

Prepared in cooperation with Bernalillo County

Assessing *Escherichia coli* and Microbial Source Tracking Markers in the Rio Grande in the South Valley, Albuquerque, New Mexico, 2020–21



Scientific Investigations Report 2023–5019

Cover.

Left: Joseph Beman wading the Rio Grande at Rio Bravo bridge.

Top right: Kimberly Beisner sampling the Rio Grande.

Middle right: Kate Wilkins kayaking on the Rio Grande.

Bottom right: Sediment in the Rio Grande.

Assessing *Escherichia coli* and Microbial Source Tracking Markers in the Rio Grande in the South Valley, Albuquerque, New Mexico, 2020–21

By Rebecca E. Travis, Kate L. Wilkins, and Christopher M. Kephart

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Conversion Factors

U.S. customary units to International System of Units

Multiply	By	To obtain
Length		
inch (in.)	2.54	centimeter (cm)
inch (in.)	25.4	millimeter (mm)
foot (ft)	0.3048	meter (m)
mile (mi)	1.609	kilometer (km)
Area		
square mile (mi ²)	259.0	hectare (ha)
square mile (mi ²)	2.590	square kilometer (km ²)
Volume		
ounce, fluid (fl. oz)	0.02957	liter (L)
Flow rate		
cubic foot per second (ft ³ /s)	0.02832	cubic meter per second (m ³ /s)
million gallons per day (Mgal/d)	0.04381	cubic meter per second (m ³ /s)

International System of Units to U.S. customary units

Multiply	By	To obtain
Length		
centimeter (cm)	0.3937	inch (in.)
millimeter (mm)	0.03937	inch (in.)
Volume		
liter (L)	33.81402	ounce, fluid (fl. oz)

Temperature in degrees Celsius (°C) may be converted to degrees Fahrenheit (°F) as follows:

$$^{\circ}\text{F} = (1.8 \times ^{\circ}\text{C}) + 32.$$

Datum

Horizontal coordinate information is referenced to the North American Datum of 1983 (NAD 83).

Supplemental Information

Specific conductance is given in microsiemens per centimeter at 25 degrees Celsius ($\mu\text{S}/\text{cm}$ at 25 °C).

Concentrations of chemical constituents in water are given in milligrams per liter (mg/L).

A water year is the period from October 1 to September 30 and is designated by the year in which the period ends.

Abbreviations

AU	assessment unit
BEMP	Bosque Ecosystem Monitoring Program
COA	City of Albuquerque
DNA	deoxyribonucleic acid
EPA	U.S. Environmental Protection Agency
HUC	hydrologic unit code
MST	microbial source tracking
NADA	Nondetects and Data Analysis (software package)
NMED	New Mexico Environment Department
NMEDSWQB	New Mexico Environment Department Surface Water Quality Bureau
NMWQS	New Mexico Water Quality Standard
NMWSC	New Mexico Water Science Center
OWML	Ohio Water Microbiology Laboratory
qPCR	quantitative polymerase chain reaction
RSD	relative standard deviation
SD	standard deviation
TMDL	total maximum daily load
USGS	U.S. Geological Survey
WWTP	wastewater treatment plant

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Abstract

The Rio Grande, in southern Albuquerque, New Mexico, is a Clean Water Act Section 303(d) Category 5 impaired reach for *Escherichia coli* (*E. coli*). The reach is 5 miles in length, extending from Tijeras Arroyo south to the Isleta Pueblo boundary. An evaluation of *E. coli* and microbial source tracking markers (human-, canine-, and waterfowl-specific sources) was conducted by the U.S. Geological Survey to determine the extent and source of fecal bacteria within the impaired reach of the Rio Grande, primarily during the dry season (November through June) in 2020 and 2021. Samples were collected in the river cross section at three locations within each site and collected during both the dry season and the wet season, thereby providing data over a range of flow conditions to better understand the extent and source of fecal bacteria. Because fecal microorganisms may readily attach to sediments, riverbed material samples were also collected. During the dry season, *E. coli* concentrations in water were primarily detected below the New Mexico Surface Water Quality Standard of 410 colony forming units per 100 milliliters and mostly human and canine sources were detected. However, approximately 40 percent of the water samples exceeded the Isleta Pueblo water quality standard of 88 colony forming units per 100 milliliters. *E. coli* concentrations in bed material were detected at low concentrations, and the bed material was a sandy substrate, with little fine-grained material, a suitable habitat that would allow for bacterial growth during the dry season. Significant spatial and temporal differences, where p-values were less than 0.05, were found in water-quality samples for *E. coli* (seasonal) and the human tracker concentrations (between sites and within a cross section of a site). Given the lack of correlation between discharge and *E. coli* concentration and the human marker being most prevalent in the study area, the sources of *E. coli* in the dry season are likely nonpoint sources. The results from this study will help decision makers determine the efficacy of their best management practices and guide new practices to improve water quality in the reach.

Introduction

The Rio Grande is a major river that spans New Mexico from its northern border to southern border (fig. 1). The Clean Water Act Section 303(d) Category 5 requires listing a stream, river, or lake as impaired if a water-quality standard is exceeded (U.S. Environmental Protection Agency [EPA], 2022a). The New Mexico Environment Department (NMED) assesses and lists impaired waters in New Mexico every 2 years (New Mexico Environment Department Surface Water Quality Bureau [NMEDSWQB], 2019a). Impairments are listed by segments known as assessment units (AUs), which are segments of surface waters with assumed homogenous water quality and are based on geology, topography, incoming tributaries, and surrounding land use/land management (NMEDSWQB, 2022a).

Several AUs on the Rio Grande have been listed by NMED over the years as impaired by *Escherichia coli* (*E. coli*), with concentrations exceeding water-quality standards (NMEDSWQB, 2019a). Since 2010, impaired AUs in the urbanized area of Albuquerque (figs. 1 and 2) have included the following, as named and numbered by NMED:

- Rio Grande (Alameda Bridge to U.S. Highway No. 550, 12.12 miles [mi] in length), AU identification number (ID): NM-2105.1_00
- Rio Grande (Tijeras Arroyo to Alameda Bridge, 15.6 mi in length), AU ID: NM-2105_51
- Rio Grande (Isleta Pueblo boundary to Tijeras Arroyo, 5.14 mi in length), AU ID: NM-2105_50

The study area extends from Bridge Boulevard (Blvd) site to the Valle de Oro site, and includes parts of the NM-2105_51 and AU NM-2105_50 AUs (fig. 2). The AU NM-2105_50 has been consistently listed as impaired for *E. coli*, which is why it is the focus of this study.

