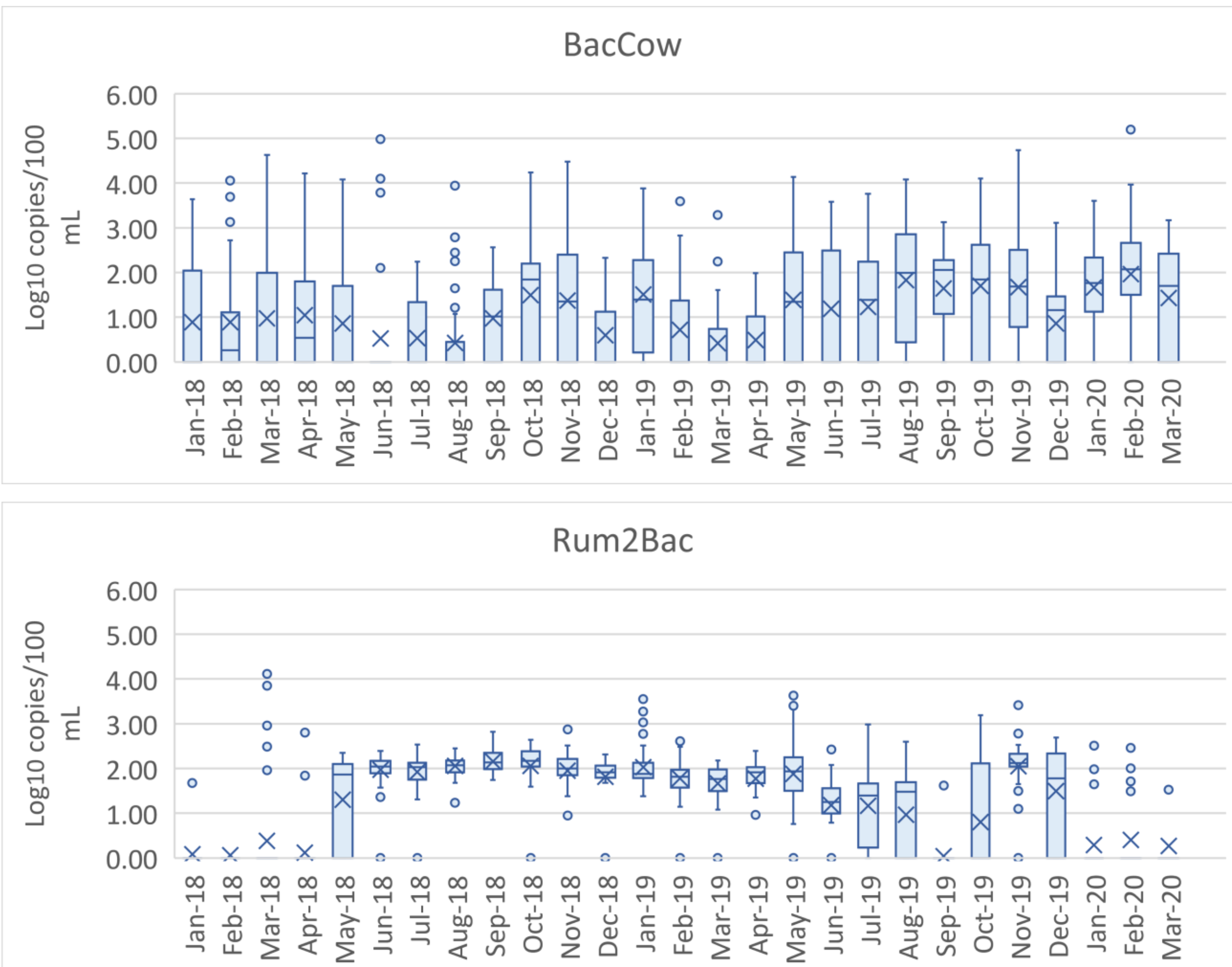

Figure 7. Temporal variation of ruminant markers across the study period for all sites.

Canine fecal pollution trends. Prior to this study, the extent to which dogs contribute to fecal pollution in the Edwards Aquifer recharge and contributing zones was unknown. While dog waste is thought to represent a lower public health risk compared to human and cattle sources, it does harbor *E. coli* making it a potential contributor to surface water impairment. Highest levels of BacCan genetic marker were observed for the pond sites (Sites 3, 4 and 5; Figure 8) that could be attributed to the large number of dogs associated with pet owning households in the nearby area as well as the unrestrained dog population in the City (AVMA 2019, City of San Antonio 2019). Site 5 exhibited a consistently high concentration of the BacCan marker between April 2018 and June 2018, and Oct 2018 and January 2019 clearly suggesting a water quality impact from pet waste management activities. Higher levels of BacCan marker were also observed for the Leon Creek sites (Sites 18 and 19) located near the Dominion neighborhood. The management of dog waste in San Antonio is left up to voluntary owner responsibility. Others report that that community education programs about good pet waste management practices can improve the water quality in areas impacted by canine fecal pollution

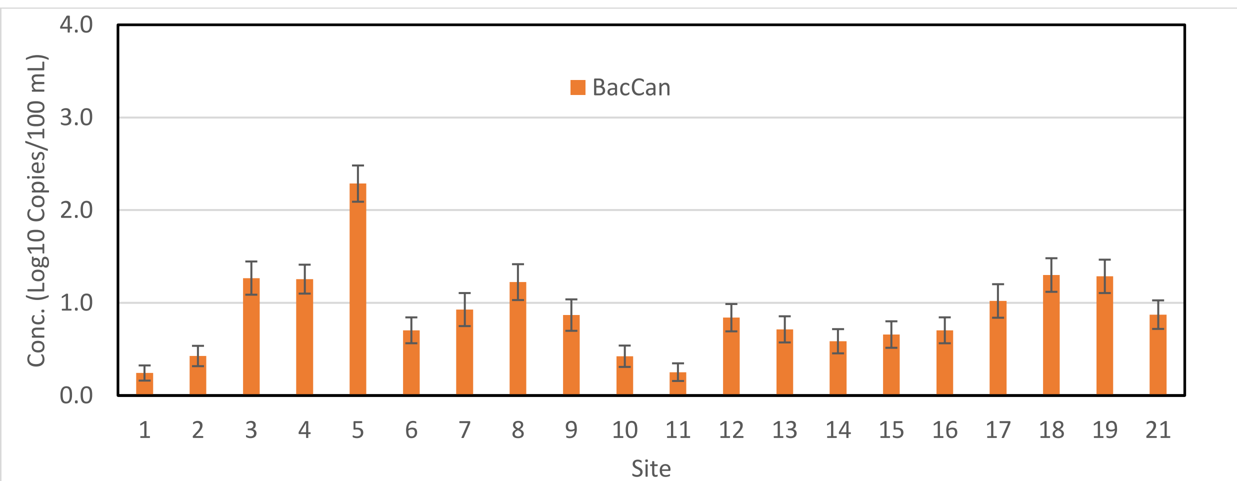

Figure 8. Spatial variation of canine marker across the study sites. Bar graph represents average for each site and error bars represent standard error.

The BacCan marker exhibited higher concentrations during the end of fall/early winter (Figure 9). Temporal changes in BacCan marker levels can be attributed to pet ownership and pet waste management practices in the Bexar County study region. In this area, it is more common for pet owners to keep their pets indoors during the winter and summer and spend more recreational time with pets outdoors during the fall and spring seasons. Studies focusing on Texas show that the highest physical activity occurs during the spring, while declines in physical activity can be attributed to the shorter days and adverse weather conditions associated with winter and hot and humid conditions during the summer months (Tucker et al. 2007). Additionally, Leon Creek is adjacent to the Leon Creek greenway, 20 miles of paved multi-use hike and bike trails following the Leon Creek in north and central San Antonio. This creek-side trail allows access to dogs on a leash and offers a variety of activities, including dog walking. High levels of dog fecal waste could be associated with the large number of pets and unrestrained dog population in the study area, as well as an increase in seasonal outdoor recreational activities, specifically along the Leon Creek and pond sites.

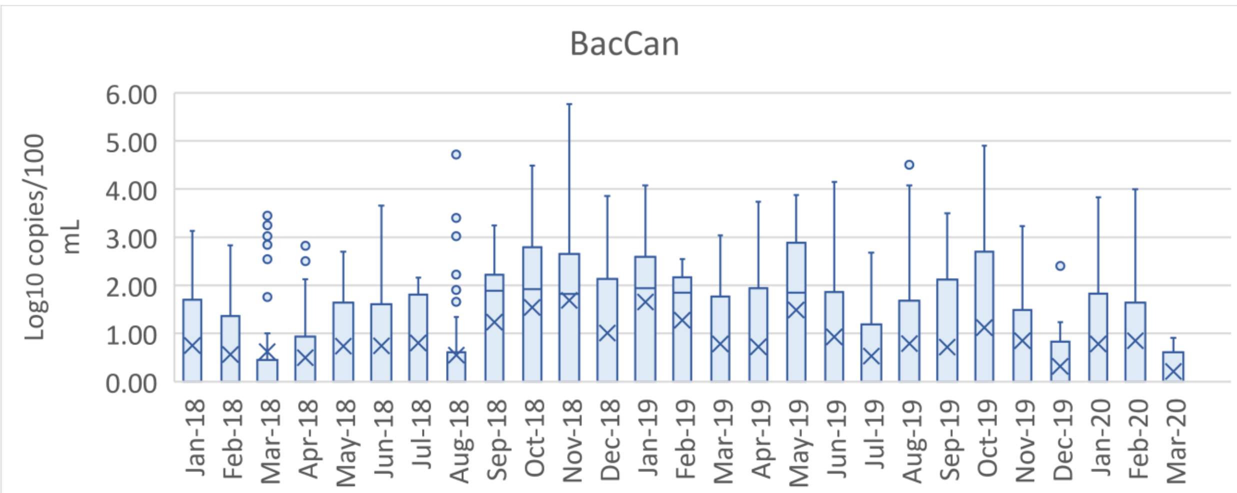

Figure 9. Temporal variation of canine marker across the study period for all sites.

Avian fecal pollution trends. Fecal waste from bird species can harbor general fecal indicator bacteria such as *E. coli*, as well as a range of pathogens that can potentially infect humans and contribute to poor water quality. Bexar County is home to several resident bird species, as well as numerous seasonal populations that migrate from the south into the north reaches during the early spring and summer that could impact local water quality. Two avian-associated genetic markers (GFD and Chicken/Duck-Bac) were used to reveal information about the potential influence of bird fecal waste in surface waters. Chicken/Duck-Bac detection in water samples varied significantly across study sites, but overall, it was detected with much less frequency than the GFD avian marker. Detection for Chicken/Duck Bac marker ranged from 2 to 80%, with highest occurrence for sites 5 and 4. Notably, both sites 4 and 5 are pond sites which are visited by birds and dog populations frequently. The Chicken/Duck-Bac marker was consistently higher in the pond sites as compared to creek sites (Figure 10). This may be attributed to the more frequent visitation of ducks and other birds at the pond sites. The concentrations of the GFD genetic marker were relatively higher at all surface water sites (except for well sites) suggesting that bird fecal pollution is a major source of concern for Edwards Aquifer. Additionally, higher levels of the avian markers were observed during the spring and fall seasons that can be attributed to bird migration patterns (Figure 11). Texas is located directly in the center of the Central Flyway, a well-defined route for migratory birds. According to a publication by the Texas Parks and Wildlife, Texas has recorded more species of birds than any other state (over 615 species), most of which are migrant (Shackelford et al. 2005). Increases in migrant birds are observed during the spring season on their transit northward and fall season during their southward passage through Bexar County, Texas.

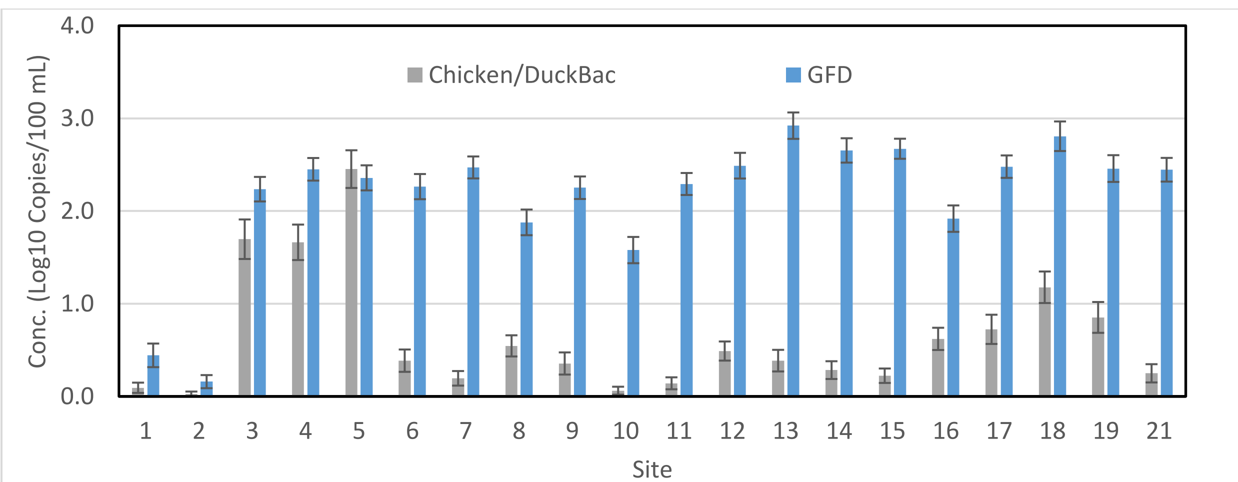

Figure 10. Spatial variation of avian markers across the study sites. Bar graph represents average for each site and error bars represent standard error.

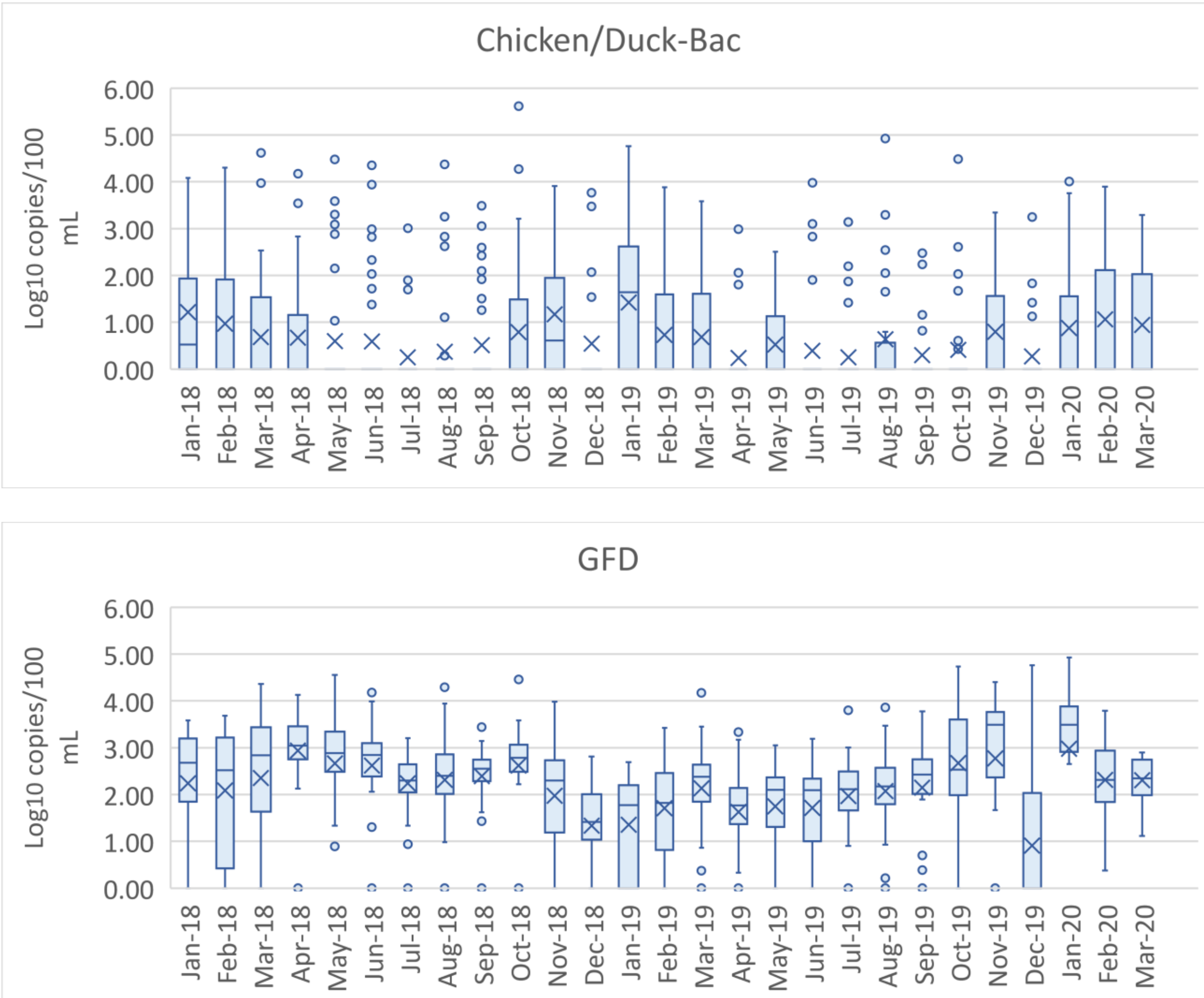

Figure 11. Temporal variation of avian markers across the study period for all sites.

Human fecal pollution trends. Human waste can potentially enter local waters in the Edwards Aquifer from sanitary sewer overflows, wastewater treatment plants, public campgrounds, potential stormwater sewer cross-connections, faulty onsite septic systems, seasonal portable restrooms at local parks, and transient camps. In addition to containing *E. coli*, these human waste sources can harbor numerous pathogens (e.g., *Shigella sonnei*, noroviruses, and *Cryptosporidium*), solids, debris, and a variety of pollutants (i.e., antibiotics, hormones, caffeine, steroids, metals, and synthetic organic compounds). The incidence of human fecal pollution in the study area was less severe compared to ruminant and avian sources, but a closer investigation reveals several interesting patterns. The two human-associated markers (HF183 and BacHum) displayed similar levels, but were slightly higher for the BacHum marker at all sites (Figure 12). For example, detection across study sites ranged from 2 to 30% for the HF183 assay. Sites 1, 2, 4 and 10 exhibited the lowest detection frequency (2%), while sites 8 and 13 exhibited the highest detection frequencies (30% and 29%, respectively) for the HF183 assay. Sites 8 and 13 also exhibited the highest detection frequencies for the BacHum assay (39% and 44%, respectively).

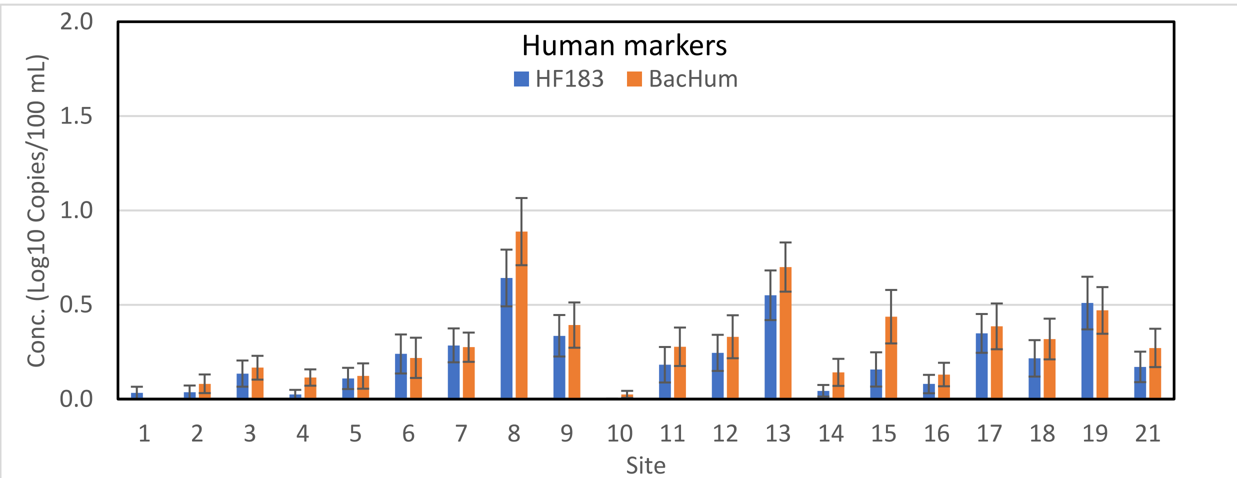

Figure 12. Spatial variation of human markers across the study sites. Bar graph represents average for each site and error bars represent standard error.

Results from this study were consistent with previous studies which showed higher marker levels for BacHum in relation to HF183 (Tanvir Pasha et al. 2019, Kapoor et al. 2018). The recent public release of USEPA draft method for HF183 assay (USEPA 2019) has led to an increased interest in the use of these qPCR based markers to characterize fecal pollution in environmental waters polluted by sewage. Studies comparing host specificities among different fecal sources for the HF183 and BacHum markers, have shown that both assays were equally sensitive to sewage (Shanks et al. 2010), but BacHum showed substantially more false-positive results for non-human fecal sources such as cat, dog, gull, and raccoon feces (Jenkins et al. 2009, Van De Werfhorst et al. 2011, Boehm et al. 2013). Although human-associated markers were detected at most sites over the study period, majority of the samples (83 %) exhibited no evidence of these markers suggesting that human fecal pollution was not a concern for these sites (Figure 13). Surface water sites located near septic tanks exhibited higher concentrations for the two human-associated markers. Studies have shown a close relationship between septic tanks and human fecal pollution as these systems have been identified as significant sources for emerging contaminants in groundwater and surface water (Gao et al. 2019, Hinkle et al. 2005, Schaider et al. 2016, Phillips et al. 2015).