When these AUs were listed by NMED as impaired from *E. coli*, a total maximum daily load (TMDL) was then calculated for each AU. A TMDL is the calculation of the maximum amount of a contaminant allowed to enter a waterbody so that the waterbody will meet and continue to meet water-quality

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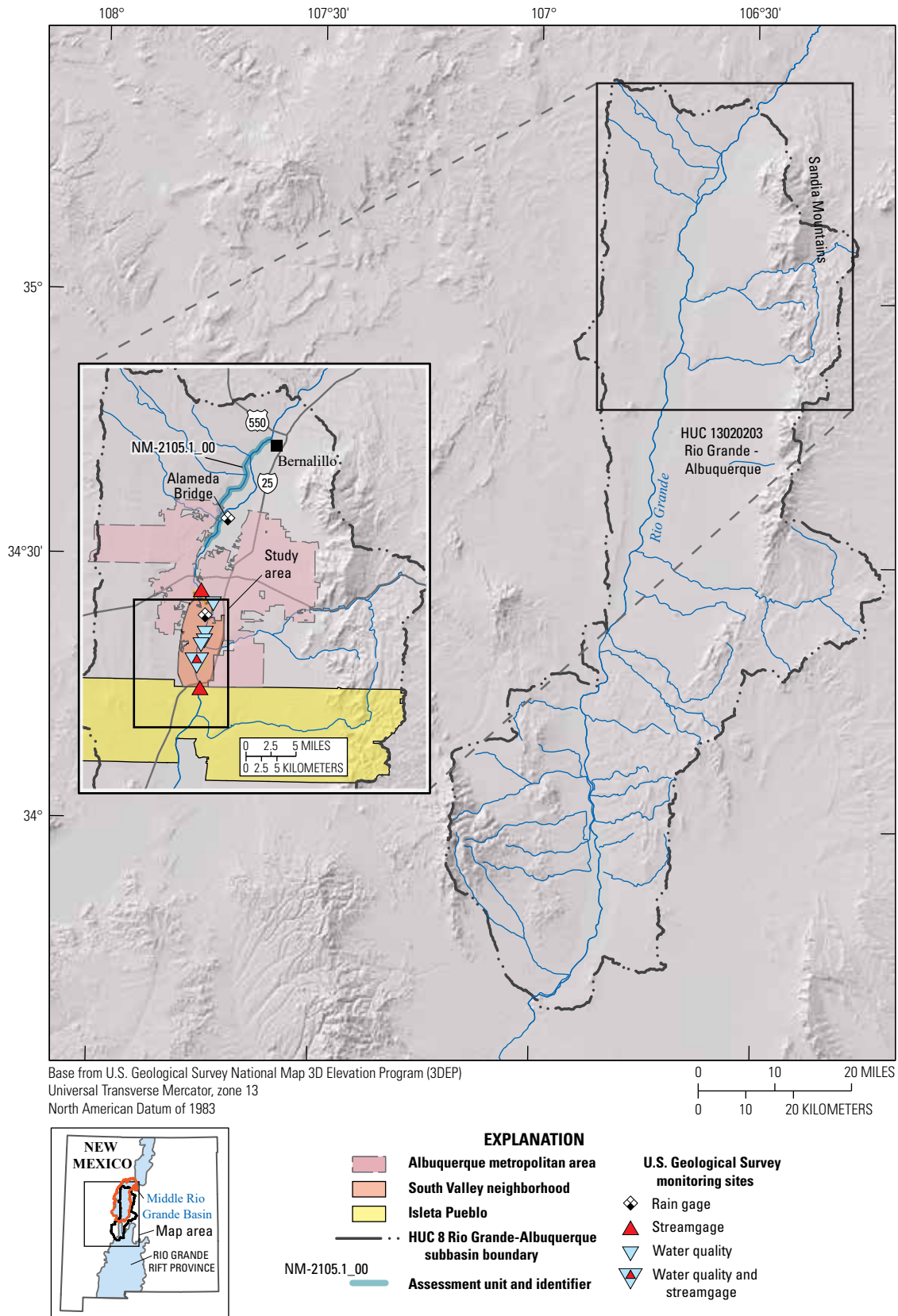


Figure 1. Region surrounding study area within the Rio Grande-Albuquerque hydrologic unit code (HUC) 8 and HUC 10 watersheds in central New Mexico.

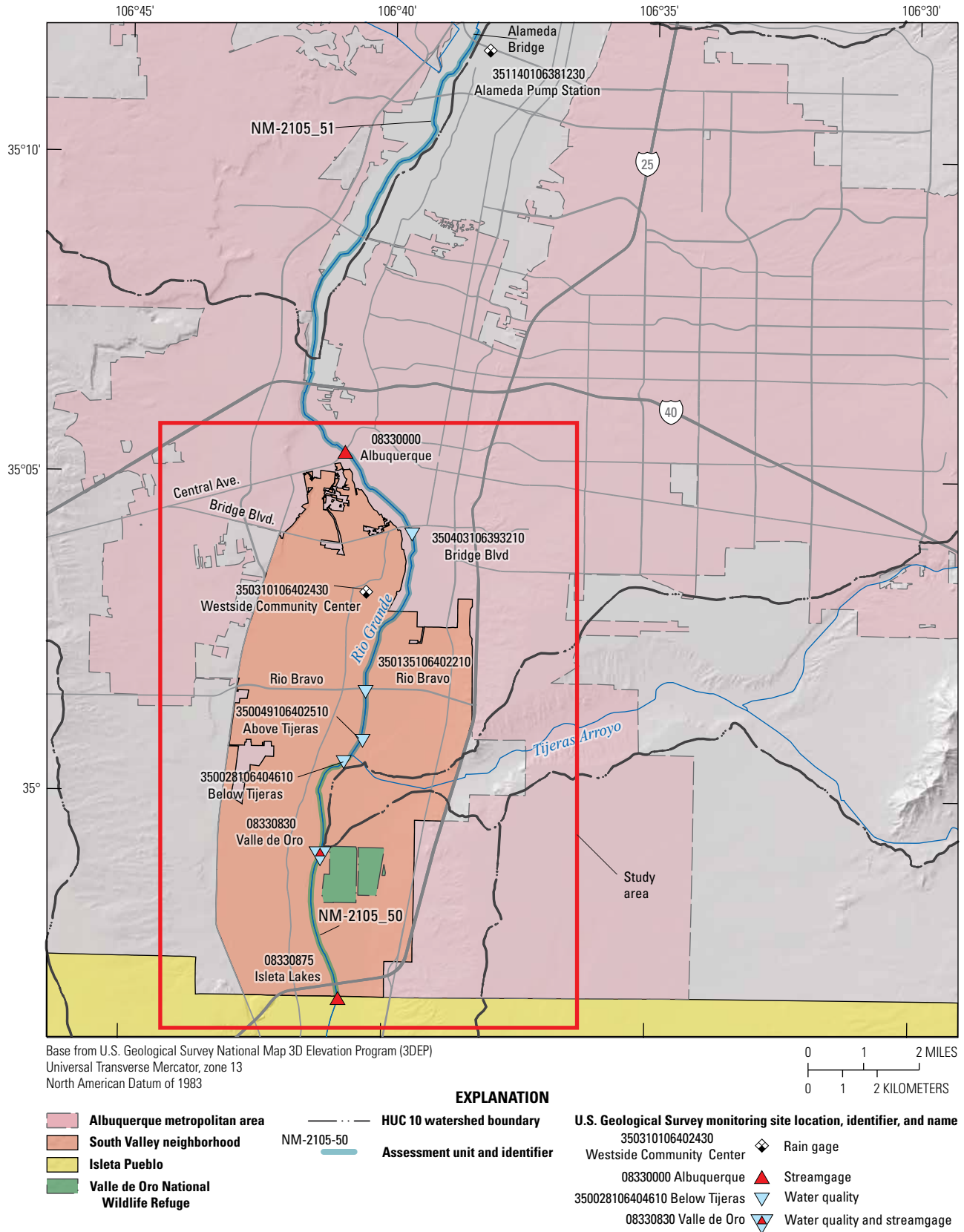


Figure 2. Study area in Albuquerque, New Mexico, the South Valley neighborhood and impaired assessment units (segments) of the Rio Grande. HUC, hydrologic unit code.