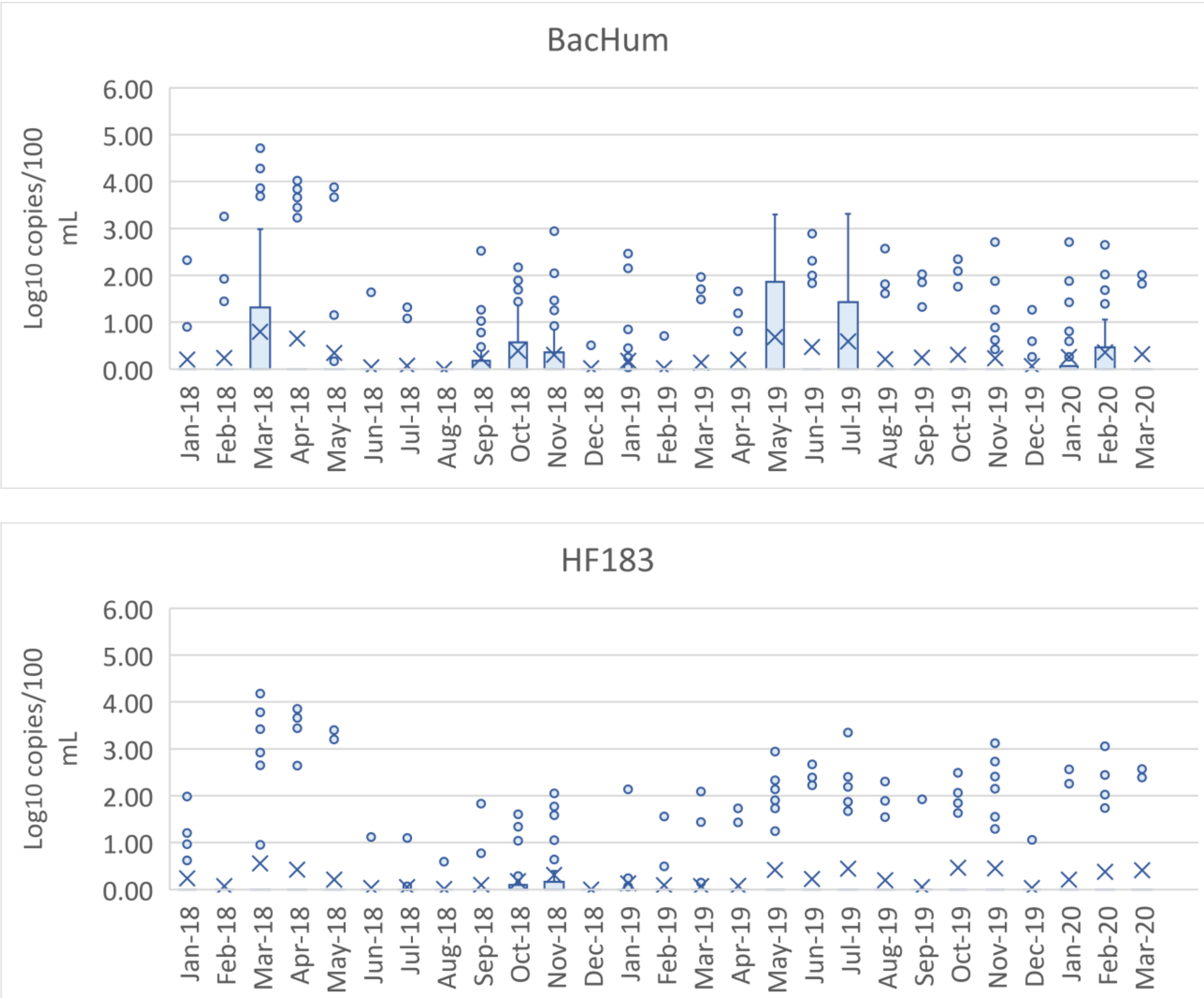

Figure 13. Temporal variation of human markers across the study period for all sites.

3.3 Water quality and correlation of markers

Water quality parameters measured at each site during the two-year study period are summarized in Table 5. The average water temperature at the study sites ranged from 20.2 to 23.3°C, and the pH ranged from 6.87 to 8.45. The groundwater sites had higher average nitrate concentrations compared to surface water sites. Among the surface water sites, site 19 had the highest average nitrate concentration (0.689 mg/L NO_3^-N). Site 19 is located in Leon Creek nearby highly populated area in Bexar County. Nitrite and ammonia concentrations were generally low for all sites. Spearman's rank correlation coefficients were calculated between each pair of qPCR markers and water quality parameters. All statistical analysis outcomes among the markers were regarded as significant at $p < 0.05$. Spearman's rank correlation analysis showed an overall strong correlation between the three general markers, BacUni, *E. coli* and Entero1, with the strongest correlations between Entero1 and BacUni (0.69) and Entero1 and *E. coli* (0.68) (Figure 14). For the host-associated markers, HF183 and BacHum showed a strong correlation (0.63), as both markers have been developed for specificity to human fecal waste (Haughland et al. 2010, Kildare et al. 2007). Results from this study are consistent with multiple previous studies that have shown the HF183 and BacHum markers to have similar sensitivity (Jenkins et al. 2009, Odagiri et al. 2015, Kapoor et al. 2018). Correlations among the other host-associated markers varied from weak to moderate. The host-associated indicators (HF183, BacHum and BacCan) also had stronger correlations with nitrate (0.27, 0.33, 0.36, respectively) and nitrite (0.30, 0.23, 0.15, respectively) nitrogen concentration than exhibited by the general indicators. These findings suggest that Bacteroidales markers are better indicators of fecal pollution compared to the conventional indicators. Nitrate has been reported as a strong predictor for the presence of human-associated markers and consistent with our study, the major source of nitrate can be attributed to human fecal pollution (Badgley et al. 2019). Additionally, given that the study area do not contain wastewater treatment plant outfalls, this result raises the concern about leaking subsurface wastewater infrastructure and/or septic tanks. Furthermore, dog fecal contamination may also be attributed as a major source of nitrogen pollution in creeks as demonstrated by positive correlation between BacCan marker levels and nitrate/nitrite concentrations.

For the water quality parameters, the temperature and DO showed moderate negative correlation (-0.44), while nitrite and nitrate had moderate positive correlation (0.46). The negative correlation between temperature and DO concentration demonstrates their interdependence, whereas the positive relationships observed between levels of nitrite and nitrate suggest a common source that may be sewage pollution. Notably, pH and ammonia did not correlate with any of the qPCR markers or other water quality parameters. One possible reason for the lack of associations could be the considerably less variation of water pH throughout the study period (average pH 7.6 ± 0.5) and high number of samples with no ammonia detected. It is important to note that different water quality parameters (e.g., temperature, nitrate, DO, and pH) are expected to vary widely with regard to runoff from surrounding land-use, which may also be related to varying pollution sources for various indicator types.

Table 5. Average values (± standard deviation) of water quality parameters at each sampling site during the study period.

Site	Water Temp (°C)	pH	Dissolved oxygen (mg/L)	Nitrite, NO_2^--N (mg/L)	Nitrate, NO_3^-N (mg/L)	Ammonia, NH_3^-N (mg/L)
1	22.7 (3.4)	6.88 (0.92)	7.69 (1.13)	0.001 (0.003)	3.155 (0.796)	0.043 (0.302)
2	22.9 (3.1)	6.96 (0.92)	7.60 (1.18)	0.000 (0.002)	2.008 (0.455)	0.033 (0.261)
3	22.5 (7.0)	8.12 (1.42)	8.49 (3.91)	0.010 (0.030)	0.268 (0.485)	0.019 (0.050)
4	22.2 (7.2)	7.84 (1.28)	7.76 (2.38)	0.012 (0.030)	0.188 (0.198)	0.034 (0.099)
5	23.1 (7.0)	8.45 (0.90)	10.30 (3.74)	0.043 (0.237)	0.185 (0.249)	0.028 (0.099)
6	23.3 (7.0)	7.75 (0.55)	11.16 (2.53)	0.016 (0.059)	0.359 (0.746)	0.047 (0.173)
7	22.0 (7.1)	7.74 (1.18)	8.44 (3.16)	0.007 (0.020)	0.208 (0.271)	0.042 (0.086)
8	21.4 (5.6)	7.32 (0.37)	6.78 (2.26)	0.022 (0.078)	0.379 (0.279)	0.043 (0.088)
9	21.4 (6.4)	7.71 (1.28)	7.16 (2.91)	0.017 (0.064)	0.383 (0.405)	0.019 (0.033)
10	22.6 (1.8)	6.87 (0.29)	5.46 (0.76)	0.005 (0.031)	0.090 (0.148)	0.156 (0.081)
11	21.9 (5.3)	7.74 (0.39)	8.99 (1.33)	0.001 (0.003)	0.139 (0.206)	0.012 (0.027)
12	20.8 (5.9)	7.67 (1.16)	9.05 (2.07)	0.001 (0.004)	0.218 (0.269)	0.016 (0.034)
13	20.2 (5.0)	7.55 (0.35)	7.07 (1.95)	0.002 (0.006)	0.473 (0.475)	0.024 (0.044)
14	21.8 (5.8)	7.66 (0.43)	9.25 (1.34)	0.002 (0.005)	0.203 (0.221)	0.007 (0.010)
15	20.9 (6.2)	7.53 (0.35)	7.56 (2.93)	0.004 (0.010)	0.306 (0.301)	0.029 (0.061)
16	22.7 (6.5)	7.71 (0.37)	9.48 (2.88)	0.005 (0.010)	0.283 (0.381)	0.019 (0.039)
17	23.2 (7.1)	7.94 (0.39)	11.53 (3.20)	0.005 (0.011)	0.280 (0.369)	0.027 (0.114)
18	22.7 (5.8)	7.48 (1.13)	9.90 (3.36)	0.013 (0.026)	0.542 (0.600)	0.030 (0.058)
19	22.1 (6.0)	7.35 (1.49)	7.82 (2.46)	0.013 (0.026)	0.689 (0.710)	0.014 (0.025)
21	20.6 (6.6)	7.68 (1.89)	8.81 (2.27)	0.021 (0.085)	0.591 (0.491)	0.034 (0.079)

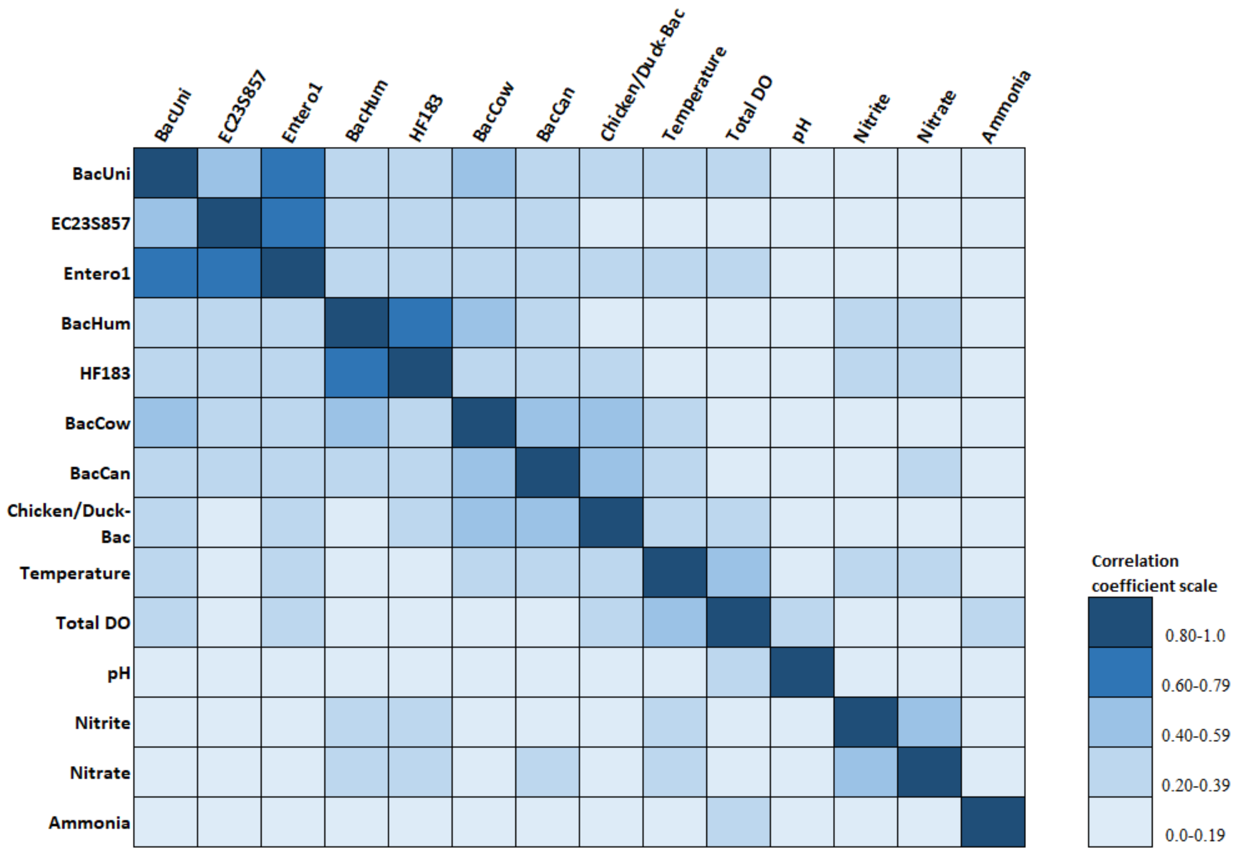

Figure 14. Heat map of Spearman's rank correlation coefficients matrix for qPCR markers and water quality parameters.

3.4 Effect of Stormwater on Marker Concentrations

Over the duration of the study, a total of 13 storm event-related samples were collected at the surface water sites to determine peak concentration loadings before, during and after storm event. Stormwater samples were collected during storm events using a Nalgene Storm Water Sampler and Mounting Kit (Thermo Scientific). For comparison purposes, samples collected during storm events were defined as "during", while samples collected before and after the rain were classified as "before" and "after", respectively. The results from non-parametric one-way analysis of variance (Kruskal-Wallis method) indicated that there is a significant difference (p-value > 0.05) between mean concentrations in at least two of the storm events for four markers: *E. coli*, Entero1, BacHum, and BacCan. No significant difference (p-value < 0.05) in the mean concentrations was observed for BacUni, HF183, BacCow, Chicken/Duck-Bac, Rum2Bac, and GFD markers. Figure 15 shows the spatial variation of the average general and host-associated qPCR marker concentrations for before, during, and after stormwater events. In general, there was an increase in concentration for all fecal markers after the rain suggesting that storm events can significantly increase fecal pollution in the water bodies over the Edwards Aquifer. Among the general markers, *E. coli* and Entero1 markers showed a substantial increase during and after rain events, while the BacCan marker displayed highest concentration difference between before and after rain samples among the animal markers. Concentrations were highest for the stormwater samples across nine of the ten qPCR markers, signifying that peak concentration loadings occurred during storm-related events (Lee et al. 2014, Kapoor et al. 2018, Parker et al. 2010). The largest differences, greater than 1-log_{10} copies/100 mL, in marker concentrations between before and after stormwater samples occurred between BacCan and *E. coli*, suggesting large fluxes of increased canine fecal waste during storm events in the study area. The two human-associated markers showed increases from before to after rain, however, differences in concentrations were significant only for BacHum, and not for HF183.

Figure 15. Average concentrations for fecal markers for before, during and after stormwater events.

3.5 Proximal land use characterization analysis and spatial variation between Leon Creek and Balcones Creek marker concentrations

Factors influencing fecal contamination sources and levels differ between urban and rural areas (Ridley et al. 2014). For this study, an analysis of proximal land use using 1-km buffer zones surrounding Balcones and Leon creeks was performed in order to characterize the study sites based on septic tanks, wastewater collection infrastructure, human population, and developed vs. undeveloped land (Figure 16). In addition, land use parameters were also estimated for each site individually (Table 6) and correlated with marker occurrence at each site (Table 7). SAWS and the City of Fair Oaks Ranch underground wastewater collection infrastructure data was analyzed and estimated to service approximately 22% area of the Balcones Creek buffer zone and 75% of the Leon Creek Buffer zone. Additionally, the Leon Creek buffer zone contained a higher amount of impervious cover (>50%) relative to the buffer zone surrounding Balcones Creek (<10%). Human population for the Balcones Creek buffer zone was estimated at 202 people/km^2, while the Leon Creek buffer zone population was estimated to be more than 6.5 times greater, at 1,355 people/km^2. The number of active septic tanks for Balcones and Leon Creek buffers totaled 421 and 304 tanks, respectively. The septic tank density for the Balcones creek buffer zone is 27 tanks/km^2 and 10 tanks/km^2 for the Leon Creek buffer zone, indicating that tanks cover larger area in the Balcones buffer that is approximately 2.7 times greater than the Leon Creek buffer.

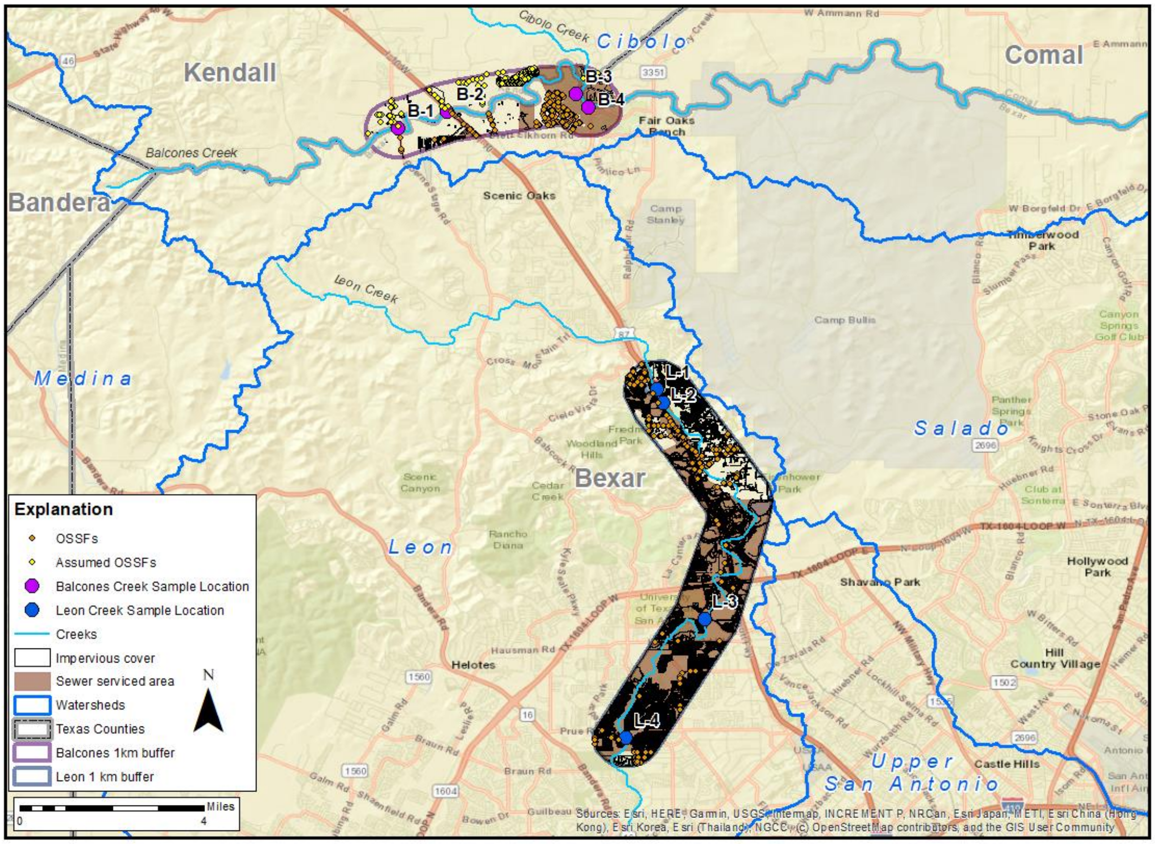

Figure 16. Proximal land use characterization analysis for the study area including 1-km buffer zones surrounding Balcones and Leon Creek and features of interest.

Table 6. Land-use variables within 1 km radius of study sites.

Site	Septic tank density (tanks/km^2)	Human population	Impervious surface (%)	Distance to nearest sewer main (km)
B-1	10	850	6.00	1.45
B-2	9	722	6.00	0.41
B-3	39	452	7.00	<0.10
B-4	36	801	7.00	<0.10
L-1	30	1488	15.00	<0.10
L-2	30	1488	15.00	<0.10
L-3	1	4401	31.00	<0.10
L-4	15	16776	32.00	<0.10

Table 7. Correlation coefficients between marker occurrence and land use.

	BacUni	BacHum	HF183	BacCow	BacCan	Chicken /Duck Bac	Entero1	E. coli
Septic tank density	-0.44	0.27	0.55	0.47	0.52	0.75	-0.59	-0.59
Human population	0.14	0.39	0.38	-0.29	-0.25	-0.09	-0.14	-0.14
Developed land	0.32	0.44	0.22	-0.66	0.07	-0.24	-0.03	-0.03
Undeveloped land	-0.37	-0.38	-0.26	0.27	0.16	0.00	-0.10	-0.10

A comparison of the average general and host-associated marker concentrations (log_{10} copies/100 mL) between Leon and Balcones Creeks is represented in Figure 17. General marker levels were similar across both creeks and showed the highest concentrations among all the qPCR markers, with concentrations above 4-log_{10} copies/100 mL for Entero1 and BacUni assays. All host-associated marker levels were detected at lower concentrations (below 3-log_{10} copies/100 mL), with the lowest levels detected for the two human-associated markers. Differences in marker levels between the Leon area and Balcones areas were evident for each host-associated marker. Leon Creek displayed higher levels for all markers, except BacCow. Higher concentrations in Leon Creek for the two human-associated markers could be attributed to the larger human population served by septic tanks or sewer infrastructure estimated for the Leon Creek buffer zone (Table 6). Both markers were positively correlated to the septic tank density, human population and the percent of impervious surface (Table 7). Studies have shown a close relationship between OSSFs and human fecal pollution as these systems have been identified as significant sources for emerging contaminants in groundwater and surface water (Gao et al. 2019, Hinkle et al. 2005, Schaider et al. 2016, Phillips et al. 2015). BacHum and HF183 concentrations were lowest for sites B-1 and L-3. The areas surrounding these two sites are occupied by the fewest number of septic tanks (Figure 16). Furthermore, these sites are located furthest from underground wastewater collection mains (Table 6), which could also explain the relatively low levels of detection for human fecal waste. An increase in septic tank density is observed surrounding sites B-3, L-1 and L-4, which showed the highest levels of both human-associated markers. Therefore, relatively higher levels at these three sites could be attributed to leaking septic tanks or breaks in underground sewer infrastructure.

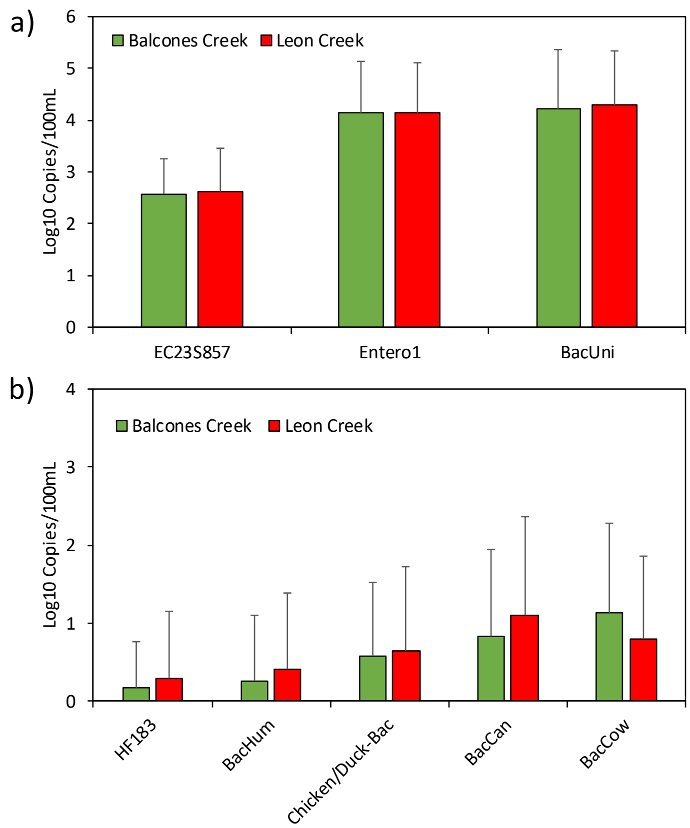

Figure 17. Comparison between Balcones Creek and Leon Creek sites for a) general indicators and b) host-associated markers using qPCR assays.

Concentrations for BacCan were higher for Leon Creek sites, while the Balcones Creek showed higher concentrations for the BacCow marker. Higher levels of BacCan within the Leon Creek area could be attributed to the large number of dogs associated with pet owning households as well as the unrestrained dog population in the City (AVMA 2019, City of San Antonio 2019). Higher levels of BacCow marker within the undeveloped Balcones sites corresponds well with the higher number of farms and ranches devoted to pasture in that area (TDA 2019, USDA 2012). The typical ranch in Bexar County is engaged in cow/calf operation; however, other types of livestock operations found in the county include horses, sheep, goats, dairy cows and stocker calves (Bexar CAD 2019). Additionally, high BacCow marker levels could have also resulted from a lack of host specificity as mentioned earlier (Somnark et al. 2018, Kildare et al. 2007). Therefore, high detections for BacCow observed in the study area could be attributed to the large number of animal ranchlands surrounding the Balcones Creek sites and detections for BacCow observed at the Leon Creek sites could be attributed to false-positive results from non-cow fecal sources due to a lack of host specificity.

Among the Balcones Creek sites, site B-3 showed the highest concentrations for BacCan, BacCow and the two human-associated markers, HF183 and BacHum. The Cibolo Creek, which flows south through Kendall County, converges with Balcones Creek approximately 1 mile upstream of site B-3 and has a history of significant fecal contamination often exceeding the state's water quality standards for safe contact recreation. A previous study suggested that human fecal contamination may likely occur as a result of treated effluent discharged into Cibolo Creek from two wastewater treatment plants (WWTPs) located in Boerne, Texas (Tanvir Pasha et al. 2020). Thus, relatively high concentrations for the host-associated markers at Site B-3 may be attributed to the contribution of fecal pollution from Cibolo Creek.

3.6 Implications for the Edwards Aquifer

Currently, there is an incomplete understanding of the sources of fecal pollution found in the watersheds of the Edwards Aquifer, a primary source of drinking water to the City of San Antonio and its surrounding communities. Segments of both the Upper Cibolo Creek, which converges with Balcones, and Leon Creek, both of which flow through the Aquifer recharge and contributing zones, have been identified as impaired (USEPA 2014). Bacterial pathogens have been determined to be the number one cause of river and stream impairments in Texas (TCEQ 2019, EPA 2014). This study is the first to examine and compare fecal contamination at surface water sites in the recharge and contributing zones of the Edwards Aquifer within Bexar County using a molecular MST approach targeting Bacteroidales 16S rRNA genes for host-associated qPCR assays. The presence of Bacteroidales in most of the water samples suggested that methods targeting this bacterial order (or group) can track sources of fecal pollution in environmental waters. Host-associated Bacteroidales signals were detected at all sites, although their relative occurrence varied by site and by month.

The microbial source tracking study undertaken in the Edwards Aquifer region confirms that fecal pollution is present on the landscape and is highly reflective of the architecture of the environment.

The predominant source of fecal pollution in the watershed is from bird and ruminant-derived sources. Other individual sources that encompassed lesser proportions of fecal pollution included dog wastes and human-derived wastes. In the surface waters, the concentration of fecal sources increased during the wet periods, indicating the possible influence of runoff during stormwater events. One of the goals of the study was to examine the primary sources of fecal contamination at specific sites as well as determine any site specific differences. While the predominant source of bacterial contamination at all stations was bird-derived, there were several site-specific differences noted. Sites 5, 17 and 18 had a significantly higher proportion of fecal contamination compared to other sites. Site 18 is located in the Leon Creek close to Interstate-10, which is a very busy road. Also, there is a residential neighborhood nearby and a golf course uphill. The runoff resulting from these activities may be a source of fecal contamination in the creek. Site 5 is a large pond/lake located right outside residential subdivision with septic tanks, which may contribute to fecal contamination during overflow events.

We initially selected *E. coli* O157:H7 as a pathogen target, since the high populations of *E. coli* had been previously observed and may yield an infectious strain. Following more than a year of testing via multiplex PCR, we were unable to detect any pathogenic *E. coli* O157:H7. Multiplex PCR amplifies more than one gene simultaneously; in this case, we were testing for three genes that are specifically present in pathogenic *E. coli* O157:H7. These genes include: O157, coding for a cell wall epitope; H7, a flagellar epitope; and intimin, which is required for *E. coli* to adhere to the intestinal epithelium. While we observed positive results for both cell epitopes, the presence of DNA containing the intimin gene was never observed.

Each of the methods employed here had strengths and weaknesses, and in general, MST methods are still evolving and improving. Currently there is no standard method that is appropriate to answer all MST queries. The Bacteroidales qPCR was fairly comprehensive and answered questions about dominant sources and the variety of sources of fecal pollution in the watershed. While some of these sources may be difficult to manage, such as animal wastes, the study also provided evidence of sources in the watershed that can be controlled or mitigated for, such as human-derived sources that are a public health risk. This information will provide the basis for continued education and public outreach regarding sources of bacterial contamination in the watershed. The scientific information gained from this study will allow the opportunity for resource managers to re-evaluate and modify current on-site septic system management programs to specifically target resources toward actions that will be most effective at reducing bacteria levels in the watershed. While these results should not be extrapolated to other geographic regions, information gained from the use of these tools, the overall approach and lessons learned can be applied to other watersheds.

Human and animal fecal contaminants are among the major concerns for public health since pathogens could be present. The *Bacteroidales* marker assays, when combined with land use and weather information, can allow for a better understanding of the sources and fluxes of fecal inputs in urban and rural areas, which can help us to understand when these fecal sources may impact water quality. Data collected from the present study were used to identify and quantify the sources of human and animal fecal pollution and perform an analysis of temporal and seasonal trends. It has been determined that the major fecal contributing sources differ between urban and rural areas. In our study area, major urban sources include sewage overflows and dog waste, while rural sources include waste from livestock facilities and farms, and leaking septic tanks. Effective mitigation measures should be directed towards public outreach efforts to educate pet owners on the negative effects of pet waste on water quality and the importance of proper pet waste disposal, as well as efforts to control and reduce the large unrestrained dog population in the City. Efforts to mitigate fecal pollution outside of the City should focus on the implementation of BMPs aimed at controlling and mitigating waste associated with animal ranches, agricultural practices and wildlife. Future studies focusing on a more detailed assessment of rural and urban areas associated with clusters of OSSFs and underground sewer infrastructure are recommended to determine appropriate measures for mitigating human fecal pollution from these sources.

The information provided in this report will assist City of San Antonio with pollution source location and best management practices for mitigating fecal bacterial contributions to the Edwards Aquifer.

4.0 CONCLUSIONS AND RECOMMENDATIONS

The following conclusions have been drawn from this study for the recharge and contributing zones of the Edwards Aquifer within Bexar County:

- The three general markers (*E. coli*, enterococci and BacUni) exhibited highest levels and a similar spatial distribution pattern across the sampling sites. Among the host-associated markers, low levels were observed for the human-associated markers and the highest levels for Rum2Bac/BacCow and GFD, suggesting a higher percentage of fecal pollution to be coming from the large bovine/ruminant and bird populations in the study area.

- The predominant sources of fecal contamination identified in the Edwards Aquifer study area were, in ranked decreasing order of presence: avian including gull, ducks etc. (85%), ruminant including cattle and deer (67%), dog (40%) and human-derived (17%).

- The cow/ruminant marker concentrations were higher for Balcones Creek sites due to farms and ranches in the area. Higher levels of canine-derived contamination was observed for pond sites near residential areas and Leon Creek sites, that can likely be attributed to poor pet waste management practices in the area.

- The concentrations of the GFD genetic marker were relatively higher at all surface water sites (except for well sites) suggesting that bird fecal pollution is a major source of concern for Edwards Aquifer. The Chicken/Duck-Bac marker was consistently higher in the pond sites as compared to creek sites.

- Human-associated *Bacteroidales* markers were detected mostly at surface water sites near densely populated urban areas and/or rural areas with high septic tank density, suggesting that their presence is the result of larger human population served by septic tanks or sewer infrastructure.

- One of the most important findings of the study was the absence of pathogenic *E. Coli* O157:H7 from the water samples collected from all the sampling sites.

- The highest fecal bacterial levels based on general markers were observed for site 5 and 18. Site 5 is a pond site in the contributing zone nearby a densely populated subdivision while site 18 is on Leon Creek in the contributing zone located next to Interstate-10. Also, there is golf course nearby in the Dominion neighborhood. The runoff resulting from these activities may be a source of fecal contamination in the creek.

- The lowest fecal bacterial levels were observed for well water sites (Sites 1, 2 and 10) indicating that the natural biogeochemical processes are somewhat effective in decreasing the concentrations of surface-derived microbial contaminants in the groundwater.

- Chicken/Duck-Bac, BacCan and BacCow exhibited higher concentrations during the spring season and the end of fall/early winter and were all lowest during the summer months.

- The surface runoff resulting from wet weather day event is a major contributor of fecal contamination in the creeks during storm events.

- There were strong correlations between the general indicators and between the human-associated markers. Temperature and dissolved oxygen, and nitrite and nitrate showed moderate correlations. pH and ammonia did not correlate with any of the qPCR targets or other water quality parameters.

- Data from this study have established a baseline for fecal pollution sources in the Edward Aquifer recharge and contributing zones and can be used for the recommendation and implementation of best management practices that can accurately and cost effectively prevent, control, and remediate fecal pollution events and maintain water quality.

Recommendations for the City of San Antonio include:

- Secure resources/funding to implement necessary improvements in management programs and enforcement mechanisms that will mitigate the public health risk by reducing animal and human-derived sources and other readily controllable sources of fecal contamination, including:
    - Domestic Pet Waste - Education and outreach to homeowners regarding proper disposal of domestic pet waste.
    - Urban Wildlife Populations – Education and outreach to homeowners about practices that discourage attraction of urban wildlife, particularly deers.
    - Bird Fecal Waste – Identify birds that are polluting the water and develop bird relocation efforts to reduce hazards associated with large bird populations.
    - On-Site Septic Systems
        - Ongoing homeowner education regarding septic system maintenance and homeowner inspections of septic systems.
        - Investigate, identify, and repair or replace problematic septic systems in the contributing zone.

- Improve storm water management programs, including the promotion of Low Impact Development (LID) such as the reduction of effective impervious surfaces, dispersion of storm water runoff to vegetated areas, and Best Management Practices that are appropriate to the site-specific conditions.

- Use the results from this study to evaluate current wastewater infrastructure and on-site septic system management programs and water quality monitoring plans in the recharge and contributing zones. Re-examine implementation strategies and modify if necessary to achieve long-term water quality objectives.

- Implement a change in drainage architecture which supports a more diverse biological habitat around the creeks that could produce a reduction in downstream bacterial input. For example, pouring of concrete channels around the creek to avoid runoff from directly entering the stream.

- Continue outreach (including dissemination of related study results) to the public about nonpoint source pollutant sources and steps that can be taken to mitigate those sources that are human-derived and controllable through improved management programs and enforcement mechanisms that will benefit ecosystem and public health.

- Continue emphasis on improving MST methodologies, including efforts that will encourage accessibility and use of these tools in a streamlined and cost-effective manner.

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APPENDIX A

Pictures of sampling sites.

Site 1

Site 2

Site 3

Site 4

Site 5

Site 6

Site 7

Site 8

Site 9

Site 10

Site 11

Site 12


Site 13


Site 14


Site 15


Site 16


Site 17


Site 18

Site 19

Site 20

Site 21

APPENDIX B

MST marker concentrations (log_{10} copies/100 mL) for sampling sites.

Site 1

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.84	0.52	0.00	0.00
2/8/2018	1.43	0.00	0.00	0.00	0.00	0.00	1.86	0.00	0.00	0.00
2/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.95	2.31	0.00	0.00
3/8/2018	0.93	0.00	0.00	0.00	0.00	0.00	1.54	1.35	0.00	0.00
3/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.96	0.00
4/5/2018	4.53	0.00	0.00	0.00	1.51	2.24	4.62	3.50	0.00	3.33
4/19/2018	1.16	0.00	0.00	0.00	0.00	0.00	1.91	2.63	0.00	0.00
5/3/2018	0.82	0.00	0.00	0.00	0.00	0.00	2.25	2.56	0.00	0.00
5/17/2018	0.00	0.00	0.00	0.00	1.51	0.00	0.00	1.73	2.00	0.00
5/31/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.49	2.20	2.21	0.00
6/14/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	2.07	2.24	0.00
6/28/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.94	0.00	2.08	0.00
9/21/2018	1.41	0.00	0.00	0.00	0.00	0.00	2.39	2.23	1.77	1.43
10/5/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.76	2.21	2.10	0.00
10/19/2018	0.78	0.00	0.00	0.00	0.00	0.00	2.19	1.57	2.35	0.00
11/2/2018	1.43	0.00	0.00	0.00	0.00	0.00	1.64	2.20	2.31	0.00
11/16/2018	0.00	0.00	1.58	0.00	0.00	0.00	1.62	1.37	1.38	0.00
12/14/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.16	1.27	2.05	0.00
12/21/2018	2.01	0.00	0.00	0.00	0.00	0.00	1.92	1.90	1.91	0.00
1/11/2019	1.76	0.00	0.00	0.00	0.00	0.00	1.63	1.32	1.80	0.00
1/25/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.07	1.11	2.17	0.00
2/8/2019	0.00	0.00	0.00	0.00	1.86	0.00	1.63	1.82	1.97	0.00
2/22/2019	1.84	0.00	0.00	0.00	0.00	0.00	2.19	1.53	1.87	0.71
3/8/2019	1.58	0.00	0.00	0.00	0.00	0.00	2.18	2.25	1.46	0.00
3/22/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.83	1.83	1.27	0.00
4/5/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.41	1.56	0.00	0.39
4/22/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.00	2.49	1.84	0.00
5/3/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.32	2.04	1.64	0.00
5/21/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.00	2.08	2.23	0.00
5/31/2019	1.98	0.00	0.00	0.00	0.00	0.00	2.46	2.15	1.04	0.93
6/14/2019	0.00	0.00	0.00	0.00	2.00	0.00	1.43	2.07	0.00	0.00

6/14/2019	1.26	0.00	0.00	0.00	1.80	0.00	2.11	1.76	1.28	1.00
6/28/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.59	1.88	1.44	0.00
7/12/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.32	0.00	1.54	0.00
7/26/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.12	2.53	0.00	0.10
8/9/2019	0.00	0.00	0.00	1.70	0.00	0.00	2.30	1.17	1.48	0.93
8/23/2019	2.11	0.00	0.00	2.86	0.00	0.00	2.11	1.36	0.97	0.33
8/23/2019	5.25	0.00	0.00	1.80	0.00	0.00	1.63	3.34	0.00	2.34
9/6/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.41	2.38	0.00	0.00
9/20/2019	0.00	0.00	0.00	2.15	0.00	0.00	1.35	2.68	0.00	0.00
10/4/2019	0.00	0.00	0.00	1.44	0.00	0.00	1.24	2.62	1.95	0.00
10/18/2019	1.85	0.00	0.00	0.00	0.93	0.00	1.33	0.00	2.18	2.73
11/1/2019	0.00	0.00	0.00	0.74	0.00	1.10	1.34	0.00	2.08	1.67
11/15/2019	0.00	0.00	0.00	1.12	0.00	1.13	1.71	2.36	1.65	0.00
11/27/2019	0.00	0.00	0.00	0.00	1.18	0.00	1.09	0.00	0.00	2.63
12/13/2019	1.10	0.00	0.00	1.76	0.00	0.00	1.84	0.00	1.78	0.00
1/3/2020	1.08	0.00	0.00	1.42	0.00	0.00	0.00	2.59	0.00	2.75
1/17/2020	0.00	0.00	0.00	0.00	0.86	0.00	1.96	2.05	0.00	0.00

Site 2

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.75	0.00	0.00	0.00
2/8/2018	0.83	0.00	0.00	0.00	0.00	0.00	1.61	1.82	0.00	0.00
2/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.56	2.60	0.00	0.00
3/8/2018	1.14	0.00	0.00	0.00	0.00	0.00	1.97	2.17	0.00	0.00
3/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
4/5/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.36	2.16	0.00	0.00
4/19/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.58	2.42	0.00	0.00
5/3/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.48	1.93	0.00	0.00
5/17/2018	1.59	0.00	0.00	0.00	0.00	0.00	2.66	2.08	1.93	0.89
5/31/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.00	2.81	2.11	0.00
6/14/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.53	2.30	2.12	0.00
6/28/2018	0.80	0.00	0.00	0.00	0.00	0.00	2.11	2.27	0.00	0.00
7/26/18	0.00	0.00	0.00	0.00	1.34	0.00	2.53	2.16	2.27	0.00
8/9/18	1.97	0.00	0.00	0.00	0.00	0.00	2.31	2.13	1.90	0.00
8/24/2018	2.50	0.00	0.00	0.00	0.00	0.00	2.06	2.42	2.14	0.00

9/7/2018	2.30	0.00	0.00	0.00	0.00	0.00	2.28	2.24	2.45	0.00
9/21/2018	0.93	0.00	0.00	0.00	1.91	0.00	2.06	1.41	2.11	0.00
10/5/2018	1.59	0.00	0.00	0.00	0.00	0.00	1.79	1.20	1.59	0.00
10/19/2018	1.70	0.00	0.00	0.00	0.00	0.00	1.98	1.14	2.33	0.00
11/2/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.95	2.22	2.23	0.00
11/16/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.31	1.43	1.93	0.00
12/14/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.73	1.77	0.00
12/21/2018	2.01	0.00	0.00	0.00	1.81	0.00	1.78	2.61	2.23	0.00
1/11/2019	2.51	0.00	0.00	0.00	2.02	0.00	1.88	1.22	1.82	0.00
1/25/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.71	1.46	1.93	0.00
2/8/2019	0.00	0.00	0.00	0.00	2.01	0.00	1.82	1.42	1.57	0.00
2/22/2019	1.63	0.00	0.00	0.00	1.58	0.00	2.46	1.96	1.58	0.00
3/8/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.24	2.58	1.76	1.32
3/22/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.91	1.25	1.62	0.00
4/5/2019	0.00	0.00	0.00	0.00	2.00	0.00	2.32	2.10	2.03	0.00
4/22/2019	1.72	0.00	0.00	0.00	1.94	0.00	2.69	0.00	1.94	0.00
5/3/2019	2.15	0.00	0.00	0.00	0.00	0.00	2.34	1.79	1.77	0.00
5/21/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.14	1.94	2.12	0.79
5/31/2019	1.39	0.00	0.00	0.00	2.22	0.00	1.96	0.00	1.69	0.00
6/14/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.82	1.78	0.94	0.00
6/28/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.26	0.00
7/12/2019	0.00	0.00	0.00	0.00	1.86	0.00	2.60	2.05	1.34	0.00
7/26/2019	0.00	1.82	0.00	1.43	0.00	0.00	2.00	1.93	1.72	0.00
8/9/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.51	1.28	0.00	0.00
8/23/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.30	0.00	0.00	0.00
9/6/2019	0.00	0.00	0.00	4.08	0.00	0.00	2.23	1.34	1.56	0.00
9/20/2019	5.32	0.00	0.00	1.91	2.35	0.00	3.91	5.71	0.00	1.16
10/4/2019	0.00	0.00	0.00	1.56	0.00	0.00	1.27	2.38	0.00	0.70
10/18/2019	0.00	0.00	1.92	0.00	0.00	0.00	0.84	2.25	0.00	0.39
11/1/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.41	0.00	2.55	3.19
11/15/2019	1.46	0.00	0.00	0.00	0.00	0.00	1.69	2.53	1.73	0.00
11/27/2019	0.00	1.88	0.00	1.66	0.00	0.00	1.33	2.49	2.31	0.00
12/13/2019	0.00	0.62	0.00	1.57	0.00	0.00	0.89	2.31	2.08	0.00
1/3/2020	0.00	0.00	0.00	0.00	0.00	0.00	1.42	2.42	0.00	0.00
1/17/2020	0.00	0.00	0.00	1.16	0.00	0.00	1.36	2.06	1.39	0.00
1/31/2020	1.21	0.00	0.00	1.19	1.56	1.40	1.62	2.22	0.00	0.00
2/14/2020	0.00	0.00	0.00	0.00	0.00	0.00	1.76	2.29	0.00	0.00
2/28/2020	1.80	0.00	0.00	0.00	0.00	0.00	1.86	2.17	1.68	0.00