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standards for that contaminant (EPA, 2022b). The TMDL for *E. coli* ranges from 1.89×10^{11} colony forming units per 100 milliliters per day (cfu/100 mL/day) in low flow conditions to 5.27×10^{12} cfu/100 mL/day in high flow conditions within the segment of the Rio Grande between Alameda Bridge and Isleta Pueblo, which includes two AUs listed above and extends upstream and downstream from the study area (NMEDSWQB, 2019b).

The New Mexico Water Quality Standard (NMWQS) 20.6.4.105 New Mexico Administrative Code (New Mexico Water Quality Control Commission, 2010) designates the Rio Grande in the study area for the following uses: irrigation, marginal warm water aquatic life, livestock watering, public water supply, wildlife habitat, and primary contact. Primary contact (NMWQS 20.6.4.900), which is defined as water use “in which there is prolonged human contact” for recreational or ceremonial purposes, has a more stringent standard for *E. coli* for a single sample (410 colony forming units per 100 milliliters [cfu/100 mL] or a 30-day geometric mean of 126 cfu/100 mL). NMWQS 20.6.4.105 excludes the waters of Isleta Pueblo, which has its own water-quality standards that are allowed to be lower than State standards because of their ceremonial use of water (EPA, 2021a). The Isleta Pueblo standard for *E. coli* for a single sample is 88 cfu/100 mL and a 30-day geometric mean of 47 cfu/100 mL for primary contact ceremonial and recreational use (EPA, 2021a).

In 2018, after implementation of best management practices in the early to mid-2010s, the northern two of these three AUs were no longer listed as impaired for *E. coli* on the Section 303(d) list, but the impairment listing for *E. coli* remained on the downstream AU, from the Isleta Pueblo boundary to the Tijeras Arroyo (NMEDSWQB, 2019a). The most downstream AU was the initial study area targeted for the project in 2019, whereas the upstream portion (Bridge Blvd to Tijeras Arroyo) was added to include an upstream area that was not listed for *E. coli*. However, the two upstream AUs were relisted for *E. coli* in 2020 following a July 2017–May 2018 study conducted by Ciudad Soil and Water Conservation Service (Fluke, 2018; NMEDSWQB, 2021). With that relisting, the study area encompassed two impaired AUs for *E. coli*.

While *E. coli* concentrations are expected to be elevated during storm events and during the wet season (July 1–October 31), results from previous monitoring, discussed in the “Previous Monitoring” section, have also shown elevated *E. coli* concentrations during the dry season (November 1–June 30), of up to 3,076 most probable number per 100 milliliters (MPN/100 mL) (Bosque Ecosystem Monitoring Program [BEMP], 2021). Therefore, additional information was needed to determine the *E. coli* source during the dry season.

The objective of this study, conducted by the U.S. Geological Survey (USGS), was to determine the extent and source of fecal-origin bacteria, in both water and bed material, within these impaired AUs of the Rio Grande during the dry season. The extent and source of fecal-origin bacteria were evaluated by using microbial source tracking (MST), a method used for identifying sources of fecal contamination

in the environment. MST methods are based on the concept that intestinal systems of different warm-blooded animals have specific microbial populations to account for differences in diet and physiology. Therefore, different MST markers are associated with different sources (Francy and Stelzer, 2012). In this study, human-, canine-, and waterfowl-specific sources were evaluated.

Purpose and Scope

The purpose of this report is to evaluate the concentrations and sources, in both water and bed material, of fecal bacteria in the impaired reach of the Rio Grande. Spatial and temporal variabilities were analyzed, and instantaneous loads were calculated. Learning more about the extent of fecal-origin bacteria and its sources during the dry season will enable decision makers to focus best management practices to reduce *E. coli* concentrations from both point sources (discrete and discernible) and nonpoint sources (diffuse) in the Rio Grande watershed.

Study Area

The study area is in the unincorporated South Valley of Albuquerque, New Mexico (figs. 1 and 2), which is more agricultural than the metropolitan area of Albuquerque and has been for over 400 years (Visit Albuquerque, 2021). Historically, the southern part of the South Valley is more rural, and residences rely on groundwater wells and septic systems instead of city water and sewer systems. The current septic use based on Bernalillo County wastewater atlas data is estimated to be approximately 2,100 permitted systems in the South Valley and approximately 980 unpermitted systems (D McGregor, Bernalillo County, written commun., 2022). Five water-quality sites located within two impaired AUs along the Rio Grande were sampled in this study (table 1, fig. 2).

Watershed Characterization

Hydrologic unit codes (HUC) are units by which the watersheds of the United States are classified. The HUCs are defined at different levels with smaller HUCs nested within larger HUCs, from the smallest geographic area (cataloging unit) to the largest geographic area (region) (USGS, 2022a). Albuquerque and the South Valley are within the HUC 8 watershed 13020203, the Rio Grande-Albuquerque watershed, with an area of 3,216 square miles (mi²) (fig. 1). The HUC 10 watershed where the study sites are located is HUC 1302020303 (City of Albuquerque–Rio Grande, 269 mi²). Directly upstream from the sites is HUC 1302020301 (Arroyo de Las Calabacillas–Rio Grande, 330 mi²) (fig. 3). HUC 1302020302 (Tijeras Arroyo, 132 mi²) feeds into the Rio Grande between the Above Tijeras and Below Tijeras Arroyo sites (figs. 2 and 3).

Table 1. Study area sites, including water-quality sampling sites, streamgages, and precipitation gages.

[Horizontal coordinate information is referenced to the North American Datum of 1983. USGS, U.S. Geological Survey; NM, New Mexico; Blvd, boulevard; nr, near]

USGS site number	USGS site name	Short name	Station type	Decimal latitude	Decimal longitude
08330000	Rio Grande at Albuquerque, NM	Albuquerque	Continuous discharge	35.08916667	-106.6806944
350403106393210	Rio Grande below Bridge Blvd	Bridge Blvd	Water quality, sediment, discrete discharge	35.06765	-106.6590583
350135106402210	Rio Grande below Rio Bravo	Rio Bravo	Water quality, sediment, discrete discharge	35.02640278	-106.673
350049106402510	Rio Grande above Tijeras Arroyo	Above Tijeras	Water quality, sediment, discrete discharge	35.01361944	-106.6737139
350028106404610	Rio Grande below Tijeras Arroyo	Below Tijeras	Water quality, sediment, discrete discharge	35.00798611	-106.6795694
08330830	Rio Grande at Valle de Oro, NM	Valle de Oro	Water quality, sediment, continuous discharge	34.98333333	-106.6865556
08330875	Rio Grande at Isleta Lakes nr Isleta, NM	Isleta Lakes	Continuous discharge	34.94666667	-106.6802778
350310106402430	Westside Community Center nr Albuquerque, NM	Westside Community Center	Precipitation	35.05277778	-106.6733333
351140106381230	Alameda Pump Station Rain Gage	Alameda Pump Station	Precipitation	35.19444444	-106.6366667

The three HUC 10 watersheds contributing to the study area vary in land cover (fig. 3; Dewitz, 2021). The Arroyo de Las Calabacillas-Rio Grande watershed, located directly upstream from the sites, contains 87 percent shrubland and is mostly undeveloped. The Tijeras Arroyo watershed contains approximately 94 percent shrubland and forested land. The City of Albuquerque-Rio Grande watershed is 34 percent developed and 47 percent shrubland with a small component (4 percent) of pasture, hay, and crops.