Site 3

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	4.15	0.90	1.98	0.00	0.00	3.03	3.92	2.79	0.00	2.87
2/8/2018	5.22	0.00	0.00	2.72	1.40	3.94	4.05	1.86	0.00	3.22
2/22/2018	4.48	1.45	0.00	0.66	0.60	2.95	3.57	2.32	0.00	2.86
3/8/2018	4.53	0.00	0.00	0.00	0.00	1.96	4.02	2.36	0.00	2.71
3/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.55	0.00	0.00	0.00
4/5/2018	3.20	0.00	0.00	0.00	0.00	0.00	3.78	0.00	0.00	2.55
4/19/2018	4.89	0.00	0.00	2.33	0.00	2.51	4.95	3.03	0.00	2.75
5/3/2018	5.47	0.00	0.00	3.95	2.65	3.59	5.00	3.82	0.00	3.56
5/17/2018	4.67	0.00	0.00	2.64	1.59	2.88	3.76	2.74	2.01	2.54
5/31/2018	5.25	0.00	0.00	4.08	2.17	4.48	5.25	3.47	2.25	3.07
6/14/2018	5.19	0.00	0.00	4.10	2.45	3.94	5.24	4.52	2.04	2.96
6/28/2018	3.80	1.63	1.12	2.24	1.46	2.33	3.82	1.98	2.31	2.58
7/12/2018	4.74	0.00	0.00	2.12	0.00	1.92	4.19	2.80	2.09	2.03
7/26/18	5.26	0.00	0.00	0.95	2.01	1.70	4.60	3.95	2.42	2.36
8/9/18	4.70	0.00	0.00	0.00	1.22	0.00	4.21	2.32	2.03	2.33
8/24/2018	5.41	0.00	0.00	1.65	3.40	2.62	4.83	3.02	1.83	2.15
9/7/2018	5.06	0.00	0.00	1.68	2.79	3.48	5.61	5.04	1.32	3.44
9/21/2018	4.76	0.00	0.00	0.00	2.08	2.06	4.13	2.29	2.29	2.66
10/5/2018	5.67	0.00	0.00	2.63	2.97	3.21	5.06	2.83	2.01	3.43
10/19/2018	6.86	0.00	0.00	4.24	4.28	5.62	5.21	4.67	2.27	4.46
11/2/2018	5.41	0.47	0.00	1.97	3.29	3.73	5.38	4.14	2.24	2.74
11/16/2018	5.93	0.00	0.00	2.53	0.00	2.90	3.76	2.54	1.85	1.45
12/14/2018	4.21	0.00	0.00	0.00	2.16	0.00	3.84	2.36	2.09	1.24
12/21/2018	3.91	0.50	0.00	0.00	0.00	0.00	3.55	1.69	1.81	1.01
1/11/2019	4.15	0.00	0.30	0.00	1.23	0.00	3.95	1.89	1.68	1.31
1/25/2019	4.86	0.00	0.00	1.27	0.00	2.96	4.19	1.77	1.88	0.75
2/8/2019	4.47	0.00	0.00	0.00	2.13	2.88	4.32	1.26	1.72	2.70
2/22/2019	5.18	0.00	0.00	1.33	2.03	3.89	4.07	2.82	1.97	2.92
3/8/2019	4.93	0.00	0.00	1.02	2.33	2.82	4.24	1.72	2.01	2.74
3/22/2019	4.81	0.00	0.00	2.24	2.04	2.86	4.90	4.22	1.73	2.54
4/5/2019	5.16	0.00	0.00	1.36	0.00	0.00	4.64	0.00	2.14	1.78
4/19/2019	4.47	0.00	0.00	0.00	0.00	0.00	4.23	2.83	1.97	2.32
5/3/2019	5.34	0.00	0.00	0.00	0.00	0.00	4.57	3.62	2.10	1.30
5/21/2019	5.54	0.00	0.00	1.71	2.17	2.51	4.80	3.27	1.67	2.16

5/31/2019	5.93	0.00	0.00	2.81	2.45	3.11	5.03	4.02	1.01	2.37
6/14/2019	5.29	0.00	0.00	3.12	3.16	2.98	4.75	4.13	1.42	2.70
6/28/2019	5.02	0.00	0.00	3.59	4.15	3.98	4.48	3.79	0.00	2.55
7/12/2019	4.59	0.00	0.00	1.65	2.62	2.19	3.37	2.94	1.24	1.89
7/26/2019	4.91	0.00	0.00	2.28	0.00	1.48	4.49	2.42	1.75	1.46
8/9/2019	0.00	0.00	0.00	0.00	0.00	0.00	3.10	1.59	0.00	0.00
8/23/2019	5.34	0.00	0.00	1.53	1.72	0.00	5.02	3.13	0.00	1.81
9/6/2019	5.31	0.00	0.00	2.22	1.62	0.00	4.84	1.63	1.62	0.00
9/20/2019	5.51	1.85	0.00	2.21	0.00	0.00	2.75	3.05	0.00	2.45
10/4/2019	6.10	0.00	0.00	0.00	0.00	0.00	3.24	3.67	0.00	1.66
10/18/2019	5.92	0.00	0.00	1.72	0.00	0.00	3.07	3.74	0.00	1.81
11/1/2019	4.14	0.00	0.00	2.00	1.20	0.00	2.57	3.08	2.16	3.22
11/15/2019	5.09	0.00	3.12	2.07	0.00	0.00	2.93	3.83	2.12	2.83
11/27/2019	1.73	0.00	0.00	0.00	0.00	0.00	1.61	0.00	2.38	0.00
12/13/2019	2.28	0.00	1.06	0.00	0.93	0.00	1.55	2.74	2.69	0.00
1/3/2020	5.03	0.00	0.00	2.05	0.00	0.00	3.76	3.16	0.00	2.65
1/17/2020	5.10	0.41	0.00	1.22	0.00	0.00	3.54	4.13	0.00	4.20
1/31/2020	4.70	0.00	0.00	0.00	0.00	0.00	2.03	2.78	0.00	3.06
2/14/2020	4.61	0.00	0.00	2.69	0.00	1.53	3.59	2.77	1.51	2.31
2/28/2020	5.25	0.00	0.00	0.00	0.00	2.35	4.08	2.59	0.00	2.33

Site 4

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	4.60	0.00	0.00	3.70	1.95	3.36	3.95	2.48	0.00	3.53
2/8/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
2/22/2018	5.67	0.00	0.00	4.63	3.02	4.62	5.16	4.37	0.00	4.36
3/8/2018	0.00	0.00	0.00	0.00	0.00	0.83	0.00	0.00	0.00	1.87
3/22/2018	5.51	0.00	0.00	3.59	2.13	3.54	5.09	4.44	0.00	3.38
4/5/2018	5.56	0.00	0.00	4.22	2.50	4.17	5.19	4.60	0.00	3.74
4/19/2018	4.32	0.00	0.00	0.00	0.00	0.00	4.35	3.01	0.00	2.95
5/3/2018	4.75	0.00	0.00	1.42	0.00	2.15	4.78	2.77	2.10	2.24
5/17/2018	4.68	0.00	0.00	0.00	0.00	0.00	4.90	2.55	1.73	2.82
5/31/2018	4.68	0.00	0.00	0.00	0.00	0.00	4.35	0.00	1.92	2.60
6/14/2018	4.55	0.00	0.00	0.00	1.81	0.00	4.28	2.22	1.85	2.34
6/28/2018	4.82	0.00	0.00	0.00	2.16	0.00	4.56	2.58	2.10	2.18

7/12/2018	5.22	0.00	0.00	0.00	0.00	0.00	4.74	3.31	2.17	2.38
7/26/18	5.08	0.00	0.00	2.45	2.23	2.65	4.31	3.29	2.24	2.71
8/9/18	4.97	0.00	0.00	2.25	0.00	2.83	4.60	3.86	2.21	3.06
8/24/2018	5.27	0.00	0.00	2.56	2.72	2.60	4.92	4.42	2.14	2.87
9/7/2018	5.32	0.00	0.00	1.87	2.39	2.42	4.79	3.19	2.16	2.67
9/21/2018	5.02	0.67	BDL	0.81	1.97	0.00	4.64	1.91	2.39	3.26
10/5/2018	6.06	0.00	0.00	3.80	3.79	4.27	5.24	4.55	2.15	3.58
10/19/2018	5.57	0.44	BDL	1.87	2.25	1.65	5.30	4.16	1.87	2.81
11/2/2018	4.74	0.00	0.00	0.00	1.79	1.55	4.09	2.72	2.02	2.81
11/16/2018	4.40	0.00	0.00	0.94	2.17	2.14	4.09	1.93	2.09	2.36
12/14/2018	5.23	0.00	0.00	1.15	0.00	1.54	4.47	2.08	1.85	2.01
12/21/2018	4.51	0.00	0.00	2.29	2.33	2.62	4.46	2.29	2.03	2.27
1/11/2019	4.89	0.00	0.00	2.85	2.48	3.16	4.31	2.14	1.51	1.79
1/25/2019	4.63	0.00	0.00	2.27	2.54	2.87	4.78	3.21	1.21	3.07
2/8/2019	3.73	0.00	0.00	1.65	2.18	2.57	3.75	2.00	1.75	2.47
2/22/2019	4.85	0.00	0.00	3.28	3.04	3.58	4.85	3.10	1.86	2.86
3/8/2019	3.78	0.00	0.00	0.93	0.74	2.01	4.28	2.40	2.11	2.53
3/22/2019	4.39	0.00	0.00	0.65	0.00	1.81	5.09	2.83	1.63	1.76
4/19/2019	5.29	0.92	0.00	1.10	2.48	2.21	5.12	4.04	1.09	2.31
5/3/2019	4.30	0.00	0.00	0.00	0.00	0.00	3.68	2.45	1.89	1.87
5/21/2019	5.13	0.00	0.00	1.83	0.00	1.77	5.03	3.52	0.00	2.18
5/31/2019	5.12	0.00	0.00	2.46	0.00	1.90	5.32	3.43	0.79	2.14
6/14/2019	4.06	0.00	0.00	0.00	1.99	0.00	4.56	3.08	1.56	1.56
6/28/2019	4.96	0.00	0.00	0.00	0.00	0.00	5.16	3.08	1.55	2.41
7/12/2019	4.92	0.00	0.00	0.00	0.00	0.00	5.04	3.24	1.59	1.73
7/26/2019	4.90	0.00	0.00	3.21	1.58	3.34	4.85	3.13	0.00	2.33
8/9/2019	4.58	0.00	0.00	2.98	2.19	2.54	5.09	4.34	1.98	2.59
8/23/2019	2.60	0.00	0.00	2.26	0.00	0.00	1.36	2.54	0.00	0.41
9/6/2019	5.86	0.00	0.00	3.13	2.28	2.48	2.56	3.54	0.00	2.05
9/20/2019	0.00	0.00	0.00	0.00	0.00	0.00	2.10	3.57	0.00	0.00
10/4/2019	5.52	0.00	0.00	2.99	0.00	2.61	2.64	4.08	0.00	2.41
10/18/2019	4.87	0.00	0.00	2.05	2.04	1.99	3.63	4.19	2.31	3.49
11/1/2019	4.92	0.00	1.35	3.10	2.15	3.17	2.75	3.99	2.10	3.66
11/15/2019	0.00	0.00	0.00	1.58	0.87	0.00	1.37	2.50	2.26	0.00
11/27/2019	1.38	0.00	0.00	0.00	0.00	0.00	1.37	2.60	2.35	2.09
12/13/2019	5.03	0.00	0.00	2.17	0.00	0.00	2.49	3.01	1.65	3.52
1/3/2020	5.07	1.42	0.00	1.80	0.00	0.00	3.60	4.91	0.00	3.37
1/17/2020	4.63	0.59	0.00	2.90	2.07	2.95	2.16	3.54	0.00	3.81
1/31/2020	4.91	1.05	0.00	2.01	1.80	2.06	3.92	2.89	0.00	2.32
2/14/2020	4.57	0.00	0.00	1.80	0.89	0.00	3.91	2.92	0.00	2.15

2/28/2020	4.60	0.00	0.00	3.70	1.95	3.36	3.95	2.48	0.00	3.53

Site 5

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	5.44	0.00	0.62	3.50	3.13	4.08	4.01	3.17	1.67	3.31
2/8/2018	2.80	0.00	0.00	0.00	0.00	0.67	2.02	0.82	0.00	0.00
2/22/2018	5.12	0.00	0.00	3.13	2.83	3.94	4.24	3.64	0.00	3.68
3/8/2018	5.18	0.00	0.00	3.45	3.12	3.97	4.65	3.37	0.00	3.52
3/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.85	2.22	0.00	0.00
4/5/2018	5.18	0.00	0.00	2.57	2.53	2.16	5.04	3.93	0.00	2.71
4/19/2018	5.46	0.00	0.00	2.81	2.82	2.83	5.57	4.68	0.00	2.96
5/3/2018	4.97	0.00	0.00	2.86	2.52	3.09	5.33	3.77	2.05	3.42
5/17/2018	4.51	0.00	0.00	2.93	2.70	3.30	4.45	3.00	1.90	2.55
5/31/2018	4.05	0.00	0.00	2.64	2.42	2.89	4.06	2.97	1.80	2.50
6/14/2018	4.33	0.00	0.00	2.10	2.11	2.82	4.04	2.12	2.15	2.62
6/28/2018	5.65	0.00	0.00	3.78	3.66	4.35	5.49	4.25	2.07	3.45
7/12/2018	4.77	0.00	0.00	2.24	2.01	3.01	4.47	3.21	1.70	2.71
7/26/18	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.94
8/9/18	5.15	0.00	0.00	2.79	3.02	3.25	3.82	3.10	1.81	2.08
8/24/2018	5.67	0.00	0.00	3.94	4.72	4.37	4.88	3.04	1.94	2.85
9/7/2018	5.82	0.00	0.00	1.87	2.03	1.91	5.49	3.77	2.16	1.62
9/21/2018	5.32	0.00	0.00	1.14	0.00	0.00	4.83	2.90	2.35	2.58
10/5/2018	4.81	0.00	0.00	2.19	2.17	2.64	4.19	3.11	2.59	2.75
10/19/2018	5.32	0.54	BDL	2.51	3.31	3.06	4.96	3.36	2.38	2.49
11/2/2018	5.71	1.25	0.00	2.84	4.09	3.53	5.65	4.49	2.36	2.77
11/16/2018	7.67	2.94	2.05	4.48	5.76	5.53	6.09	4.48	1.98	3.98
12/14/2018	5.57	0.00	0.00	2.03	3.85	3.76	5.46	2.94	1.73	2.81
12/21/2018	4.93	0.00	0.00	2.33	3.25	3.48	4.48	1.91	2.07	2.14
1/11/2019	4.69	0.00	0.00	1.91	2.73	3.02	4.38	1.72	1.90	2.11
1/25/2019	5.96	0.00	0.00	3.78	4.08	4.76	5.09	2.79	1.38	0.00
2/8/2019	4.86	0.00	1.61	1.44	2.26	3.02	4.44	3.01	1.54	2.75
2/22/2019	4.43	0.00	0.00	3.59	1.87	0.00	4.29	2.15	1.77	2.46
3/8/2019	2.91	0.00	0.00	0.00	0.00	1.70	2.76	2.27	1.80	1.03
3/22/2019	5.07	0.00	0.00	2.31	3.01	3.07	4.54	2.22	1.47	2.73
4/5/2019	4.71	0.00	0.00	1.99	3.74	2.99	4.50	2.40	1.67	1.83

4/19/2019	4.31	0.00	0.00	0.65	0.00	0.00	4.10	2.77	1.44	2.15
5/3/2019	6.01	0.00	0.00	2.64	3.04	2.90	5.05	3.90	2.06	2.57
5/21/2019	4.42	0.00	1.75	1.23	0.00	1.73	3.99	2.56	2.14	1.73
6/14/2019	5.32	2.00	0.00	3.51	3.67	3.10	4.78	3.36	1.09	2.51
6/28/2019	5.17	0.00	0.00	1.85	0.00	0.00	6.09	3.70	1.13	1.89
7/12/2019	4.68	0.00	0.00	0.00	2.68	0.00	5.55	3.77	1.03	2.23
7/26/2019	4.95	0.00	0.00	3.06	0.00	0.00	4.56	2.36	0.00	2.38
8/9/2019	5.26	0.00	0.00	2.58	2.90	2.05	4.17	2.49	1.59	2.17
8/23/2019	5.72	0.00	0.00	1.89	1.74	1.65	5.96	3.52	0.00	1.60
9/6/2019	4.68	0.00	0.00	1.62	0.00	0.00	2.33	2.43	0.00	1.89
9/20/2019	5.77	0.00	0.00	2.82	2.16	2.38	2.64	3.21	0.00	2.26
10/4/2019	5.49	0.00	0.00	1.74	0.00	0.00	2.40	3.01	0.00	2.47
10/18/2019	7.20	0.00	0.00	4.10	4.90	4.48	2.98	4.71	0.00	2.96
11/1/2019	5.43	0.00	0.00	3.94	3.23	3.34	3.12	4.27	2.13	3.67
11/15/2019	4.85	0.00	0.00	3.97	2.58	2.71	3.18	3.62	3.41	3.42
11/27/2019	0.00	0.00	0.00	0.00	0.84	1.64	0.00	0.00	2.13	0.00
12/13/2019	1.78	0.00	0.00	1.18	0.00	1.27	1.05	2.24	2.60	0.00
1/3/2020	5.22	0.00	0.00	3.49	2.95	3.64	3.02	3.92	0.00	3.68
1/17/2020	5.79	0.00	0.00	3.53	3.04	4.06	2.37	4.05	0.00	3.99
1/31/2020	5.36	0.00	0.00	3.03	2.66	3.76	1.72	3.21	1.98	3.87
2/14/2020	5.42	0.00	0.00	3.74	2.61	3.44	3.53	3.02	0.00	2.20
2/28/2020	5.57	0.00	0.00	3.97	3.40	3.60	4.00	2.63	0.00	2.65

Site 6

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicke n Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	2.96	0.00	0.00	2.18	0.00	0.00	3.33	0.59	0.00	2.96
2/8/2018	2.69	0.00	0.00	0.00	0.00	0.62	3.25	2.70	0.00	2.55
2/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.08	2.68	0.00	2.55
3/8/2018	3.16	0.00	0.00	0.00	0.00	0.00	4.02	2.50	0.00	2.96
3/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
4/5/2018	3.90	0.00	2.64	0.00	0.00	0.00	4.69	2.81	0.00	2.75
4/19/2018	3.82	0.00	0.00	1.35	1.54	0.00	4.88	2.65	0.00	2.97
5/3/2018	4.40	3.88	0.00	2.22	0.00	0.00	5.01	2.75	1.63	3.25
5/17/2018	4.09	0.00	0.00	0.00	2.00	0.00	4.31	2.23	2.06	2.13
5/31/2018	4.03	0.00	0.00	0.00	1.55	0.00	4.07	1.74	2.02	2.47

6/14/2018	3.88	0.00	0.00	0.00	0.00	0.00	4.02	2.61	2.06	2.06
6/28/2018	3.49	0.00	0.00	0.00	0.00	0.00	3.93	2.45	2.22	3.42
7/12/2018	4.5	0.00	0.00	1.46	0.00	0.00	4.67	2.77	2.01	2.64
7/26/18	4.40	0.00	0.00	0.00	0.00	0.00	3.92	1.91	2.07	2.08
8/9/18	4.85	0.00	0.00	0.00	0.00	0.00	4.08	2.40	2.45	2.87
8/24/2018	5.37	0.00	0.00	0.00	0.00	0.00	4.96	2.99	2.31	2.15
9/7/2018	4.84	0.00	0.00	1.11	2.00	0.00	5.07	3.23	2.19	1.84
9/21/2018	4.71	0.00	0.00	0.00	0.00	0.00	3.56	1.29	1.82	2.56
10/5/2018	4.48	0.00	0.00	BDL	0.00	0.00	4.46	2.02	2.26	2.86
10/19/2018	4.32	0.00	0.00	1.88	2.77	2.52	4.69	3.39	1.79	2.59
11/2/2018	4.18	0.00	0.00	0.00	0.00	0.00	3.76	1.88	2.01	2.55
11/16/2018	3.86	0.00	0.00	0.00	0.00	0.00	2.92	1.17	1.85	0.99
12/14/2018	3.56	0.00	0.00	1.04	0.00	0.00	3.70	2.37	1.93	1.35
12/21/2018	3.72	0.00	0.00	0.00	0.00	0.00	3.61	2.18	1.91	1.27
1/11/2019	3.68	0.00	0.00	0.00	0.00	0.00	3.89	2.73	1.49	0.00
1/25/2019	4.15	0.00	0.00	0.99	0.00	0.00	4.21	2.96	2.29	2.13
2/8/2019	3.22	0.00	0.00	0.00	1.85	0.00	3.65	1.97	1.90	2.37
2/22/2019	2.70	0.00	0.00	0.00	1.62	0.00	3.40	1.16	1.69	2.10
3/8/2019	1.89	0.00	0.00	0.00	2.16	0.00	2.45	1.73	1.71	0.86
3/22/2019	4.14	0.00	0.00	1.00	0.00	0.00	3.99	2.73	1.08	2.18
4/5/2019	4.89	0.00	0.00	0.19	0.00	0.00	4.62	0.00	1.79	1.79
4/19/2019	4.03	0.00	0.00	0.00	0.00	0.00	3.91	2.46	2.02	1.44
5/3/2019	4.60	0.00	0.00	0.00	0.00	1.82	4.99	4.17	2.04	2.13
5/21/2019	4.85	0.00	0.00	BDL	2.50	0.00	4.54	2.82	2.00	2.55
5/31/2019	5.74	0.00	0.00	0.00	2.36	0.00	4.76	2.97	1.66	3.05
6/14/2019	5.11	1.97	2.39	3.22	3.14	2.83	4.80	3.26	1.66	2.95
6/28/2019	4.69	0.00	0.00	1.78	1.84	0.00	3.97	2.49	0.00	3.19
7/12/2019	5.29	0.00	0.00	2.57	1.58	3.14	4.56	2.98	1.08	2.79
7/26/2019	5.27	3.31	3.35	3.49	0.00	0.00	4.53	3.11	1.55	3.80
8/9/2019	4.93	0.00	2.37	2.31	0.00	2.20	4.33	2.78	1.64	2.81
8/23/2019	5.31	0.00	0.00	3.46	2.04	3.37	4.81	2.80	0.00	2.25
9/6/2019	4.42	0.00	0.00	2.18	0.00	0.00	1.63	0.00	0.00	2.59
9/20/2019	5.51	0.00	0.00	2.32	2.38	0.00	2.67	2.98	0.00	2.41
10/4/2019	5.98	0.00	0.00	2.43	2.36	0.00	3.04	2.15	0.00	2.21
10/18/2019	5.82	2.40	2.18	2.07	2.33	0.00	3.03	4.70	0.00	1.57
11/1/2019	4.89	0.00	0.00	1.72	0.00	0.00	3.01	3.67	2.12	3.52
11/15/2019	4.12	0.00	0.00	1.88	0.00	0.00	3.12	4.33	2.40	4.32
11/27/2019	1.57	0.00	0.00	1.33	0.00	0.00	1.32	2.32	2.36	0.00
12/13/2019	0.00	0.00	0.00	1.38	0.00	0.00	1.58	2.53	2.35	0.00
1/3/2020	0.00	0.00	0.00	0.00	0.00	0.00	1.22	1.98	0.00	0.00

1/17/2020	5.16	0.00	0.00	2.33	0.00	1.63	5.14	4.79	0.00	3.81
1/31/2020	4.88	0.25	0.00	1.76	0.00	0.00	2.87	3.17	0.00	3.59
2/14/2020	4.52	0.00	0.00	2.23	1.99	1.48	4.20	3.72	0.00	2.36
2/28/2020	3.88	0.00	0.00	0.00	0.00	1.21	3.32	2.32	0.00	1.58

Site 7

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	2.70	0.00	0.00	1.58	0.00	0.00	3.54	2.00	0.00	2.42
2/8/2018	1.09	0.00	0.00	0.00	0.00	0.00	1.91	1.10	0.00	0.00
2/22/2018	2.75	0.00	0.00	1.04	0.00	1.86	3.96	3.08	0.00	2.25
3/8/2018	4.07	0.00	0.95	1.20	0.00	0.00	4.38	2.45	0.00	2.52
3/22/2018	3.88	0.00	0.00	0.00	0.00	0.00	4.30	2.64	0.00	2.82
4/5/2018	4.65	0.00	0.00	1.58	0.00	0.00	4.36	3.08	0.00	3.23
4/19/2018	4.50	0.00	0.00	1.33	0.00	0.00	4.42	2.58	0.00	3.10
5/3/2018	4.50	0.00	0.00	0.00	0.00	0.00	4.83	2.98	0.00	3.34
5/17/2018	5.07	0.00	0.00	1.32	1.77	0.00	5.20	2.31	2.25	2.67
5/31/2018	4.73	1.15	0.00	1.46	0.94	0.00	5.09	2.53	2.33	3.37
6/14/2018	4.27	0.00	0.00	0.00	1.57	0.00	4.90	2.11	2.05	3.36
6/28/2018	3.53	0.00	0.00	0.00	0.00	0.00	4.04	1.92	1.57	2.98
7/12/2018	5.01	0.00	0.00	1.90	0.00	0.00	4.70	3.18	2.05	3.02
7/26/18	4.18	0.00	0.00	0.00	1.88	0.00	3.60	2.64	1.74	2.76
8/9/18	4.48	0.00	0.00	0.00	0.00	0.00	4.56	2.91	1.90	2.91
8/24/2018	5.05	0.00	0.00	0.00	1.71	0.00	4.95	3.40	2.15	2.56
9/7/2018	4.97	0.00	0.00	0.45	2.58	0.00	4.95	3.30	1.97	2.41
9/21/2018	5.36	0.86	0.72	0.94	0.00	0.00	4.12	3.04	2.12	3.14
10/5/2018	5.08	1.44	1.34	2.24	0.00	0.00	4.99	2.53	2.11	3.37
10/19/2018	5.20	BDL	0.00	2.52	4.01	0.00	5.38	4.19	2.23	3.36
11/2/2018	5.73	0.70	0.64	3.31	4.39	2.09	6.21	5.37	2.87	2.56
11/16/2018	4.87	0.09	0.00	1.75	0.00	0.00	4.19	2.59	1.42	2.04
12/14/2018	3.98	0.00	0.00	0.63	2.30	0.00	4.12	2.95	1.91	2.25
12/21/2018	4.06	0.00	0.00	0.00	0.00	0.00	3.91	1.87	1.94	1.51
1/11/2019	4.51	0.45	0.00	1.92	1.91	0.00	4.49	2.39	1.82	2.02
1/25/2019	4.40	0.00	0.00	1.44	0.00	0.00	4.00	2.35	1.78	0.72
2/8/2019	4.33	0.00	0.00	0.41	0.00	0.00	3.82	2.03	1.53	3.42
2/22/2019	4.12	0.00	0.00	0.00	0.00	0.00	3.98	2.25	1.51	2.87

3/8/2019	4.75	0.00	0.00	0.00	0.00	0.00	4.82	1.80	1.58	2.69
3/22/2019	5.02	0.00	0.00	0.30	0.00	0.00	4.80	2.36	2.14	4.17
4/5/2019	5.22	0.00	0.00	0.34	0.00	0.00	4.50	0.00	1.41	3.33
4/19/2019	4.49	0.00	1.73	BDL	0.00	0.00	4.12	2.77	1.82	1.43
5/3/2019	4.82	2.02	1.73	1.14	3.34	0.00	4.77	3.76	2.00	2.56
5/21/2019	4.98	0.00	0.00	0.58	0.00	0.00	4.49	1.80	1.62	1.68
5/31/2019	4.70	0.00	0.00	0.00	1.88	0.00	4.62	3.21	0.76	2.21
6/14/2019	4.76	0.00	0.00	0.00	1.68	0.00	4.08	3.04	1.34	0.81
6/28/2019	4.84	0.00	0.00	3.18	3.13	0.00	3.94	4.43	2.42	2.09
7/12/2019	5.26	0.00	0.00	2.52	0.00	0.00	4.98	3.38	1.20	1.95
7/26/2019	5.02	1.72	0.00	0.00	0.00	0.00	4.42	2.94	1.55	2.14
8/9/2019	5.14	0.00	0.00	2.77	1.65	2.70	4.64	2.74	1.64	1.80
8/23/2019	3.94	0.00	0.00	0.87	0.00	0.00	3.83	1.91	1.53	2.05
9/6/2019	2.76	0.00	0.00	2.32	0.00	1.16	1.07	2.39	0.00	0.79
9/20/2019	5.22	0.00	0.00	2.07	0.00	0.00	1.36	2.43	0.00	2.43
10/4/2019	5.68	0.00	0.00	0.00	0.00	0.00	2.43	2.30	0.00	2.48
10/18/2019	3.67	0.00	0.00	0.00	3.25	0.00	1.67	3.21	0.00	1.40
11/1/2019	4.89	1.27	0.00	2.47	1.10	0.00	2.83	5.30	1.55	3.74
11/15/2019	4.91	0.00	2.84	2.60	2.65	0.00	3.80	5.11	2.09	4.00
11/27/2019	1.92	0.92	0.00	0.00	0.00	1.30	1.25	0.00	2.30	2.45
12/13/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.87	0.00	2.13	1.98
1/3/2020	4.32	0.00	0.00	1.70	0.00	0.00	3.00	3.38	1.76	3.33
1/17/2020	4.92	0.95	0.00	2.48	2.98	0.00	3.50	4.56	0.00	3.78
1/31/2020	4.40	0.00	0.00	1.89	0.00	0.00	2.80	3.69	0.00	3.48
2/14/2020	2.60	0.00	0.00	2.19	0.00	0.00	2.53	2.50	0.00	0.37
2/28/2020	4.85	1.39	1.74	2.53	0.00	0.00	3.70	2.83	1.80	2.18

Site 8

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/23/2018	3.89	2.32	1.20	2.24	1.77	1.82	4.07	3.57	0.00	2.80
2/8/2018	3.83	1.92	0.00	1.13	1.52	0.00	3.59	1.62	0.00	2.36
2/22/2018	5.39	3.26	2.00	2.61	2.46	0.00	5.23	3.90	1.69	2.81
3/8/2018	5.98	3.68	2.93	2.46	0.00	1.92	4.71	3.06	0.00	2.71
3/22/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
4/5/2018	5.66	4.02	3.85	1.80	0.00	0.00	5.30	3.02	0.00	3.23

4/19/2018	5.95	3.84	0.00	2.44	0.00	0.00	4.91	3.16	0.00	2.82
5/3/2018	5.36	3.67	3.40	1.70	0.00	0.00	4.77	2.89	0.00	2.85
5/17/2018	4.96	0.16	0.00	0.00	0.00	0.00	4.04	2.95	1.71	2.62
5/31/2018	3.88	0.00	0.00	0.00	0.00	0.00	3.68	2.51	2.12	2.32
6/14/2018	4.92	0.00	0.00	0.00	2.08	1.72	5.09	2.95	2.04	2.29
6/28/2018	3.59	0.00	0.00	0.00	0.00	0.00	3.70	3.17	2.03	2.11
7/12/2018	5.01	0.00	0.00	1.55	0.00	1.90	4.89	3.10	2.03	2.62
7/26/18	4.77	0.00	0.00	0.00	0.00	0.00	3.88	3.40	1.77	1.80
8/9/18	4.85	0.00	0.00	0.84	0.00	0.00	4.08	3.11	2.11	1.72
8/24/2018	5.30	0.00	0.00	1.25	0.00	0.00	4.55	2.96	1.91	2.10
9/7/2018	4.99	0.15	0.00	BDL	2.14	0.00	5.13	4.16	2.23	2.39
9/21/2018	5.52	0.00	0.00	0.00	0.00	0.00	4.77	2.98	2.34	2.64
10/5/2018	5.41	0.00	0.00	0.00	1.87	0.00	5.22	3.24	2.35	3.09
10/19/2018	5.68	1.69	1.04	2.77	4.49	1.46	6.11	4.52	2.64	3.02
11/2/2018	6.28	2.04	2.19	2.83	4.84	1.60	6.19	5.06	2.22	2.39
11/16/2018	5.25	0.00	0.21	1.22	2.62	1.96	4.71	2.69	1.70	1.33
12/14/2018	4.46	0.00	0.00	1.40	2.14	0.00	4.09	2.14	2.31	1.57
12/21/2018	5.05	0.00	0.00	1.95	2.74	2.14	4.29	2.09	0.00	1.41
1/11/2019	4.81	0.00	0.00	2.19	1.70	2.49	4.59	2.32	2.10	1.52
1/25/2019	5.02	0.00	0.00	1.77	0.00	1.00	4.44	2.79	1.52	0.00
2/8/2019	5.35	0.00	0.00	1.31	2.39	1.65	4.92	2.91	1.91	2.46
2/22/2019	3.89	0.00	1.56	0.00	2.27	0.00	3.90	2.80	1.14	2.41
3/8/2019	3.26	0.00	0.00	0.00	0.00	0.00	3.57	2.73	1.78	2.18
3/22/2019	5.23	1.99	1.44	1.61	2.23	2.23	5.15	3.73	1.83	2.44
4/5/2019	4.94	0.00	0.00	0.73	0.00	0.00	4.50	2.98	2.17	1.60
4/19/2019	5.05	0.00	0.00	1.03	2.48	0.00	4.63	2.96	1.99	1.65
5/3/2019	3.28	0.00	0.00	0.00	2.92	0.00	2.82	2.25	0.00	0.00
5/21/2019	6.84	3.30	2.94	2.16	3.79	2.37	6.24	5.58	2.25	2.56
5/31/2019	5.97	2.25	0.00	1.69	3.15	2.04	5.44	5.12	0.00	1.52
6/14/2019	1.30	0.00	0.00	0.00	0.00	0.00	4.74	3.26	1.63	0.00
6/28/2019	5.88	0.00	0.00	1.61	0.00	0.00	5.26	4.06	1.56	1.15
7/12/2019	5.77	2.60	2.19	1.68	1.95	0.00	5.08	3.42	0.00	1.69
7/26/2019	6.01	1.80	0.00	1.88	0.00	0.00	5.01	3.03	0.00	1.05
8/9/2019	5.09	0.00	0.00	0.00	0.00	0.00	4.59	2.93	1.14	1.60
8/23/2019	6.04	0.00	0.00	2.00	0.00	0.59	6.86	6.73	1.99	1.79
9/6/2019	5.88	2.08	0.00	0.00	0.00	0.00	2.68	3.66	0.00	2.39
9/20/2019	5.99	0.00	0.00	0.00	2.32	0.00	4.46	6.38	0.00	0.00
10/4/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
10/18/2019	6.05	0.00	0.00	0.00	0.00	0.44	3.69	5.12	0.00	2.07
11/1/2019	4.89	0.00	2.15	0.90	0.00	0.00	3.57	4.00	2.11	3.35

11/15/2019	5.66	0.00	2.73	2.98	3.01	1.02	4.37	5.34	2.78	3.40
11/27/2019	0.00	0.88	0.00	0.00	1.07	0.00	1.43	2.36	2.41	0.00
12/13/2019	1.65	0.00	0.00	0.00	0.00	0.00	1.55	0.00	0.00	0.00
1/3/2020	4.98	0.00	0.00	2.08	0.00	0.00	2.98	3.51	0.00	3.28
1/17/2020	6.21	2.70	2.40	3.16	3.83	1.10	4.75	5.93	0.00	3.08
1/31/2020	0.00	0.00	0.00	0.00	0.00	0.00	1.02	0.00	0.00	0.00
2/14/2020	5.63	2.65	2.44	3.00	3.31	0.00	4.57	4.08	1.79	1.40
2/28/2020	5.07	0.91	0.00	2.25	0.99	0.00	3.72	2.77	0.00	1.76

Site 9

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	3.91	0.00	0.00	0.00	0.00	1.76	4.63	2.03	0.00	2.83
2/15/2018	5.37	0.00	0.00	4.06	1.95	4.30	4.55	2.85	0.00	3.64
3/1/2018	4.31	2.98	1.04	0.87	0.00	1.66	4.32	2.48	0.00	3.13
3/15/2018	4.27	1.64	0.00	2.18	0.00	1.55	5.01	3.87	0.00	3.04
3/29/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
4/12/2018	4.52	3.45	3.66	1.09	0.00	0.00	4.50	2.07	0.00	2.91
4/30/2018	4.81	0.00	0.00	1.25	0.00	0.00	5.44	3.06	0.00	3.01
5/10/2018	5.04	0.00	0.00	0.00	0.00	0.00	4.88	2.92	0.00	3.04
5/24/2018	4.19	0.00	0.00	0.00	0.00	0.00	4.57	2.90	1.86	3.00
6/7/2018	4.77	0.00	0.00	0.00	0.00	0.00	5.00	2.62	2.35	2.86
6/21/2018	5.02	0.00	0.00	3.85	0.92	2.99	5.16	3.66	2.14	2.87
7/13/2018	4.11	0.00	0.00	0.00	1.79	0.00	4.74	2.60	1.95	2.65
7/19/2018	4.78	0.00	0.07	0.00	0.00	0.00	3.81	2.34	1.62	2.24
8/2/18	4.43	0.00	0.00	0.00	0.00	1.14	4.19	2.64	2.10	2.17
8/16/2018	5.14	0.00	0.00	0.00	0.00	0.00	4.78	3.47	2.07	1.84
8/31/2018	4.60	0.00	0.00	0.00	0.00	0.00	4.92	2.96	1.90	2.04
9/14/2018	4.40	0.00	0.00	1.05	1.87	0.00	3.84	1.64	2.05	1.96
9/29/2018	5.29	0.78	0.00	2.26	0.00	0.00	4.63	2.68	2.39	2.50
10/12/2018	5.11	2.17	1.60	1.80	2.75	0.00	5.08	3.80	2.38	2.79
10/26/2018	4.63	1.89	1.44	1.99	2.19	2.30	4.24	3.02	1.98	2.47
11/9/2018	5.60	1.47	1.77	3.09	4.68	1.98	5.67	5.07	2.51	2.33
11/30/2018	3.45	0.00	0.00	0.00	1.86	0.00	3.03	1.40	1.87	1.14
12/16/2018	2.95	0.00	0.00	0.00	0.00	0.00	3.46	1.66	2.07	1.10
1/4/2019	4.72	0.00	0.00	2.16	3.30	0.00	4.72	3.27	2.04	1.63

1/18/2019	4.27	0.00	0.00	0.00	0.00	0.00	4.10	2.04	1.83	0.00
2/1/2019	4.06	0.00	0.00	1.19	1.99	0.00	4.29	1.97	2.06	2.46
2/15/2019	3.58	0.00	0.00	0.00	0.00	0.00	4.22	1.55	1.28	2.27
3/1/2019	4.16	0.00	0.00	0.00	1.98	0.00	3.70	2.26	1.49	2.49
3/18/2019	4.55	0.00	0.00	0.57	0.00	0.00	4.34	1.62	1.89	2.52
3/29/2019	4.43	0.00	0.00	0.00	1.50	0.00	3.64	3.18	1.81	2.39
4/12/2019	3.11	0.00	0.00	0.00	0.00	0.00	3.21	1.95	1.67	0.50
4/26/2019	5.07	0.00	1.43	0.00	2.36	0.00	5.15	3.18	2.03	1.39
5/10/2019	5.32	2.60	2.33	3.80	3.88	1.51	5.38	4.25	3.51	2.22
5/24/2019	4.85	0.00	0.00	0.09	0.00	0.00	4.78	2.95	1.99	2.00
6/7/2019	4.66	0.00	0.00	2.72	0.00	0.00	4.48	3.12	1.67	2.29
6/21/2019	5.37	0.00	2.67	3.28	1.76	0.00	5.59	3.62	1.92	2.22
7/8/2019	4.93	0.00	0.00	1.83	0.00	0.00	5.08	3.43	1.79	2.77
7/19/2019	3.91	0.00	0.00	1.74	0.00	0.00	4.88	3.33	0.00	1.65
8/2/2019	4.65	0.00	0.00	2.09	0.00	0.00	3.99	1.84	2.15	1.69
8/16/2019	1.90	0.00	0.00	0.00	0.00	0.00	5.02	2.83	1.81	2.11
8/30/2019	0.00	1.81	0.00	3.45	0.00	0.00	4.24	2.18	2.32	2.39
9/13/2019	5.53	0.00	0.00	2.44	0.00	0.00	2.18	2.36	0.00	2.54
9/27/2019	6.66	0.00	0.00	0.00	2.11	0.00	2.04	3.47	0.00	3.01
10/11/2019	3.94	0.00	0.00	0.00	0.00	0.00	0.92	2.39	0.00	2.01
10/25/2019	5.59	2.40	2.11	3.98	3.82	0.00	3.59	5.07	3.19	2.34
11/8/2019	6.13	0.00	0.00	2.08	1.27	0.00	2.93	3.84	2.38	3.61
11/22/2019	4.78	0.00	0.00	2.60	0.00	0.00	2.86	3.32	2.08	3.32
12/6/2019	1.56	0.00	0.00	0.00	1.13	0.00	1.37	2.28	2.30	0.00
12/20/2019	0.00	0.00	0.00	1.47	0.00	0.00	1.49	2.34	0.00	0.00
1/10/2020	3.98	0.00	0.00	1.42	0.00	0.00	2.47	2.23	0.00	3.27
1/24/2020	5.40	0.00	0.00	2.05	2.86	0.00	3.65	4.28	0.00	3.11
2/7/2020	3.49	0.00	0.00	1.80	0.00	0.00	1.87	0.00	0.00	3.55
2/21/2020	4.58	0.00	0.00	0.00	0.00	0.00	3.59	2.55	0.00	1.90
3/6/2020	4.57	0.00	0.00	0.00	0.91	0.00	3.25	2.39	1.53	2.33

Site 10

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	2.43	0.00	0.00	0.00	0.00	0.00	2.81	0.00	0.00	1.75
2/15/2018	1.98	0.00	0.00	0.00	0.00	0.00	2.80	0.78	0.00	1.74
3/1/2018	3.32	0.00	0.00	0.00	0.00	0.00	4.19	2.41	0.00	2.46
3/15/2018	3.43	0.00	0.00	0.00	0.00	0.00	4.08	2.20	0.00	2.50