There are several potential point and nonpoint sources of *E. coli* in the South Valley (fig. 4). According to the EPA, sources of fecal indicator bacteria can include leaking septic systems, wastewater treatment plants, stormwater runoff, and domestic animal and wildlife waste (EPA, 2021b). On the basis of locations of existing water infrastructure, the density of water infrastructure is lower in the southeastern portion of the study area (M. Haley, Albuquerque Bernalillo County Water Utility Authority, written commun., 2021). If there is less water infrastructure in the area, it is more likely that residences will be on septic systems. LaMotte (2018) used 1990 and 2010 census data to estimate the number of people on septic systems in the coterminous United States on the basis of

census blocks from the 2010 census. In the South Valley, the estimated number of people on septic systems was 13,320, as compared to the total population of 46,807 (fig. 4). However, Bernalillo County estimates that number to be much lower (approximately 3,000 systems) after initiatives to transition residences to municipal wastewater lines in the late 2000s (D. McGregor, Bernalillo County, written commun., 2022). Additional nonpoint sources similar in signature to leaking septic systems could include leaking sewer lines, sewer overflows into stormwater drains, illegal septage discharge from haulers into drains or the river, or public defecation into drainages in the study area.

There are several point sources within the study area. A wastewater treatment plant (WWTP) is located along the Rio Grande upstream from the confluence of Tijeras Arroyo (figs. 2 and 4), which allows for a 30-day average of 47 cfu/100 mL and a daily maximum of 88 cfu/100 mL for *E. coli* (NMEDSWQB, 2019c). The WWTP is the largest wastewater treatment plant in New Mexico, currently treating 50 to 60 million gallons of wastewater per day (Albuquerque Bernalillo County Water Utility Authority, 2023). A dog park is located just east of the Rio Grande and upstream from

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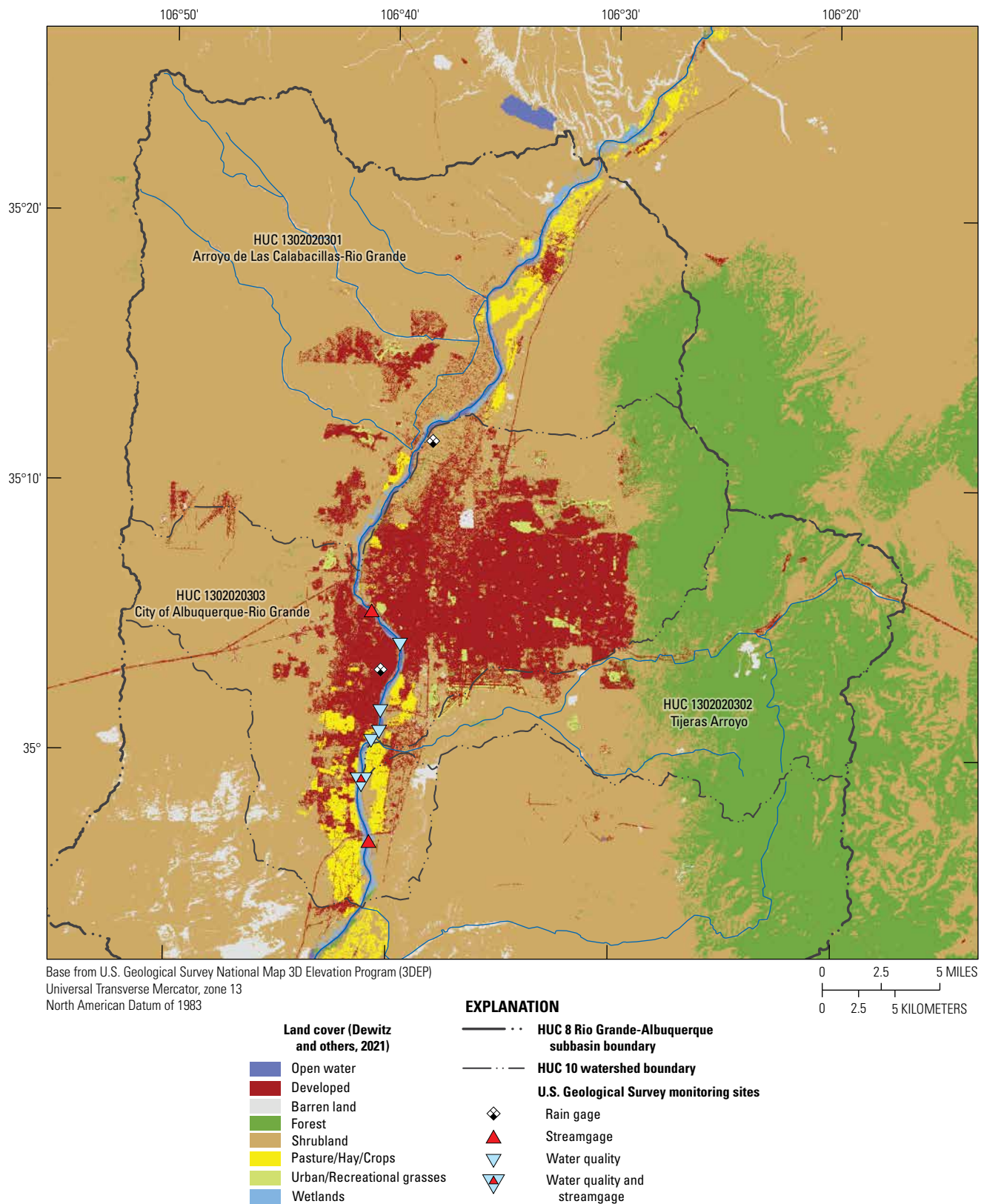
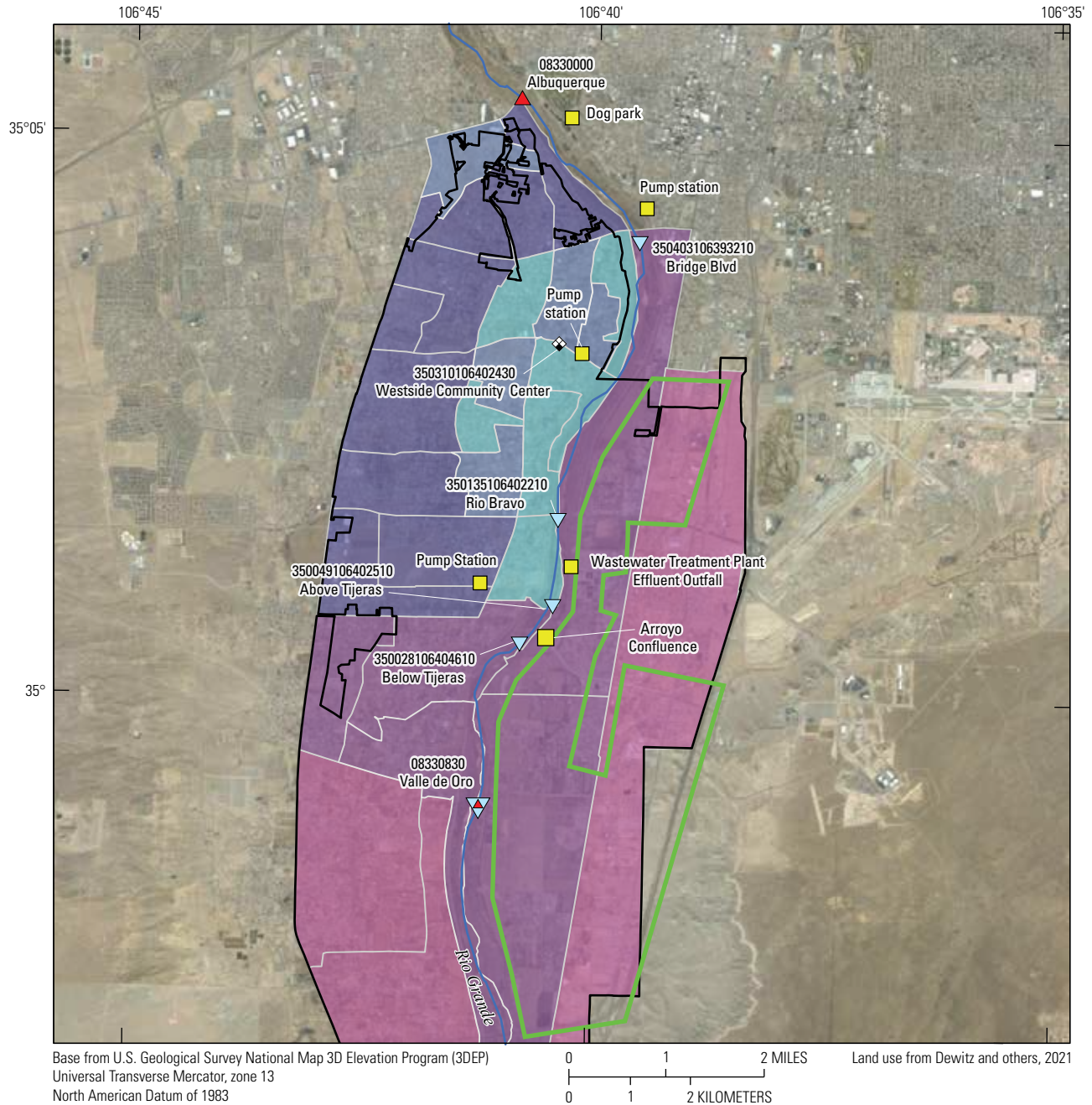