3/29/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.39	0.00	0.00	1.58
4/12/2018	3.37	0.00	0.00	0.00	0.00	0.00	4.10	1.91	0.00	2.87
4/30/2018	3.48	0.00	0.00	0.00	0.00	0.00	3.51	2.68	0.00	2.37
5/10/2018	2.10	0.00	0.00	0.00	0.00	0.00	3.04	2.46	0.00	2.55
6/7/2018	2.99	0.00	0.00	0.00	1.88	0.00	2.46	1.79	2.23	1.30
6/21/2018	2.41	0.00	0.00	0.00	0.00	0.00	3.41	0.00	1.77	2.08
7/13/2018	2.46	0.00	0.00	0.00	1.65	0.00	2.94	2.47	1.95	1.97
7/19/2018	3.31	0.00	0.00	0.00	0.00	0.00	3.32	2.49	1.90	1.33
8/2/18	2.67	0.00	0.00	0.00	1.71	0.00	2.63	1.97	2.03	0.98
8/16/2018	2.75	0.00	0.00	0.00	0.00	0.00	2.98	2.31	2.12	1.31
8/31/2018	1.03	0.00	0.00	0.00	0.00	0.00	2.26	2.59	2.31	1.16
9/14/2018	1.75	0.00	0.00	0.00	0.00	0.00	2.73	1.92	1.95	2.43
9/29/2018	2.85	0.00	0.00	0.00	0.00	0.00	3.30	2.01	2.08	3.12
10/12/2018	2.85	0.00	0.00	0.00	0.00	0.00	3.12	1.94	2.05	3.07
10/26/2018	3.53	0.00	0.00	0.00	0.00	0.00	3.54	1.62	2.08	2.38
11/9/2018	3.01	BDL	0.00	0.00	0.00	0.00	3.17	1.75	1.91	0.82
11/30/2018	2.95	0.00	0.00	0.00	0.00	1.59	2.70	1.66	2.01	0.00
12/16/2018	2.25	0.00	0.00	0.00	1.85	0.00	2.36	0.00	1.79	0.00
1/4/2019	2.84	0.00	0.00	0.00	0.00	0.00	2.85	0.83	1.99	0.00
1/18/2019	3.29	0.85	0.06	0.00	1.94	0.00	3.12	1.23	1.72	0.00
2/1/2019	3.14	0.00	0.00	0.00	0.00	0.00	3.04	0.73	1.90	0.00
2/15/2019	2.37	0.00	0.00	0.00	1.91	0.00	2.89	1.33	1.82	0.66
3/1/2019	2.58	0.00	0.00	0.00	1.58	0.00	2.87	2.05	1.50	1.12
3/18/2019	2.11	0.00	0.00	0.00	0.00	0.00	2.52	1.98	0.00	1.16
3/29/2019	2.30	0.00	0.00	0.00	0.00	0.00	2.74	1.63	1.66	1.37
4/12/2019	4.80	0.00	0.00	BDL	1.52	0.00	4.77	2.85	1.93	1.42
4/26/2019	3.87	0.00	0.00	0.00	0.00	0.00	3.99	2.64	1.62	1.03
5/10/2019	3.75	0.00	0.00	1.36	2.79	0.00	3.99	2.52	2.19	1.10
5/24/2019	3.29	0.00	0.00	0.00	0.00	0.00	3.74	1.90	1.63	1.28
6/7/2019	4.17	0.00	0.00	2.13	2.41	0.00	5.08	4.15	1.64	2.10
6/21/2019	2.35	0.00	0.00	0.00	0.00	0.00	2.79	0.00	0.99	0.00
7/8/2019	3.43	0.00	0.00	0.00	1.94	0.00	3.71	2.29	1.54	1.56
7/19/2019	3.26	0.00	0.00	1.62	0.00	0.00	3.45	1.71	0.00	1.59
8/2/2019	3.66	0.00	0.00	0.00	0.00	0.00	2.90	1.25	1.49	1.82
8/16/2019	3.02	0.00	0.00	0.00	0.00	0.00	3.00	0.00	0.00	0.21
10/11/2019	3.31	0.00	0.00	1.99	0.00	0.00	1.20	2.03	0.00	1.51
10/25/2019	3.55	0.00	0.00	2.18	0.00	0.00	1.15	3.09	0.00	1.69
11/8/2019	2.33	0.00	0.00	0.00	0.00	0.00	1.36	0.00	2.06	2.34
11/22/2019	2.57	0.42	0.00	1.54	0.00	0.00	1.39	2.02	2.05	2.77
12/6/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.85	2.61	2.33	0.00

12/20/2019	3.90	0.00	0.00	2.19	0.00	0.00	2.83	2.50	1.79	4.76
1/10/2020	2.79	0.00	0.00	1.37	0.00	1.50	1.34	2.32	0.00	3.08
1/24/2020	2.80	0.00	0.00	1.43	0.00	0.00	1.46	4.27	0.00	2.67
2/7/2020	2.99	0.00	0.00	2.01	0.00	0.00	1.43	2.14	0.00	1.72
2/21/2020	3.61	0.00	0.00	0.00	0.00	0.00	3.02	2.31	0.00	1.07
3/6/2020	3.41	0.00	0.00	0.00	0.00	0.00	2.94	2.24	0.00	1.11

Site 11

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	3.21	0.00	0.00	2.07	1.56	1.96	3.55	1.98	0.00	2.14
2/15/2018	2.64	0.00	0.00	0.00	0.00	0.00	3.04	1.85	0.00	1.70
3/1/2018	3.10	0.00	0.00	0.00	0.00	0.00	3.37	2.82	0.00	2.04
3/15/2018	2.98	0.00	0.00	0.00	0.00	0.00	3.06	2.42	0.00	1.79
3/29/2018	4.51	3.86	3.42	2.82	3.36	1.94	5.05	4.10	2.96	3.36
4/12/2018	4.03	0.00	0.00	0.00	0.00	0.00	4.32	2.54	0.00	2.13
4/30/2018	4.54	0.00	0.00	0.00	0.00	0.00	4.35	2.65	0.00	2.79
5/10/2018	5.29	0.00	3.20	0.00	0.00	0.00	3.46	0.00	0.00	2.89
5/24/2018	2.99	0.00	0.00	0.00	0.00	0.00	2.77	1.87	2.08	1.33
6/7/2018	3.65	0.00	0.00	0.00	0.00	0.00	3.33	2.29	2.39	3.10
6/21/2018	4.16	0.00	0.00	0.00	0.00	0.00	4.15	3.39	1.88	2.51
7/13/2018	3.65	0.00	0.00	0.00	0.00	0.00	3.44	2.36	1.55	2.28
8/2/18	4.31	0.00	0.00	BDL	0.00	0.00	3.95	3.65	1.23	2.89
8/16/2018	4.32	0.00	0.00	0.00	0.00	0.00	3.50	2.49	2.40	2.41
8/31/2018	4.84	0.00	0.00	0.00	0.00	0.00	4.23	3.54	1.88	3.11
9/14/2018	4.42	0.00	0.00	0.54	0.00	0.00	4.03	0.96	2.38	2.54
9/29/2018	5.04	0.00	BDL	1.24	0.00	0.00	4.16	2.39	2.28	2.86
10/12/2018	4.54	0.00	0.00	2.19	0.00	0.00	4.11	2.39	2.46	3.08
10/26/2018	3.98	0.00	0.00	0.77	0.00	0.00	3.87	2.07	2.02	3.01
11/9/2018	4.45	0.00	0.00	1.73	1.23	1.63	4.49	3.55	1.77	3.02
11/30/2018	3.60	0.00	0.00	0.00	0.00	0.00	2.86	1.59	1.80	0.77
12/16/2018	2.75	0.00	0.00	0.00	0.00	0.00	2.58	1.51	1.86	1.72
1/4/2019	3.58	0.00	0.00	1.36	2.05	0.00	3.13	2.06	1.49	2.22
1/18/2019	4.15	0.00	0.00	0.96	0.00	0.00	3.48	1.75	1.98	1.91
2/1/2019	4.10	0.00	0.00	2.40	0.00	0.00	2.97	2.14	2.02	1.26
2/15/2019	3.24	0.00	0.00	0.00	2.30	0.00	2.81	2.22	1.81	1.62

3/1/2019	3.55	0.00	0.00	1.51	0.00	0.00	3.19	2.07	2.02	1.98
3/18/2019	3.81	1.97	0.00	0.00	0.00	0.00	3.37	2.27	1.45	1.82
3/29/2019	4.38	0.00	0.00	0.59	0.00	0.00	3.99	1.88	1.60	2.32
4/12/2019	4.60	0.00	0.00	1.03	0.00	0.00	4.52	2.75	2.01	2.71
4/26/2019	4.46	0.00	0.00	1.26	0.00	0.00	4.54	3.12	2.28	0.33
5/10/2019	4.49	1.72	0.00	2.14	0.00	0.00	4.45	3.10	2.25	2.12
5/24/2019	4.58	0.00	0.00	2.88	0.00	0.00	4.22	3.07	3.24	1.84
6/7/2019	2.40	0.00	0.00	0.00	0.00	0.00	2.68	1.78	1.03	0.65
6/21/2019	3.93	0.00	0.00	0.00	0.00	0.00	3.39	2.64	1.46	1.62
7/8/2019	3.91	0.00	0.00	2.12	0.00	0.00	3.77	2.61	1.29	1.84
7/19/2019	4.72	0.00	1.68	3.43	0.00	0.00	4.02	2.40	2.98	2.44
8/2/2019	4.79	0.00	0.00	2.25	1.47	0.45	4.23	3.17	0.00	2.74
8/16/2019	5.02	0.00	0.00	0.00	0.00	0.00	4.10	1.30	1.63	2.36
8/30/2019	4.72	0.00	0.00	2.70	0.00	0.00	4.61	2.76	1.58	2.16
9/13/2019	5.48	1.32	0.00	1.97	0.00	0.00	3.27	3.80	0.00	2.80
9/27/2019	5.57	2.07	0.00	2.07	0.00	0.00	3.30	2.09	0.00	2.46
10/11/2019	5.49	0.00	0.00	0.99	0.00	0.00	3.40	3.55	0.00	3.06
10/25/2019	5.41	1.76	0.00	2.35	0.00	0.00	2.55	2.66	0.00	2.58
11/8/2019	3.50	0.00	1.36	0.00	0.00	0.00	3.03	2.88	2.28	3.14
11/22/2019	4.11	1.42	0.00	1.72	0.00	0.00	3.73	3.46	2.11	3.86
12/6/2019	0.00	0.59	0.00	1.42	0.00	0.00	1.27	2.52	2.34	0.00
12/20/2019	1.28	0.00	0.00	0.87	0.00	0.00	1.65	2.80	0.00	0.00
1/10/2020	4.22	0.00	0.00	0.00	0.00	1.53	3.27	2.49	0.00	3.93
1/24/2020	4.42	0.00	0.00	1.49	0.00	0.00	3.10	3.22	0.00	3.05
2/7/2020	4.18	0.00	0.00	1.47	0.00	0.00	2.87	3.62	0.00	3.52
2/21/2020	4.77	0.00	0.00	2.84	1.40	0.00	3.34	2.37	0.00	2.69
3/6/2020	4.91	0.00	0.00	1.88	0.00	0.00	3.51	2.56	0.00	2.90

Site 12

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	2.61	0.00	0.00	0.00	0.00	1.05	2.34	2.69	0.00	2.57
2/15/2018	3.61	0.00	0.00	0.85	0.00	1.38	3.39	3.09	0.00	3.56
3/1/2018	4.43	2.71	0.00	1.04	0.00	2.53	4.37	2.95	0.00	3.35
3/15/2018	3.49	1.51	0.00	0.00	0.00	0.95	3.52	3.05	0.00	3.17
3/29/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.21	0.00	0.00	0.00

4/12/2018	3.65	0.00	0.00	0.00	1.85	2.06	3.65	2.66	0.00	3.49
4/30/2018	3.68	0.00	0.00	0.00	0.81	0.00	3.78	2.30	0.00	3.44
5/10/2018	3.35	3.67	0.00	0.00	0.00	0.00	3.42	2.40	0.00	3.21
5/24/2018	3.29	0.00	0.00	0.00	1.76	0.00	3.38	2.15	2.10	2.60
6/7/2018	4.01	0.00	0.00	0.00	1.62	0.00	4.38	2.19	2.18	3.08
6/21/2018	3.43	0.00	0.00	0.00	0.00	0.00	3.69	2.28	1.99	2.74
8/2/18	4.13	0.00	0.00	0.00	0.00	0.00	3.90	3.34	1.84	2.53
8/16/2018	4.67	0.00	0.00	1.31	1.34	0.00	4.01	3.19	1.82	2.67
8/31/2018	3.06	0.00	0.00	0.00	0.00	0.00	2.64	2.33	2.01	1.85
9/14/2018	4.65	0.00	0.00	1.65	2.39	0.00	3.96	2.54	2.13	2.63
9/29/2018	5.08	0.00	0.00	2.12	2.29	0.00	4.21	2.73	2.52	2.82
10/12/2018	4.64	0.00	0.00	2.05	0.00	0.00	4.07	3.02	2.17	2.89
10/26/2018	3.93	0.00	0.00	1.72	0.00	1.33	3.60	2.54	0.00	2.25
11/9/2018	4.48	0.00	0.00	2.48	2.57	0.00	4.46	3.29	1.91	2.67
11/30/2018	3.77	0.00	0.00	0.00	0.00	0.00	2.86	2.00	1.99	0.91
12/16/2018	3.96	0.00	0.00	1.50	1.85	0.00	3.31	2.35	2.15	2.29
1/4/2019	4.37	0.00	0.00	2.24	2.14	1.89	4.08	2.67	1.86	2.49
1/18/2019	4.55	0.00	0.00	1.83	1.79	1.72	4.05	2.08	1.76	2.42
2/1/2019	5.00	0.00	0.00	2.63	1.85	1.77	3.88	2.86	2.36	1.80
2/15/2019	3.26	0.00	0.00	0.00	0.00	0.00	2.97	2.31	1.44	1.52
3/1/2019	2.95	0.00	0.00	0.00	2.02	0.00	2.93	1.90	1.29	1.21
3/18/2019	3.38	0.00	0.00	0.77	0.00	0.00	3.15	2.40	1.09	0.37
3/29/2019	3.81	0.00	0.00	0.69	1.47	1.63	3.64	1.97	1.83	2.47
4/12/2019	4.17	0.00	0.00	0.00	0.00	0.00	3.96	1.78	1.35	1.48
4/26/2019	4.41	0.00	0.00	1.01	1.94	0.00	4.22	2.78	1.90	2.44
5/10/2019	4.81	0.00	0.00	2.87	1.93	1.63	4.71	3.41	2.36	2.56
5/24/2019	4.81	1.65	0.00	2.37	2.10	0.00	4.41	3.12	2.01	2.12
6/7/2019	4.49	1.54	0.00	2.82	0.00	0.00	4.05	2.86	2.08	1.01
6/21/2019	4.43	0.00	0.00	0.00	0.00	0.00	4.00	3.23	1.49	2.83
7/8/2019	5.02	0.00	1.87	3.76	0.00	0.00	5.06	3.39	2.48	1.87
7/19/2019	4.93	2.29	2.22	2.40	0.00	1.42	4.50	2.81	2.01	2.69
8/2/2019	4.15	0.00	0.00	2.57	0.00	0.00	3.35	2.31	1.92	1.95
8/16/2019	5.30	0.00	0.00	3.55	2.33	0.80	4.39	3.97	1.71	2.63
8/30/2019	5.24	0.00	0.00	2.72	0.00	0.00	4.41	2.88	1.57	2.84
9/13/2019	5.49	0.00	0.00	2.42	3.28	0.00	2.55	3.33	0.00	3.23
9/27/2019	5.67	0.00	0.00	2.30	0.00	0.00	2.74	4.00	0.00	3.55
10/11/2019	5.78	0.00	0.00	1.68	0.00	0.00	2.64	3.85	0.00	3.69
10/25/2019	4.90	0.00	0.00	1.62	3.64	0.00	2.16	4.16	0.00	2.82
11/8/2019	4.05	0.00	1.41	1.49	0.00	1.47	2.29	3.65	2.12	3.75
11/22/2019	4.72	0.00	2.41	1.60	1.76	1.06	2.67	4.49	2.33	4.40

12/6/2019	0.00	0.00	0.00	1.66	0.94	1.13	1.40	1.94	2.05	0.00
12/20/2019	0.00	0.00	0.00	1.38	0.00	0.00	1.16	2.53	0.00	0.00
1/10/2020	2.94	0.00	0.00	0.00	0.00	0.00	1.76	3.16	0.00	3.67
1/24/2020	4.39	0.00	0.00	0.95	0.00	0.00	2.69	4.48	0.00	3.92
2/7/2020	3.81	0.00	0.00	1.60	0.00	0.00	2.03	3.76	0.00	3.36
2/21/2020	4.73	2.02	2.45	2.09	0.00	0.00	3.02	3.33	0.00	2.73
3/6/2020	5.09	1.82	2.39	2.74	0.00	1.64	3.19	3.61	1.68	2.87

Site 13

Date of sample collection	**Bac Uni**	**Bac Hum**	**HF183**	**Bac Cow**	**Bac Can**	**Chicke n Duck Bac**	**Entero 1**	***E. coli***	**Rum2 Bac**	**GFD**
1/30/2018	3.32	0.90	0.96	0.00	0.00	0.00	3.45	2.68	0.00	3.58
2/15/2018	2.90	0.00	0.00	0.87	0.00	0.00	2.64	2.16	0.00	3.22
3/1/2018	3.98	1.16	0.00	0.79	2.84	0.00	3.85	2.89	0.00	3.51
3/15/2018	3.91	0.00	1.08	2.54	0.97	0.55	3.82	2.93	0.00	3.83
3/29/2018	2.43	0.00	0.00	0.00	0.00	0.00	2.88	2.48	0.00	0.00
4/12/2018	3.97	0.00	0.00	0.00	0.00	0.00	4.02	2.67	0.00	3.62
4/30/2018	4.20	3.69	3.44	1.49	0.00	0.00	4.39	3.11	0.00	4.11
5/10/2018	4.45	3.68	3.28	2.46	0.00	0.00	4.41	3.05	0.00	4.56
5/24/2018	5.27	0.00	0.00	2.38	1.90	2.26	6.16	3.96	1.96	4.00
6/7/2018	4.46	0.00	0.00	0.00	1.38	1.38	4.57	2.63	1.93	3.91
6/21/2018	4.18	0.00	0.00	0.00	0.00	0.00	4.26	1.71	2.34	3.75
7/13/2018	4.17	0.00	0.00	0.00	0.00	0.00	4.19	2.72	2.04	2.50
7/19/2018	4.72	1.08	0.00	1.33	1.15	0.00	4.45	4.27	2.19	2.44
8/2/18	4.62	0.00	0.00	0.00	0.00	0.00	3.96	3.02	1.92	3.94
8/16/2018	4.84	0.00	0.00	0.00	0.00	0.00	4.18	3.00	2.25	3.53
8/31/2018	4.55	0.00	0.00	0.00	0.00	0.00	4.09	3.02	2.18	4.29
9/14/2018	4.25	0.00	0.00	0.90	1.45	0.00	3.57	2.07	2.03	2.09
9/29/2018	4.49	0.93	0.77	1.23	0.00	0.00	3.77	2.24	2.13	2.77
10/12/2018	4.46	BDL	BDL	BDL	0.00	0.00	3.49	3.02	2.14	2.34
10/26/2018	4.82	BDL	0.10	2.04	2.87	0.00	4.70	3.03	2.12	3.06
11/9/2018	5.84	0.00	0.00	2.82	4.33	3.91	3.86	2.76	2.09	2.88
11/30/2018	5.02	0.00	0.00	1.22	2.20	1.90	4.22	2.77	1.69	2.07
12/16/2018	4.06	0.00	0.00	BDL	0.00	0.00	3.63	2.00	2.02	2.34
1/4/2019	4.11	0.00	0.00	1.12	0.00	0.00	3.84	2.38	1.84	2.07
1/18/2019	4.56	0.88	0.00	0.12	0.00	0.00	4.08	2.34	2.06	2.24

2/1/2019	4.15	0.00	0.00	BDL	2.22	0.00	3.35	1.85	2.14	1.73
2/15/2019	2.36	0.00	0.00	0.00	0.00	0.00	2.50	1.84	1.74	0.60
3/1/2019	3.91	0.00	0.00	0.00	0.00	0.00	3.72	2.39	2.01	2.78
3/18/2019	4.46	0.00	0.00	0.00	1.91	0.00	4.03	2.17	1.75	3.45
3/29/2019	4.23	0.00	0.00	0.57	1.84	0.00	4.05	2.45	1.76	2.64
4/12/2019	3.74	0.00	0.00	0.00	0.00	0.00	4.41	1.93	1.71	1.78
4/22/2019	4.71	0.81	0.00	1.51	0.00	0.00	4.47	2.92	1.95	2.65
4/26/2019	4.86	1.66	0.00	0.83	0.00	0.00	4.40	2.63	0.96	3.17
5/10/2019	4.70	1.14	0.00	2.48	1.63	0.00	4.50	3.13	2.28	2.34
5/24/2019	4.67	2.07	1.90	0.63	0.00	0.00	4.46	2.45	1.98	2.58
6/7/2019	4.81	1.83	0.00	2.35	1.91	0.00	4.66	3.11	1.25	3.17
6/21/2019	5.20	2.31	0.00	2.33	0.00	0.00	4.79	3.15	1.15	2.97
7/8/2019	4.12	1.98	2.40	0.00	2.41	0.00	3.62	2.94	1.67	1.76
7/19/2019	5.27	1.25	1.67	2.88	1.74	0.00	4.30	2.74	1.92	2.90
8/2/2019	5.00	1.71	0.00	3.46	0.00	0.00	4.36	2.94	2.60	3.30
8/16/2019	4.75	1.74	0.00	0.00	0.00	0.00	4.14	2.62	0.00	2.83
8/30/2019	5.28	0.00	2.35	1.99	0.00	0.00	4.61	3.53	0.00	3.33
9/13/2019	5.88	0.00	0.00	2.04	0.00	0.00	2.53	3.95	0.00	3.23
9/27/2019	4.61	2.02	0.00	2.32	0.00	0.00	1.28	0.00	0.00	2.91
10/11/2019	5.66	2.34	2.15	1.70	0.00	0.00	2.69	4.00	0.00	4.18
10/25/2019	5.01	0.00	1.85	2.42	1.85	0.00	2.76	4.62	2.24	4.18
11/8/2019	4.46	0.00	1.55	1.65	0.00	1.17	2.76	3.66	2.12	3.73
11/22/2019	4.78	0.00	0.00	2.68	0.00	0.00	2.91	4.12	2.29	4.32
12/6/2019	0.00	0.26	0.00	1.53	0.00	1.42	1.36	2.07	2.19	0.00
12/20/2019	1.74	0.00	0.00	0.00	0.00	0.00	1.43	0.00	0.00	0.00
1/10/2020	4.59	0.88	2.42	2.10	1.86	2.00	2.22	3.46	1.66	3.94
1/24/2020	5.16	1.87	2.56	3.09	1.95	2.99	2.79	4.75	0.00	4.08
2/7/2020	4.08	0.61	1.79	2.06	0.00	1.69	1.82	4.16	0.00	3.63
2/21/2020	5.19	1.68	0.00	2.26	0.85	1.95	3.51	2.70	0.00	3.02
3/6/2020	4.47	0.00	0.00	1.88	0.00	0.00	2.95	2.57	0.00	2.77