Figure 3. Land cover in the study area in Albuquerque, New Mexico, South Valley neighborhood. HUC, hydrologic unit code.



EXPLANATION

<p>Estimated persons on septic (LaMotte, 2018)</p> <ul style="list-style-type: none"> 0 to 75 76 to 150 151 to 500 501 to 1,500 1,501 to 2,500 <p> Area of limited wastewater infrastructure (M. Haley, Albuquerque Bernalillo County Water Utility Authority, written commun., 2021)</p>	<p> South Valley</p> <p>U.S. Geological Survey monitoring location, identifier, and name</p> <ul style="list-style-type: none"> 350310106402430 Westside Community Center ◆ Rain gage 08330000 Albuquerque ▲ Streamgage 350028106404610 Below Tijeras ▼ Water quality 08330830 Valle de Oro ◄ Water quality Pump station ■ Potential source of <i>Escherichia coli</i>
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Figure 4. Potential sources of *Escherichia coli*, areas of limited wastewater infrastructure, and estimated number of persons on septic systems in the study area in Albuquerque, New Mexico, the South Valley neighborhood.

Bridge Blvd. Three pump stations convey stormwater from channels and storm drainage systems throughout the city and county into the Rio Grande. The pump stations primarily contribute to the Rio Grande during storm events. One pump station is directly upstream from Bridge Blvd and two other pump stations are downstream from Bridge Blvd, one near the Westside Community Center precipitation gage and the other upstream from and to the west of the Above Tijeras site. Both downstream pump stations are operated by the City of Albuquerque (COA). The Rio Grande and its surrounding bosque (a riparian cottonwood forest) are largely designated by the COA as open space and contain a large network of trails for hiking, including dog walking (City of Albuquerque Parks and Recreation Department Open Space Division, 2014). Bernalillo County Public Works has implemented several initiatives to reduce stormwater pollution, including an initiative to reduce dog waste in the watershed (Bernalillo County, 2022) that includes dog waste depositories across the bosque. Waterfowl fecal matter can be a source of *E. coli*, and the Rio Grande and bosque are also a major flyway for migratory birds. Many birds inhabit the area during the winter months (typically November through March) at places like Valle de Oro National Wildlife Refuge, which is located near the southern border of the study area (fig. 2; Sullivan and others, 2009; Herzenberg, 2018).

Hydrology

The Rio Grande-Albuquerque watershed is part of the Rio Grande Rift physiographic province, with mountains forming the eastern and western boundaries. To the west, the mountains are volcanic and to the east, the mountains are uplifted igneous and sedimentary geologic units. The valley floor consists of compacted sands and gravels of the Santa Fe Group, which is Paleogene in age and locally includes Quaternary aged alluvium (Natural Resources Conservation Service, 2002).

The Santa Fe Group aquifer is an unconfined aquifer that underlies the Rio Grande, and the depth to groundwater varies widely throughout the Middle Rio Grande Basin (fig. 1), a geologic basin that contains the study area (Bartolino and Cole, 2002). The Santa Fe Group thickness ranges from 1,400 to 14,000 feet (ft), and the depth to groundwater ranges from less than 2 ft near the Rio Grande, where groundwater and surface water are closely connected (Bartolino and Cole, 2002), to about 1,200 ft in areas west of Albuquerque.

In the Rio Grande-Albuquerque watershed, the Rio Grande is a highly regulated system with the flow controlled by dams and reservoirs upstream and downstream from the study area. Flow in the Rio Grande varies widely throughout the year, with the highest flows typically occurring during spring snowmelt (Bartolino and Cole, 2002). Discharges in 2020 and 2021 were historically low during the dry season and were often statistically below the 25th percentile (548 cubic feet per second [ft^3/s]) of historical flows at USGS 08330000 (Albuquerque) (fig. 5; Jian and others, 2008). Discharges for

53 percent of the days during the dry season were below the 25th percentile from November 1, 2019, through June 30, 2020, and those for 63 percent of days during the dry season were below the 25th percentile from November 1, 2020, through June 30, 2021. When the flow is very low at USGS 08330830 (Valle de Oro), diurnal fluctuations based on the effluent discharge can be observed.