Site 14

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	3.94	0.00	0.00	0.00	1.42	1.85	3.81	2.65	0.00	3.47
2/15/2018	3.31	0.00	0.00	0.73	1.24	1.93	4.01	2.14	0.00	3.26

3/1/2018	4.47	1.44	1.09	1.36	0.00	0.00	4.04	3.37	0.00	3.46
3/15/2018	4.65	0.00	0.00	2.03	0.00	0.00	5.00	3.91	0.00	3.20
3/29/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.01	2.16	0.00	0.00
4/12/2018	3.75	3.23	0.00	0.00	0.00	0.00	4.11	2.54	0.00	3.40
4/30/2018	4.25	0.00	0.00	0.48	0.00	0.00	4.63	3.13	0.00	4.13
5/10/2018	4.60	0.00	0.00	0.00	0.00	0.00	5.09	2.61	0.00	4.41
5/24/2018	4.00	0.00	0.00	1.44	0.78	0.00	4.36	2.11	2.09	3.44
6/7/2018	3.68	0.00	0.00	0.00	0.00	0.00	4.01	2.42	1.86	3.02
6/21/2018	3.81	0.00	0.00	0.00	1.80	0.00	3.67	2.08	1.92	2.97
7/13/2018	4.60	0.00	0.00	0.00	0.00	0.00	4.50	2.66	1.73	2.80
7/19/2018	5.12	0.00	0.00	0.00	2.02	0.00	4.38	2.41	1.52	2.92
8/2/18	3.92	0.00	0.00	0.00	0.00	0.00	3.71	2.83	2.11	2.03
8/16/2018	4.29	0.00	0.00	0.00	0.00	0.00	4.03	2.44	2.09	2.61
8/31/2018	3.11	0.00	0.00	0.00	0.00	0.00	2.86	2.35	2.06	2.03
9/14/2018	4.21	0.00	0.00	0.84	0.00	0.00	3.59	1.35	1.93	2.45
9/29/2018	4.45	0.48	BDL	2.36	2.10	0.00	3.36	1.73	2.82	2.37
10/12/2018	4.76	0.00	0.00	2.06	3.10	0.00	4.08	2.83	2.47	2.71
10/26/2018	4.31	0.00	0.00	1.78	0.00	0.00	3.89	2.39	2.38	2.76
11/9/2018	4.45	0.00	0.00	1.95	0.00	0.00	3.95	2.23	2.15	2.87
11/30/2018	3.42	0.00	0.00	1.30	0.00	2.28	2.96	1.60	0.95	2.62
12/16/2018	3.60	0.00	0.00	1.02	0.00	0.00	3.02	2.03	1.76	1.56
1/4/2019	5.34	0.00	BDL	3.88	2.71	1.84	5.28	3.77	3.55	2.52
1/18/2019	4.56	0.00	0.00	3.19	1.69	2.31	3.76	2.57	2.77	1.88
2/1/2019	4.46	0.00	0.00	2.83	0.00	2.65	3.30	2.33	2.61	1.79
2/15/2019	3.22	0.00	0.00	0.00	0.00	0.00	3.07	1.57	1.92	1.81
3/1/2019	2.97	0.00	0.00	0.00	1.70	0.00	3.08	2.18	2.08	2.18
3/18/2019	3.62	0.00	0.00	0.00	0.00	0.00	3.77	1.88	1.53	2.71
3/29/2019	3.96	0.00	0.00	0.00	1.79	0.00	4.37	1.82	1.78	2.93
4/12/2019	4.42	0.00	0.00	0.00	2.39	0.00	4.22	1.97	1.43	2.30
4/26/2019	4.51	0.00	0.00	0.00	0.00	0.00	4.12	2.30	1.89	2.15
5/10/2019	4.40	0.00	0.00	2.05	2.23	0.00	4.24	2.76	1.88	1.75
5/24/2019	4.59	0.00	0.00	0.74	0.00	0.00	4.51	2.12	1.46	2.72
6/7/2019	4.42	0.00	0.00	0.00	0.00	0.00	4.95	2.78	0.00	2.16
6/21/2019	4.44	0.00	0.00	0.00	0.00	0.00	4.82	2.54	1.18	2.09
7/8/2019	4.55	0.00	0.00	0.00	1.82	0.00	4.18	2.61	1.08	2.08
7/19/2019	5.13	0.00	0.00	0.00	0.00	0.00	4.74	1.78	1.47	2.37
8/2/2019	3.66	0.00	0.00	0.00	0.00	0.00	4.54	1.58	1.13	1.81
8/16/2019	4.56	0.00	0.00	1.78	0.00	0.00	5.19	2.19	1.81	2.14
8/30/2019	5.14	0.00	0.00	1.66	0.00	0.00	5.10	3.27	0.00	2.66
9/13/2019	5.63	0.00	0.00	1.86	0.00	0.00	3.25	3.85	0.00	2.51

9/27/2019	5.22	0.00	0.00	2.15	0.00	0.00	2.88	2.61	0.00	2.73
10/11/2019	5.78	0.00	0.00	0.00	0.00	0.00	3.59	3.03	0.00	3.57
10/25/2019	4.78	0.00	0.00	1.85	0.00	0.00	3.12	4.21	2.15	4.21
11/8/2019	3.55	0.00	1.29	0.00	0.00	0.00	2.27	3.04	1.67	3.57
11/22/2019	5.71	0.96	0.00	4.74	3.03	1.17	3.64	5.85	2.44	4.00
12/6/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.83	2.65	2.37	0.00
12/20/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.00	2.35	1.60	0.00
1/10/2020	4.95	0.00	0.00	2.21	1.82	0.00	3.94	3.77	0.00	4.33
1/24/2020	5.52	0.00	0.00	1.23	0.00	1.30	3.25	3.94	0.00	4.31
2/7/2020	5.19	0.00	0.00	1.91	0.00	0.00	2.75	2.30	0.00	3.78
2/21/2020	5.35	1.55	0.00	1.85	0.00	0.00	3.40	2.50	1.49	3.02
3/6/2020	4.85	0.00	0.00	0.00	0.00	0.00	3.37	2.62	0.00	2.68

Site 15

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	3.21	0.00	0.00	0.65	0.00	0.00	3.58	1.78	0.00	2.21
2/15/2018	4.65	0.00	0.00	0.98	0.00	0.00	4.59	2.07	0.00	2.99
3/1/2018	4.23	0.00	0.00	0.85	0.60	0.00	4.36	2.30	0.00	3.12
3/15/2018	4.21	0.00	0.00	0.48	0.00	0.00	4.08	2.60	0.00	3.93
3/29/2018	4.68	4.71	4.18	2.18	1.76	0.00	4.72	3.84	0.00	3.79
4/12/2018	4.94	3.66	0.00	0.61	0.00	1.09	4.39	2.21	0.00	3.03
4/30/2018	5.52	0.00	0.00	2.48	0.00	1.02	6.12	3.06	0.00	4.09
5/10/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.96	2.42	0.00	2.48
5/24/2018	4.86	0.00	0.00	0.00	1.55	0.00	4.58	2.93	2.31	3.50
6/7/2018	5.52	0.00	0.00	0.00	0.83	0.00	6.19	3.27	2.17	4.18
6/21/2018	4.08	0.00	0.00	0.00	0.00	0.00	5.31	2.43	2.17	2.83
7/13/2018	5.13	0.00	0.00	2.09	0.00	0.00	4.90	2.92	2.02	2.16
7/19/2018	5.30	0.00	0.22	1.36	1.31	0.00	4.54	3.00	1.91	2.48
8/2/18	4.57	0.00	0.00	0.00	0.00	0.00	3.75	2.95	2.07	2.00
8/16/2018	4.98	0.00	0.00	0.00	0.00	0.00	4.19	3.00	2.08	2.40
8/31/2018	5.07	0.00	0.00	0.05	1.90	0.00	4.92	2.89	2.09	3.57
9/14/2018	4.68	0.43	0.00	1.33	3.24	0.00	3.74	2.42	2.38	2.57
9/29/2018	4.90	0.00	0.00	2.33	0.00	0.00	4.46	2.83	2.68	2.91
10/12/2018	3.85	0.00	0.00	BDL	0.00	0.00	3.42	2.56	2.01	2.22
10/26/2018	4.03	0.00	0.00	1.44	0.00	0.00	3.84	2.28	1.98	2.67

11/9/2018	3.77	0.00	0.00	1.01	0.00	0.00	3.55	1.94	1.74	2.01
11/30/2018	3.54	0.00	0.00	0.00	0.00	0.00	3.13	1.84	0.00	1.68
12/16/2018	3.52	0.00	0.00	0.00	2.11	0.00	3.19	2.36	1.89	1.21
1/4/2019	5.03	0.00	0.24	3.45	1.98	2.16	5.02	3.44	3.27	2.69
1/18/2019	4.42	0.00	0.00	2.53	1.70	1.89	3.92	2.42	2.51	2.07
2/1/2019	4.11	0.00	0.00	1.39	0.00	1.45	3.80	2.04	2.30	1.89
2/15/2019	3.97	0.00	0.00	0.00	1.93	0.00	4.18	2.16	0.00	2.53
3/1/2019	2.92	0.00	0.00	0.00	1.58	0.00	3.20	2.08	1.85	1.95
3/18/2019	2.95	0.00	0.15	0.00	0.00	0.00	2.99	2.17	2.18	2.41
3/29/2019	4.00	1.49	0.00	0.00	0.00	0.00	4.29	2.61	2.11	2.79
4/12/2019	4.31	0.00	0.00	0.80	0.00	0.00	4.84	2.94	1.94	2.62
4/26/2019	4.52	1.70	0.00	0.59	0.00	0.00	4.51	3.30	1.73	2.13
5/10/2019	5.29	2.19	1.25	4.14	3.30	0.00	5.34	4.14	3.63	2.78
5/24/2019	3.83	0.00	0.00	0.00	0.00	0.00	3.39	2.09	1.67	0.00
6/7/2019	4.34	0.00	0.00	0.00	0.00	0.00	5.09	2.76	0.00	2.18
6/21/2019	4.51	2.00	0.00	2.18	1.75	0.00	4.87	3.09	1.24	2.34
7/8/2019	4.65	0.00	0.00	0.00	0.00	0.00	5.17	3.10	0.92	2.51
7/19/2019	4.54	0.00	0.00	0.00	0.00	0.00	4.58	1.90	1.64	2.37
8/2/2019	4.52	0.00	0.00	2.60	0.00	0.00	4.77	2.64	1.80	2.55
8/16/2019	5.16	2.57	2.31	1.63	0.00	0.00	4.48	2.66	0.00	2.52
8/30/2019	5.57	1.71	0.00	2.08	4.50	0.54	4.73	3.05	0.00	2.52
9/13/2019	5.92	0.00	0.00	0.00	0.00	0.00	2.50	3.93	0.00	2.20
9/27/2019	4.74	0.00	0.00	0.00	0.00	0.00	1.55	3.02	0.00	3.29
10/11/2019	6.16	0.00	0.00	2.58	0.00	1.81	3.37	4.52	0.00	4.73
10/25/2019	3.24	0.00	0.00	1.84	0.96	0.00	2.50	4.53	2.22	2.80
11/8/2019	4.99	0.00	0.00	2.10	1.71	1.87	3.32	3.96	2.40	3.49
11/22/2019	4.79	2.71	0.00	3.16	0.00	0.00	3.84	4.40	1.97	4.04
12/6/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.27	2.12	1.65	3.10
12/20/2019	1.07	0.00	0.00	0.00	0.00	0.00	2.38	2.61	0.00	2.53
1/24/2020	4.54	0.00	0.00	2.84	1.25	0.00	3.97	5.64	2.51	3.53
2/7/2020	3.13	0.00	0.00	0.00	0.00	0.00	1.73	2.53	0.00	3.01
2/21/2020	4.45	0.00	0.00	2.13	0.90	0.00	3.35	3.47	0.00	1.92
3/6/2020	4.24	0.00	0.00	0.00	0.00	0.00	3.00	2.73	0.00	2.06

Site 16

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
2/15/2018	0.00	0.00	0.00	0.00	0.00	0.00	1.72	3.01	0.00	2.66
3/1/2018	3.07	0.00	0.00	0.00	0.00	1.96	3.95	2.89	0.00	2.78
3/15/2018	4.49	0.00	0.00	1.79	0.00	1.89	4.88	2.74	1.55	2.75
3/29/2018	4.24	0.00	0.00	0.00	0.00	0.00	4.57	3.32	0.00	3.98
4/12/2018	4.99	0.00	0.00	1.48	0.98	1.58	5.42	3.40	0.00	2.83
4/30/2018	4.87	0.00	0.00	0.00	1.88	0.89	5.42	2.97	0.00	3.55
5/10/2018	4.67	0.00	0.00	0.00	0.00	0.00	5.01	2.59	0.00	3.07
5/24/2018	4.65	BDL	0.00	2.11	1.64	1.03	4.78	2.71	2.21	2.79
6/7/2018	4.47	0.00	0.00	0.00	2.69	2.03	5.03	2.38	2.02	2.65
6/21/2018	3.61	0.00	0.00	0.00	1.47	0.00	4.38	1.92	1.36	2.61
7/13/2018	4.52	0.00	0.10	0.00	0.00	0.00	4.36	2.71	1.75	2.82
7/19/2018	5.21	0.00	0.00	0.00	0.00	0.00	4.28	2.46	2.15	2.41
8/2/18	4.51	0.00	0.00	0.00	0.00	0.00	5.04	2.37	2.33	2.41
8/16/2018	5.02	0.00	0.00	0.00	0.00	0.00	4.18	2.72	2.22	2.42
8/31/2018	4.60	0.00	0.00	0.00	0.00	1.11	4.73	3.06	2.05	2.56
9/14/2018	4.35	0.00	0.00	0.56	0.00	0.00	3.80	2.17	1.93	2.59
9/29/2018	4.98	0.00	0.00	1.03	0.00	0.00	4.20	2.43	2.10	2.48
10/12/2018	5.37	0.54	0.00	2.23	2.37	0.00	5.18	2.91	2.56	2.92
10/26/2018	4.47	0.00	0.13	2.33	2.47	1.56	4.40	2.85	2.11	2.53
11/9/2018	4.67	0.00	0.40	1.83	1.87	0.00	4.40	3.14	1.98	1.82
11/30/2018	4.04	0.00	0.00	0.69	0.00	1.32	3.56	1.30	2.08	2.99
12/16/2018	4.16	BDL	0.00	0.73	2.01	2.07	3.94	1.95	2.05	2.00
1/4/2019	5.30	2.46	2.29	3.35	3.77	3.22	5.27	3.84	3.03	2.47
1/18/2019	4.42	0.00	0.00	1.74	2.55	2.60	4.02	2.14	2.29	1.20
2/1/2019	3.82	0.00	0.00	1.78	0.00	1.64	3.74	1.72	2.14	1.13
2/15/2019	2.83	0.00	0.00	0.00	1.59	0.00	3.13	2.08	1.71	1.40
3/1/2019	4.00	0.00	0.00	0.76	0.00	0.00	3.97	1.88	2.06	2.63
3/18/2019	3.71	0.00	0.00	0.00	1.65	1.78	3.66	1.88	2.04	1.74
3/29/2019	4.37	0.00	0.00	0.00	0.00	1.42	4.36	2.14	1.85	2.61
4/12/2019	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
4/26/2019	4.29	1.29	0.00	1.76	1.93	0.00	4.43	3.08	2.39	1.98
5/10/2019	4.58	0.00	0.00	2.68	3.18	0.00	4.54	3.52	2.56	2.08
5/24/2019	5.23	0.00	0.00	1.36	0.00	0.00	5.00	2.65	1.87	0.09
6/7/2019	5.65	0.00	0.00	0.00	0.00	0.00	4.79	3.85	1.22	1.90
6/21/2019	5.93	0.00	0.00	0.00	0.00	0.00	5.10	2.99	1.56	1.91

7/8/2019	4.50	2.12	0.00	1.35	0.00	0.00	4.54	2.65	1.73	0.90
7/19/2019	5.12	0.00	0.00	0.00	0.00	0.00	5.21	2.40	0.00	2.20
8/2/2019	4.67	0.00	0.00	0.00	0.00	0.00	4.83	2.26	0.00	1.83
8/16/2019	4.98	0.00	0.00	0.00	0.00	0.00	5.14	1.88	0.00	1.90
8/30/2019	0.00	0.00	0.00	0.00	0.00	0.00	3.76	0.00	0.00	0.00
9/13/2019	6.06	0.00	0.00	2.21	0.00	0.00	2.79	2.86	0.00	2.46
9/27/2019	5.84	0.00	0.00	0.00	2.15	2.25	3.24	0.00	0.00	2.09
10/11/2019	4.75	0.00	0.00	2.09	0.00	0.00	2.93	2.85	0.00	2.18
10/25/2019	4.53	0.00	1.63	2.37	2.93	1.67	3.34	4.82	2.39	3.44
11/8/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.31	2.32	1.22	0.00
11/22/2019	2.85	0.00	0.00	0.00	1.02	1.59	2.20	2.09	1.50	2.79
12/6/2019	1.38	0.00	0.00	0.00	1.15	0.00	1.11	4.18	2.25	0.00
12/20/2019	0.00	0.00	0.00	1.46	0.00	0.00	1.23	0.00	0.00	1.89
12/20/2019	0.00	0.00	0.00	0.00	0.00	0.00	1.41	2.10	0.00	0.00
1/11/2020	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1/24/2020	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
2/7/2020	1.11	0.00	0.00	0.00	0.00	0.00	0.94	2.97	2.00	2.35
2/21/2020	5.28	1.02	0.00	3.50	0.00	2.46	4.12	2.91	0.00	2.14
3/6/2020	4.68	0.00	0.00	2.60	0.82	1.31	3.56	2.95	0.00	2.36

Site 17

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	4.11	0.00	0.00	0.00	0.00	1.80	3.74	1.85	0.00	2.84
2/15/2018	2.63	0.00	0.00	0.52	0.00	0.00	2.61	1.62	0.00	1.94
3/1/2018	3.75	0.00	0.00	0.00	0.00	0.00	3.82	2.40	0.00	3.07
3/15/2018	3.00	0.00	0.00	1.89	1.00	0.00	3.52	2.39	0.00	3.19
3/29/2018	4.58	4.28	2.65	2.72	3.25	1.00	5.23	4.52	2.49	3.28
4/12/2018	5.58	0.00	0.00	3.35	1.41	1.18	5.33	4.13	2.81	3.32
4/30/2018	4.82	0.00	0.00	2.08	0.00	0.00	5.20	2.83	1.84	3.46
5/10/2018	4.18	0.00	0.00	1.92	0.00	0.00	4.13	2.83	1.86	3.29
5/24/2018	4.41	0.00	0.00	0.00	1.83	0.00	4.67	2.10	2.35	2.57
6/7/2018	4.97	0.00	0.00	0.00	0.00	0.00	5.17	2.68	1.94	3.33
6/21/2018	4.7	0.00	0.00	4.99	0.00	0.00	4.83	2.37	2.26	3.91
7/13/2018	5.21	0.00	1.10	1.84	2.04	0.00	5.14	3.23	1.84	2.05
7/19/2018	6.29	0.00	0.00	1.19	0.00	0.00	4.95	3.43	2.48	2.31