An intricate system of riverside drains, irrigation canals, and community irrigation ditches known as acequias line the Rio Grande. The irrigation canals and ditches are primarily used during irrigation season in places where water from the river is diverted by the Middle Rio Grande Conservancy District (Bartolino and Cole, 2002). The riverside drains parallel both sides of the river and capture lateral groundwater flow derived as seepage loss from the river and irrigation return flow (as shallow groundwater flow), which help keep the river at a stable level.

Within the study area, the main tributaries of the Rio Grande are the WWTP effluent outfall, which was considered a tributary inflow because the water is originally drawn from the Santa Fe aquifer system (fig. 4; Bartolino and Cole, 2002), and Tijeras Arroyo. With its 132 mi^2 drainage area, Tijeras Arroyo is one of the largest arroyos in the Albuquerque area and is intermittent in flow, typically during storms. At streamgage USGS 08330600 Tijeras Arroyo near Albuquerque, N. Mex., discharge was measured during only 40 days in 2021 (USGS, 2022b). Tijeras Arroyo serves as the primary channel for snowmelt and stormwater from areas east of the Sandia Mountains (fig. 1; City of Albuquerque Parks and Recreation Department Open Space Division, 2014). Although not widely reported on, Tijeras Arroyo does contribute a notable amount of sediment into the Rio Grande during storm events. During flash floods between 1937 and 1947 (Copeland, 1995), 26 suspended sediment samples were collected from Tijeras Arroyo. The average suspended sediment concentration for Tijeras Arroyo was 58,000 milligrams per liter (mg/L) at an average discharge of 300 ft^3/s , and the average size class breakdown was 19 percent clay, 69 percent silt, and 12 percent sand.

Climate

The climate in this study area is semiarid, with mostly sunny days and low humidity (Thorn and others, 1993). From water year 2015 through 2021, annual precipitation ranged from 5.19 to 10.13 inches (in.) at the Alameda Pump Station precipitation gage in northern Albuquerque (table 1, fig. 2) and from 4.70 to 10.61 in. at the Westside Community Center precipitation gage in southern Albuquerque (table 1, fig. 2; USGS, 2021). In the Middle Rio Grande Basin, the rainy season is from July through October, with 45–62 percent of annual precipitation falling within those months. Thunderstorms tend to be localized and short lived, making the precipitation spatially and temporally variable throughout the Middle Rio Grande Basin (Bartolino and Cole, 2002).

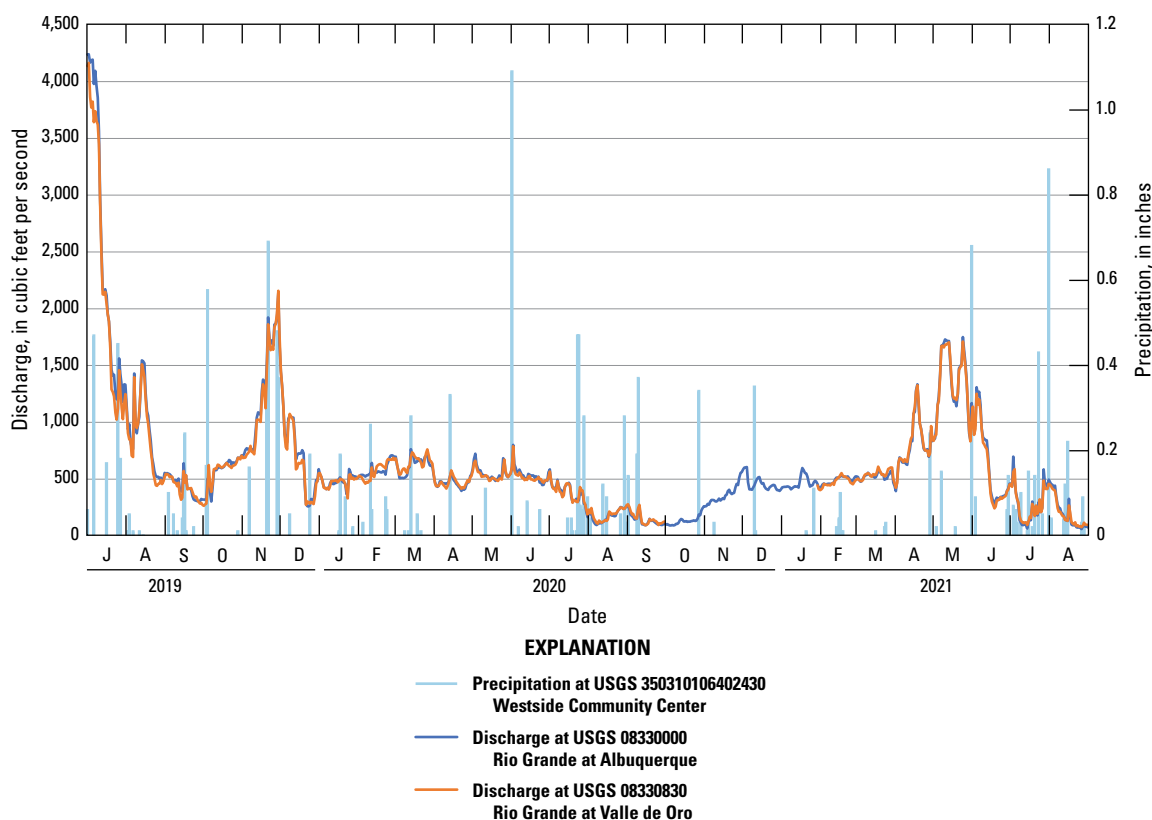


Figure 5. Discharge at U.S. Geological Survey (USGS) 08330000 Rio Grande at Albuquerque and USGS 08330830 Rio Grande at Valle de Oro and precipitation at USGS 350310106402430 Westside Community Center during the duration of the study. Data from USGS (2021).

Previous Monitoring

Since the mid-2000s, several entities, including Bernalillo County, the Albuquerque Metropolitan Arroyo Flood Control Authority, and the COA have monitored *E. coli* and its sources in the study reach and the surrounding watershed. Data associated with these studies that fall within the parameters of the current study, specifically, samples collected from sites in or near the study reach and samples collected during the dry season, are compiled in Travis (2023). A 2005 study utilized ribotypes and antibiotic resistance profiles to determine sources of *E. coli* in the Rio Grande, and the results indicated a variety of sources, with the predominant ones being avian and canine (Parsons Water & Infrastructure, Inc., 2005). In a 2018 study, the COA contracted Daniel B. Stephens and Associates, Inc., to study fecal bacteria and its sources in the Rio Grande as it runs through the city (COA, 2019); genetic markers indicated that avian and human sources were the largest contributors of fecal-origin bacteria to the river during nonstormwater events. *E. coli* was detected in all 12 dry season samples and ranged from 10 to 336 MPN/100 mL, where MPN/100 mL is assumed to be equivalent to cfu/100 mL (Noble and others, 2004). The highest *E. coli* concentration (336 MPN/100 mL) was detected at Rio Grande at the Isleta Pueblo northern boundary (figs. 1 and 2; COA, 2019).