8/2/18	4.35	0.00	0.00	0.00	0.00	0.00	4.23	2.34	1.86	2.22
8/16/2018	5.68	0.00	0.59	1.21	0.00	0.29	4.70	3.41	2.07	1.22
8/31/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.68	0.00
9/14/2018	5.17	0.19	0.00	1.51	2.25	1.51	4.30	2.22	1.76	2.65
9/29/2018	4.83	0.00	0.00	1.50	0.00	0.00	4.07	2.42	2.16	2.53
10/12/2018	4.75	0.93	0.00	2.25	2.60	0.00	4.44	2.71	2.54	2.78
10/26/2018	4.23	0.42	0.00	2.08	0.00	0.00	3.96	2.40	2.19	2.28
11/9/2018	4.46	0.88	1.05	2.40	2.09	0.00	4.64	3.04	2.38	1.85
11/30/2018	4.44	0.00	0.00	2.14	2.50	1.80	3.42	2.14	2.20	2.68
12/16/2018	4.13	0.00	0.00	1.82	0.00	0.00	4.09	2.25	0.00	1.68
1/4/2019	5.40	2.15	2.14	3.24	3.92	3.18	5.32	3.97	3.11	2.43
1/18/2019	4.66	0.24	0.22	2.59	3.17	3.21	3.84	2.54	2.14	0.00
2/1/2019	4.44	0.71	0.49	2.59	2.28	2.74	3.94	2.12	2.48	1.43
2/15/2019	3.78	0.00	0.00	0.00	0.00	0.00	3.66	2.06	1.90	1.83
3/1/2019	4.81	0.00	0.00	1.18	0.00	2.49	4.34	2.17	0.00	2.93
3/18/2019	4.53	0.00	0.00	0.00	2.25	3.11	4.18	3.21	2.10	2.37
3/29/2019	4.87	1.70	2.09	0.00	0.00	0.00	4.09	2.55	2.04	3.07
4/12/2019	4.56	0.00	0.00	0.00	0.00	0.00	4.00	2.45	2.02	2.00
4/26/2019	5.05	1.73	0.00	1.53	2.75	0.00	4.72	3.11	2.09	2.06
5/10/2019	5.42	2.74	2.14	3.72	3.68	0.00	5.62	4.29	3.36	2.45
5/24/2019	4.55	0.00	0.00	1.96	2.13	0.00	4.30	2.82	1.67	2.37
6/7/2019	5.51	1.96	0.00	2.72	0.00	0.00	4.54	2.95	1.31	1.86
6/21/2019	5.09	2.89	2.49	3.05	0.00	0.00	5.34	3.65	1.90	2.28
7/8/2019	5.01	0.00	0.00	0.00	1.91	0.00	4.36	2.89	1.23	2.22
7/19/2019	5.40	0.00	2.31	3.00	0.00	0.00	4.49	2.40	1.40	2.62
8/2/2019	5.32	0.00	0.00	3.00	0.00	0.00	4.44	3.33	2.01	2.53
8/16/2019	5.44	0.00	0.00	1.21	0.00	0.00	4.71	2.89	0.00	2.40
8/30/2019	5.00	0.00	1.54	2.99	3.64	2.62	4.18	2.52	0.00	1.64
9/13/2019	6.15	0.00	0.00	0.00	0.00	0.00	2.24	4.00	0.00	2.60
9/27/2019	6.09	0.00	0.00	2.34	0.00	0.00	2.51	4.80	0.00	2.42
10/11/2019	6.25	0.00	0.00	2.34	0.00	0.00	3.18	2.87	0.00	3.17
10/25/2019	5.51	0.00	0.00	2.63	0.00	0.00	3.76	5.31	2.37	3.94
11/8/2019	5.01	0.00	0.00	1.01	0.83	1.39	2.66	4.04	2.47	3.79
11/22/2019	5.36	0.00	0.00	2.14	1.51	2.16	3.13	3.70	2.27	3.63
12/6/2019	0.00	0.00	0.00	1.39	0.82	0.00	1.00	2.43	1.63	2.17
12/20/2019	0.00	0.00	0.00	1.28	0.00	0.00	1.40	3.17	1.20	0.00
1/10/2020	3.94	0.00	0.00	1.95	0.00	0.00	2.51	3.05	0.00	4.02
1/24/2020	4.57	0.00	0.00	2.93	0.00	1.26	2.81	3.95	1.99	2.97
2/7/2020	5.76	0.00	0.00	3.69	4.00	3.29	3.69	5.47	2.46	3.79
2/21/2020	5.77	0.00	0.00	5.19	3.24	3.90	3.80	3.44	0.00	2.56

3/6/2020	4.42	0.00	0.00	3.17	0.00	2.16	2.44	2.69	0.00	1.81

Site 18

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicke n Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	4.53	0.00	0.00	1.96	2.79	3.60	4.27	2.27	0.00	3.30
2/15/2018	4.12	0.00	0.00	1.95	1.76	2.17	3.83	1.60	0.00	3.50
3/1/2018	4.62	0.00	0.00	1.68	0.00	1.49	4.42	2.63	0.00	2.86
3/15/2018	4.61	0.00	0.00	2.80	2.54	1.35	4.34	3.82	0.00	3.56
3/29/2018	5.06	4.36	4.28	4.09	3.60	2.11	5.61	4.47	4.11	3.55
4/12/2018	3.98	0.00	0.00	0.00	0.00	0.00	4.02	2.65	0.00	2.72
4/30/2018	3.90	0.00	0.00	0.00	0.00	0.00	3.88	2.01	0.00	4.04
5/10/2018	4.09	0.00	0.00	1.43	0.00	0.00	4.16	2.19	0.00	3.34
5/24/2018	4.64	0.00	0.00	1.43	1.56	2.23	5.17	2.61	2.34	4.52
6/7/2018	4.34	0.00	0.00	0.00	0.00	0.00	4.72	2.32	2.07	3.99
6/21/2018	4.47	0.00	0.00	0.00	0.00	2.07	4.26	3.08	1.76	3.03
7/13/2018	4.60	0.00	0.00	0.00	1.85	0.00	3.75	1.90	2.04	2.23
7/19/2018	4.91	0.00	0.00	0.00	1.28	0.00	4.04	2.99	2.14	1.91
8/2/18	4.95	0.00	0.00	0.00	0.00	0.00	4.14	2.63	2.18	2.84
8/16/2018	4.32	0.00	0.00	0.00	0.00	0.00	3.76	2.43	2.15	2.43
8/31/2018	4.96	0.00	0.00	0.88	0.00	0.00	4.12	2.34	1.92	4.40
9/14/2018	5.47	1.27	0.70	2.07	2.69	3.06	5.46	2.71	1.74	2.59
9/29/2018	5.55	0.47	BDL	0.43	0.00	2.10	4.92	3.31	2.29	3.00
10/12/2018	5.45	0.42	BDL	0.71	1.58	0.00	4.70	2.52	2.06	3.55
10/26/2018	4.74	1.52	0.11	1.58	2.14	0.00	4.21	2.73	2.16	2.87
11/9/2018	5.32	0.75	1.71	2.01	3.90	1.21	5.37	4.03	1.88	2.68
11/30/2018	4.33	0.00	0.00	1.40	2.15	0.00	3.74	2.23	2.03	2.71
12/16/2018	3.87	0.00	0.00	BDL	0.00	0.00	3.76	2.44	2.03	1.41
1/4/2019	4.85	0.00	0.00	0.75	2.38	2.43	4.78	3.00	1.83	2.35
1/18/2019	4.37	0.00	0.00	0.84	1.93	1.57	3.71	2.21	1.65	1.79
2/1/2019	4.88	0.00	0.00	1.12	2.37	0.70	4.63	3.02	1.97	0.71
2/15/2019	4.40	0.00	0.00	0.88	2.00	1.39	4.25	1.95	1.61	2.83
3/1/2019	4.13	0.00	0.00	0.75	0.00	0.00	3.90	2.10	2.08	2.44
3/18/2019	4.38	0.00	0.00	0.00	2.29	2.25	3.79	3.21	1.77	2.08
3/29/2019	4.37	0.00	0.00	0.00	0.00	1.86	4.00	2.40	1.42	2.44
4/12/2019	4.17	0.00	0.00	0.00	0.00	0.00	3.93	2.54	2.09	1.44

4/26/2019	4.25	0.00	0.00	0.00	2.22	0.00	3.96	2.04	1.66	1.73
5/10/2019	5.15	2.38	1.73	3.45	3.56	1.68	5.11	3.96	3.40	2.35
5/24/2019	4.75	1.91	0.00	1.97	0.00	0.00	4.43	2.76	1.18	1.53
6/7/2019	5.07	1.90	0.00	2.49	0.00	0.00	5.03	3.92	1.02	2.06
6/21/2019	4.98	0.00	0.00	0.00	0.00	0.00	4.64	2.23	0.96	2.11
7/8/2019	5.24	1.31	0.00	2.06	2.59	0.00	5.61	3.26	1.39	1.91
7/19/2019	5.17	0.00	0.00	0.00	0.00	0.00	4.35	2.49	1.85	2.97
8/2/2019	4.78	0.00	0.00	1.89	0.00	0.00	4.39	2.36	0.00	2.07
8/16/2019	6.34	0.00	0.00	2.93	4.07	4.93	5.91	4.23	1.96	4.00
8/30/2019	5.79	0.00	0.00	3.18	3.43	3.29	5.98	3.72	1.63	3.47
9/13/2019	6.09	0.00	0.00	1.82	0.00	0.00	2.43	4.26	0.00	2.42
9/27/2019	6.14	0.00	0.00	2.27	3.05	0.82	2.46	3.37	0.00	3.59
10/11/2019	6.66	0.00	0.00	1.81	0.00	0.43	3.12	3.85	0.00	6.58
10/25/2019	5.47	0.00	1.73	3.09	3.86	2.03	4.23	5.54	2.10	4.33
11/8/2019	5.42	0.00	1.40	2.52	0.00	1.78	3.35	4.34	2.28	3.80
11/22/2019	5.39	0.00	0.00	2.65	1.37	2.03	3.10	4.42	1.70	3.77
12/6/2019	1.71	0.00	0.00	0.00	0.91	0.00	1.43	2.39	2.11	0.00
12/20/2019	4.01	0.00	0.00	2.12	0.00	1.83	2.61	2.94	0.00	4.29
1/10/2020	4.74	0.00	0.00	2.34	0.00	2.08	2.60	2.96	0.00	4.65
1/24/2020	4.75	0.92	0.00	2.31	1.99	2.85	2.86	4.68	0.00	3.50
2/7/2020	4.18	0.00	0.00	2.79	1.22	2.13	2.74	3.48	1.87	0.00
2/21/2020	5.26	0.00	0.00	1.75	2.28	3.19	3.84	4.77	1.71	1.18
3/6/2020	5.08	0.00	0.00	1.87	0.83	2.94	3.42	2.44	0.00	2.11

Site 19

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicken Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
2/15/2018	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.85
3/1/2018	4.50	0.00	0.00	0.00	0.00	1.66	4.32	3.11	0.00	2.50
3/15/2018	0.00	0.00	0.00	0.00	0.00	0.00	2.77	2.59	0.00	2.06
3/29/2018	4.67	3.89	3.78	3.72	3.06	0.00	5.53	4.19	3.85	3.51
4/12/2018	4.04	0.00	0.00	0.00	0.00	0.00	3.78	2.53	0.00	2.63
4/30/2018	5.76	4.05	3.44	1.80	0.00	1.57	6.65	4.40	0.00	3.46
5/10/2018	3.56	0.00	0.00	0.00	0.00	0.00	3.65	2.47	0.00	2.91
5/24/2018	4.18	0.00	0.00	0.00	1.82	0.00	4.27	2.94	1.60	2.91
6/7/2018	4.22	0.00	0.00	0.00	0.00	0.00	4.40	1.40	1.93	2.65
6/21/2018	4.66	0.00	0.00	0.00	0.00	0.00	4.28	2.95	1.66	2.63

7/13/2018	4.72	0.00	0.00	0.00	0.00	0.00	4.00	2.67	1.31	1.76
7/19/2018	4.75	1.32	0.15	0.00	1.35	0.00	3.83	2.93	2.53	2.10
8/2/18	4.74	0.00	0.00	0.00	1.66	0.00	4.14	2.70	2.33	2.31
8/16/2018	4.62	0.00	BDL	0.97	2.00	0.00	4.48	2.33	1.92	2.17
8/31/2018	5.35	0.00	0.00	1.07	0.00	0.00	4.33	2.81	2.01	2.86
9/5/2018	4.63	0.00	0.00	1.29	2.27	0.00	4.54	3.71	2.42	2.11
9/14/2018	4.65	1.02	0.00	0.07	2.14	0.00	4.06	2.63	2.06	2.26
9/29/2018	4.89	0.00	0.00	0.00	2.09	1.26	3.93	2.62	2.64	3.01
10/12/2018	4.96	0.00	0.00	0.00	0.00	0.00	4.16	2.52	2.17	2.71
10/16/2018	5.14	1.40	0.17	1.92	3.37	0.00	5.13	3.67	2.17	3.05
10/26/2018	4.75	0.96	0.10	1.97	2.33	0.00	4.52	2.91	2.28	2.92
11/9/2018	5.37	0.92	0.00	2.40	3.98	0.00	5.50	4.21	2.27	2.65
11/30/2018	4.38	0.00	0.00	1.03	2.52	1.91	3.63	2.04	2.07	3.24
12/16/2018	3.52	0.00	0.00	0.10	0.00	0.00	3.06	1.91	1.89	0.00
1/4/2019	4.71	0.00	0.00	1.23	3.00	2.94	4.13	2.71	1.91	2.03
1/18/2019	5.14	0.00	0.00	0.90	2.61	3.58	3.89	2.48	1.87	0.00
2/1/2019	4.07	0.00	0.00	0.00	1.75	0.00	3.59	2.07	2.14	0.00
2/15/2019	3.76	0.00	0.00	0.00	0.00	0.00	3.42	2.60	1.53	2.14
3/1/2019	4.15	0.00	0.00	0.06	0.00	0.00	3.85	2.08	1.71	2.33
3/18/2019	3.49	0.00	0.00	1.08	0.00	1.04	3.10	1.96	1.49	2.14
3/29/2019	4.68	0.00	0.00	0.65	0.00	0.00	4.19	1.81	1.78	2.39
4/12/2019	4.67	0.00	0.00	0.00	0.00	0.00	4.19	2.63	2.03	1.11
4/26/2019	4.94	0.00	0.00	1.64	3.14	3.07	4.53	2.75	1.67	1.83
5/10/2019	5.05	2.00	2.21	3.23	3.57	0.00	5.01	3.82	3.24	2.32
5/24/2019	4.54	1.93	2.16	0.00	0.00	0.00	3.87	3.14	1.00	1.32
6/7/2019	4.81	1.86	2.22	0.64	1.99	0.00	4.76	3.77	0.00	2.47
6/21/2019	4.66	0.00	0.00	0.00	0.00	0.00	3.69	2.80	1.05	1.67
7/8/2019	5.09	2.01	0.00	1.70	0.00	1.87	4.52	2.75	0.96	2.01
7/19/2019	5.17	0.00	0.00	0.00	0.00	0.00	4.37	2.42	1.56	3.00
8/2/2019	4.91	0.00	0.00	1.98	1.66	0.00	4.14	2.03	0.00	2.39
8/16/2019	4.60	0.00	0.00	2.34	1.70	0.00	4.16	2.67	1.67	2.29
8/30/2019	5.08	0.00	1.89	3.07	2.72	2.68	4.67	3.14	1.57	3.86
9/13/2019	6.26	0.00	0.00	1.43	2.37	0.00	2.72	3.86	0.00	2.18
9/27/2019	5.94	0.00	0.00	2.05	3.50	2.23	2.50	3.44	0.00	3.77
10/11/2019	6.35	0.00	0.00	0.00	0.00	0.00	3.74	3.54	0.00	4.58
10/25/2019	5.55	0.00	2.49	2.99	3.87	2.05	3.98	5.43	2.33	4.02
11/8/2019	4.94	0.00	0.00	1.14	1.43	1.78	2.77	3.82	1.10	3.73
11/22/2019	4.91	0.00	0.00	3.50	2.89	3.35	2.90	4.79	1.89	4.21
12/6/2019	0.00	0.00	0.00	1.41	0.00	0.00	1.52	2.24	2.40	0.00
12/20/2019	6.07	1.26	0.00	3.11	2.40	3.25	4.55	4.75	1.34	3.67

1/10/2020	5.93	0.80	2.26	3.61	3.10	4.01	4.04	5.50	0.00	4.93
1/24/2020	4.74	0.88	0.00	1.64	0.00	2.23	2.79	3.63	0.00	3.56
2/7/2020	4.98	0.00	3.06	2.19	1.72	2.46	3.23	3.23	0.00	2.26
2/21/2020	5.22	0.00	2.02	2.58	0.00	1.53	3.47	3.48	0.00	2.46
3/6/2020	5.19	2.01	2.57	1.46	0.00	3.29	3.31	2.61	0.00	2.69

Site 21

Date of sample collection	Bac Uni	Bac Hum	HF183	Bac Cow	Bac Can	Chicke n Duck Bac	Entero 1	*E. coli*	Rum2 Bac	GFD
1/30/2018	3.28	0.00	0.00	0.00	1.75	0.00	4.03	2.40	0.00	2.49
2/15/2018	2.81	0.00	0.00	0.00	0.00	0.00	3.01	1.10	0.00	2.48
3/1/2018	3.40	1.36	0.00	0.00	0.00	0.00	3.95	2.73	0.00	3.30
3/15/2018	3.68	0.00	0.00	0.00	0.00	0.00	3.61	2.60	0.00	3.71
3/29/2018	4.32	3.69	3.47	2.98	3.45	1.69	5.31	4.52	2.99	3.47
4/12/2018	3.87	0.00	0.00	1.68	0.00	0.00	4.30	2.79	0.00	3.06
4/30/2018	3.60	0.00	0.00	0.00	0.00	0.00	4.00	2.89	0.00	3.60
5/10/2018	4.25	0.00	0.00	0.00	0.00	0.00	4.43	2.84	0.00	4.14
5/24/2018	4.34	0.00	0.00	0.00	0.00	0.00	4.80	3.43	2.08	3.13
6/7/2018	3.90	0.00	0.00	0.00	0.00	0.00	4.19	2.49	1.96	2.98
6/21/2018	3.96	0.00	0.00	0.00	2.08	0.00	3.89	2.38	1.79	2.95
7/13/2018	4.18	0.00	0.00	0.00	1.41	0.00	3.72	2.23	2.07	3.20
7/19/2018	4.11	0.00	0.00	0.00	1.95	0.00	3.37	2.77	2.12	2.09
8/2/18	3.79	0.00	0.00	0.00	0.00	0.00	3.32	2.73	2.34	1.66
8/16/2018	4.50	0.00	0.00	0.00	1.99	0.00	3.73	2.45	2.17	2.87
9/13/2018	4.90	2.52	1.83	1.01	1.94	0.00	4.23	3.51	1.92	1.76
9/29/2018	4.84	0.00	0.00	0.00	2.08	0.00	4.20	3.28	2.13	2.45
10/12/2018	5.17	0.00	0.00	0.71	0.00	0.00	4.21	2.60	0.00	2.97
10/16/2018	5.33	BDL	0.29	2.07	3.12	2.39	4.80	3.54	2.43	2.69
10/26/2018	4.82	1.84	1.06	1.38	2.12	2.73	4.04	2.72	0.00	2.63
11/9/2018	5.60	BDL	0.00	2.64	2.67	2.77	5.11	3.57	2.42	1.82
11/30/2018	4.26	0.10	0.70	BDL	0.00	0.00	3.08	2.06	2.14	2.27
12/16/2018	3.47	0.00	0.00	0.00	0.00	0.00	3.10	1.70	1.68	1.10
1/4/2019	4.47	0.03	0.00	1.90	2.43	2.36	4.11	2.83	1.79	1.43
1/18/2019	4.90	0.00	0.00	0.46	2.64	0.00	4.13	2.51	1.80	1.75
2/1/2019	3.99	0.00	0.00	0.00	0.00	0.00	3.25	1.37	1.88	0.00
2/15/2019	3.95	0.00	0.00	0.00	2.27	0.00	3.45	2.89	1.62	2.25

3/1/2019	3.53	0.00	0.00	0.00	0.00	0.00	3.46	2.29	1.46	2.12
3/18/2019	4.03	0.00	0.00	0.00	0.00	1.53	3.69	2.35	1.60	1.92
3/29/2019	4.32	0.00	0.00	0.00	1.54	0.00	3.89	4.24	1.20	2.35
4/12/2019	4.40	0.00	0.00	0.00	0.00	0.00	3.92	2.67	2.01	1.30
4/26/2019	4.92	0.00	0.00	BDL	1.96	0.00	4.54	2.95	2.27	1.61
5/10/2019	4.52	2.03	0.00	2.67	3.33	0.00	4.50	3.35	2.84	1.32
5/24/2019	5.27	0.00	0.00	1.34	0.00	0.00	4.75	3.69	0.91	2.31
6/7/2019	4.98	0.00	0.00	0.00	0.00	0.00	4.22	3.50	1.07	2.96
6/21/2019	1.87	0.00	0.00	0.00	0.00	0.00	2.34	2.07	0.98	0.00
7/8/2019	5.06	0.00	0.00	0.76	0.00	0.00	4.10	2.93	0.00	2.53
7/19/2019	4.96	1.47	0.00	0.00	0.00	0.00	4.11	2.20	0.00	2.33
8/2/2019	5.14	1.61	0.00	0.00	0.00	0.00	4.10	2.69	1.61	1.92
8/16/2019	4.64	0.00	0.00	0.92	0.00	0.00	4.24	2.67	0.00	2.45
8/30/2019	4.59	0.00	0.00	2.85	0.00	0.00	4.23	3.13	1.74	2.95
9/13/2019	6.04	0.00	0.00	1.94	0.00	0.00	2.62	4.06	0.00	2.41
9/27/2019	5.42	0.00	0.00	2.00	0.00	0.00	1.99	3.32	0.00	2.52
10/11/2019	6.81	0.00	0.00	2.70	0.00	0.00	3.62	4.35	0.00	2.89
10/25/2019	4.70	0.00	1.88	2.88	2.81	0.00	3.63	4.95	2.60	3.61
11/8/2019	5.30	0.00	0.00	2.34	2.15	0.00	3.64	4.85	2.53	4.21
11/22/2019	4.15	0.00	0.00	1.86	0.00	0.00	2.48	2.51	2.05	3.51
12/6/2019	1.93	0.00	0.00	0.00	0.84	0.00	1.51	1.74	2.67	0.00
12/20/2019	3.60	0.00	0.00	1.67	1.24	0.00	2.97	3.54	1.72	3.48
1/10/2020	4.10	0.00	0.00	1.77	0.00	0.00	2.27	3.05	0.00	3.37
1/24/2020	4.47	0.00	0.00	1.75	1.36	0.00	2.56	3.92	0.00	3.77
2/7/2020	4.62	0.00	0.00	0.56	0.00	0.00	3.30	2.32	0.00	1.83
2/21/2020	4.71	0.00	0.00	0.00	0.00	0.00	3.20	2.17	0.00	2.15
3/6/2020	4.47	0.00	0.00	1.53	0.00	0.00	2.65	2.35	0.00	1.96