Fluke (2018) and Fluke and others (2019) evaluated *E. coli* levels in water and riverbed sediments in the Rio Grande from Bernalillo, N. Mex., to the Valle de Oro site. During the dry season, *E. coli* concentrations in water ranged from 150 to 1,660 cfu/100 mL at Albuquerque and from 20 to 1,840 cfu/100 mL at the Valle de Oro site. Riverbed sediments were collected at Albuquerque and Rio Bravo and ranged from 0.01 to 249.93 most probable number per 100 grams of dry weight sediment (MPN/100 g_{DW}) and 2.69 to 1,538.7 MPN/100 g_{DW}, respectively, during the dry season (Fluke, 2018, Fluke and others, 2019).

Fluke and others (2019) suggested that the fine-grained bed sediment could be a reservoir for bacteria that could provide suitable habitat for regrowth, resulting in resuspension and higher concentrations of bacteria in this reach. The possibility of regrowth in these fine sediments should be considered when creating best management practices emplaced to reduce the nonpoint sources of bacteria. Sanders and others (2013) suggested that assessment of factors such as interaction with sediment, bacteria regrowth, and sources are important when studying elevated bacteria during baseflow conditions. To study *E. coli* concentrations in a comparable system, Paretti and others (2019) installed piezometers in the hyporheic zone of the Santa Cruz River, Arizona, to sample waters below the stream channel from the middle of the bed to the wetted edge

and found that the *E. coli* concentrations had both a vertical and horizontal gradient that could contribute to mass *E. coli* loading during flooding situations.

Additionally, *E. coli* monitoring has been ongoing in the study reach of the Rio Grande by the Middle Rio Grande Stormwater Quality Team and Isleta Pueblo. The Middle Rio Grande Stormwater Quality Team sampling is conducted by BEMP, and results are available through their website (BEMP, 2021). BEMP sampling followed the New Mexico Environment Department Surface Water Quality Bureau bacteriological sampling standard operating procedure, which suggests that sampling occur from one point in the river that is well mixed and more than 6 in. deep (NMEDSWQB, 2022b). From 2017 through 2020, the BEMP documented *E. coli* concentrations ranging from 10.7 to 3,076 MPN/100 mL during the dry season, with the maximum value of 3,076 collected after a storm event. Isleta Pueblo also conducts sampling, and the results are accessible through the Water Quality Data Portal (National Water Quality Monitoring Council, 2021). The Isleta Water Quality Standards indicate that bacteriological samples are collected at a point where there is adequate vertical and lateral mixing (EPA, 2021a). From 2017 through 2020, Isleta Pueblo documented *E. coli* concentrations ranging from 5.2 to 2,098 MPN/100 mL during the dry season.

Methods

For the duration of the study, water and bed material samples were collected from five sites along the Rio Grande between the USGS gages at Albuquerque (08330000, Rio Grande at Albuquerque), to the northern border of the Isleta Pueblo (08330875, Rio Grande at Isleta Lakes; [fig. 2](#)). Water was analyzed for *E. coli*, MST, and suspended sediment analysis, and bed material was analyzed for *E. coli* and MST markers. All water-quality, sediment, discharge, and precipitation data are publicly available from the USGS National Water Information System (USGS, 2021) using the USGS site numbers in [table 1](#).

Field Methods

Discrete surface-water samples, bed material samples, and field parameters were collected from five sites along the Rio Grande between April 2020 and August 2021 ([table 1](#)). Within the river cross section at each site, samples were collected from within 5 ft of the east bank, river center, and within 5 ft of the west bank to determine if there was heterogeneity within the sampling site. These east, center, and west sampling locations will collectively be referred to as “three-point sampling” at each site. This sampling method is atypical of the suggested bacteriological sampling method specified by the New Mexico Environment Department Surface Water Quality Bureau Standard Operating Procedure 9.1, where compliance sampling typically occurs at only one well-mixed

location within the stream (NMEDSWQB, 2022b). Twelve sampling events occurred at each of the sites, six events in 2020 and six events in 2021. Four sampling events each year occurred during the dry season and two sampling events each year either occurred during the wet season or during the dry season at a discharge of greater than 1,000 ft³/s.

Dry season sampling criteria for this study have been set by evaluating (1) past sampling conditions in relation to precipitation and discharge and (2) the National Pollutant Discharge Permit for the WWTP (NMEDSWQB, 2019c). The dry season, as defined by the permit, is November 1–June 30; wet season sampling, which occurs after a rain event during the time period of July 1–October 31, is defined as occurring when rainfall is greater than 0.25 in., and an antecedent dry period of at least 48 hours after a rain event greater than 0.1 in. in magnitude (NMEDSWQB, 2019c). Given the criteria of the permit for the dry season and the peaks in flow in the Rio Grande after snowmelt over the past few years, dry season sampling occurred when the following criteria were met within the greater Albuquerque area:

- sampling event is between November 1 and June 30,
- at least 48 hours after a rain event of less than 0.25 in., and
- Rio Grande discharge is less than 1,000 ft³/s.

USGS precipitation and streamgages within the study area were used to monitor the streamflow and rainfall sampling criteria.

Three-point sampling surface-water samples for *E. coli*, MST markers, and suspended sediment concentration were collected using the discrete or grab method. The USGS National Field Manual states that the isokinetic depth integrated sampling is the standard procedure and typically is the preferred method if the cross section is uneven in velocity and (or) depth (USGS, variously dated). Additionally, isokinetic samples are composited into a churn that is not able to be autoclaved, so bacteria samples are traditionally collected as a grab sample. However, Paretto and others (2018) directly compared samples collected using the isokinetic depth integrated sampling technique to grab samples and found similar *E. coli* and suspended sediment concentrations using both methods. Therefore, this study used the grab sample technique.

Water and bed material samples for *E. coli* and MST markers were collected by following the USGS protocols described in Myers and others (2014). Before going into the field for sample collection, bottle kits containing the appropriate sterilized 1-liter (L) wide-mouth polyethylene bottles and sediment jars were assembled. At a minimum, all sampling equipment was soaked in phosphate free detergent for 30 minutes and rinsed with tap water followed by deionized water (USGS, 2004). After the initial cleaning step, the *E. coli* sampling bottles were autoclaved, and the MST marker sampling bottles and jars were sterilized with a sodium hypochlorite solution, neutralized with a sodium thiosulfate solution, and rinsed with sterile deionized water (Myers and others, 2014).

To prevent contamination, the least amount of equipment was used to collect a sample, and sample collection followed the USGS National Field Manual for bacteria sample collection (USGS, variously dated). If the sites were safely wadable, the sampling bottle was submerged to allow the bottle to fill with the opening pointed slightly upward toward the current. If the sites were not wadable, the samples were collected either from a bridge (Bridge Blvd and Rio Bravo) using a DH-95 sampler and a bridge board, or, at the sites with no accessible bridge (Above Tijeras, Below Tijeras, and Valle de Oro), the grab samples were collected from a kayak. All water samples were collected with about 2.5–5 centimeters of headspace in each bottle to allow for proper mixing. The *E. coli* and MST marker samples were bagged immediately to prevent contamination and stored on ice in a cooler until analysis.

Bed sediment samples were analyzed for *E. coli* and MST markers at the Ohio Water Microbiology Laboratory (OWML) and for fine sand break at the New Mexico Water Science Center (NMWSC) Sediment Laboratory. For the *E. coli* and MST bed sediment collection, three sterile, plastic, wide-mouth 125-milliliter (mL) jars were used at each sampling location. For the fine sand break analysis, a 16-ounce plastic jar was used. The lidded sampling jar was lowered to the river bottom where the lid was removed to scoop bed sediment into the jar. The lid was then secured tightly on the jar before bringing the jar to surface. The jars were bagged immediately to prevent contamination and stored on ice in a cooler until analysis. When river stage was too high to easily sample by hand, a tool with a hose clamp attached to a long handle was used to hold the sample jar and scoop bed sediment from the river bottom.

Four known-source fecal samples were collected from fresh material and of known origin. Solid samples were collected for geese (n=2) and canine (n=1) sources. One human source, a wastewater influent sample, was collected from the WWTP. All known-source fecal samples were stored on ice immediately after collection and shipped to the OWML for processing within 24 hours. Known-source samples were only collected once throughout the study.

Physical parameters, including water temperature, pH, specific conductance, dissolved oxygen, turbidity, and barometric pressure, were recorded at each three-point sampling location within each site using either a Yellow Springs Incorporated 6920 series or 600 series multiparameter water-quality sonde (YSI Incorporated, Yellow Springs, Ohio) or an In-Situ Aqua TROLL 600 (In-Situ Inc., Fort Collins, Colorado). Following the manufacturer instructions and USGS methods, a calibration check was performed on the sonde each morning before collecting samples (USGS, variously dated). Field parameters, including turbidity, dissolved oxygen, specific conductance, pH, and water temperature, were measured by USGS staff. Two USGS streamgages, Albuquerque and Valle de Oro, provided continuous discharge throughout the study (table 1). Daily mean discharge from Albuquerque was used for the Bridge Blvd and Rio Bravo sampling sites. Daily mean discharge from Valle de Oro was used for the Above

Tijeras and Below Tijeras sampling sites, as it reflects the daily effluent outfall fluctuations from the WWTP, which is upstream from Above Tijeras. At Valle de Oro, discharge was measured during every sampling date to check the continuous streamgage at the site. Table 2 shows the discharge measurements associated with each site for each sampling event. Discharge measurements were collected using standard USGS protocols as described in Rantz and others (1982), Nolan and Shields (2000), and Turnipseed and Sauer (2010).

Analytical Methods

The analytical methods for *E. coli* and MST in both water and bed material and suspended sediment analysis in water are discussed in this section.

Escherichia coli in Water

Water samples were analyzed for *E. coli* by USGS staff within 6 hours of sample collection using the colilert method (Standard Methods Committee of the American Public Health Association, American Water Works Association, and Water Environment Federation, 2022; USGS, variously dated). The 1-L samples collected for *E. coli* were shaken prior to pouring 100 mL of sample into a sterile graduated cylinder. From the cylinder, the sample was immediately poured into a sterilized 100-mL bottle, and a reagent packet was poured into that bottle. The bottle was then shaken again until all the reagent was dissolved, and the sample was poured into a Quanti-Tray and sealed with an IDEXX Quanti-Tray Sealer PLUS (IDEXX Laboratories, Inc., Westbrook, Maine) and incubated at 35 degrees Celsius (°C) for 24–28 hours. After the incubation period, the *E. coli* density was calculated by counting the number of small and large wells whose water sample reacted with the reagent on the Quanti-Tray under a long-wave ultraviolet light at 365 nanometers. The colilert method reported results as MPN/100 mL, which has been evaluated to have a 1:1 relationship to results reported in cfu/100 mL (Noble and others, 2004).

Microbial Source Tracking Markers in Water

Samples were analyzed for MST markers at the OWML. Within 24 hours of sample collection, varying matrix-dependent volumes of water samples were filtered onto 0.4-micrometer polycarbonate filters (Whatman, Inc., Florham Park, New Jersey). The filters were aseptically placed into 2-mL screw cap vials containing 0.3 grams (g) of sterile glass beads (Sigma-Aldrich Corp., St. Louis, Missouri) and preserved at –70 °C. Filtered samples underwent deoxyribonucleic acid (DNA) extraction by use of the DNA-EZ extraction kit (GeneRite, Monmouth Junction, N.J.) and extracts were stored at 4 °C until analysis by quantitative polymerase chain reaction (qPCR) within 7 days of extraction. qPCR was analyzed using either a StepOnePlus or a QuantStudio 3

Table 2. Daily mean discharge values used for all events at five sites on the Rio Grande in Albuquerque, New Mexico.

[Data from U.S. Geological Survey (USGS, 2021). Dates shown as month/day/year. Sample times shown in 24-hour format. All sample times are local. ft³/s, cubic foot per second; Blvd, boulevard]

Short name	Sample date	Sample time	Daily mean discharge (ft³/s)	USGS site number
Bridge Blvd	4/7/2020	1040	465.7	08330000
Bridge Blvd	4/27/2020	1040	467.7	08330000
Bridge Blvd	5/13/2020	0930	520.9	08330000
Bridge Blvd	6/10/2020	0910	532.5	08330000
Bridge Blvd	7/23/2020	0900	303.9	08330000
Bridge Blvd	8/31/2020	0900	266.6	08330000
Bridge Blvd	2/8/2021	0900	458.1	08330000
Bridge Blvd	3/3/2021	0930	491.0	08330000
Bridge Blvd	3/10/2021	0900	556.2	08330000
Bridge Blvd	5/18/2021	0800	1,188.0	08330000
Bridge Blvd	6/23/2021	0900	343.4	08330000
Bridge Blvd	8/16/2021	1040	326.2	08330000
Rio Bravo	4/7/2020	1250	465.7	08330000
Rio Bravo	4/27/2020	1250	467.7	08330000
Rio Bravo	5/13/2020	1130	520.9	08330000
Rio Bravo	6/10/2020	1040	532.5	08330000
Rio Bravo	7/23/2020	1000	303.9	08330000
Rio Bravo	8/31/2020	1000	266.6	08330000
Rio Bravo	2/8/2021	1100	458.1	08330000
Rio Bravo	3/3/2021	1100	491.0	08330000
Rio Bravo	3/10/2021	1030	556.2	08330000
Rio Bravo	5/18/2021	1000	1,188.0	08330000
Rio Bravo	6/23/2021	1000	343.4	08330000
Rio Bravo	8/16/2021	1140	326.2	08330000
Above Tijeras	4/7/2020	1000	459.5	08330830
Above Tijeras	4/27/2020	1000	490.4	08330830
Above Tijeras	5/13/2020	1000	515.3	08330830
Above Tijeras	6/10/2020	0940	510.8	08330830
Above Tijeras	7/23/2020	0900	297.0	08330830
Above Tijeras	8/31/2020	1100	273.4	08330830
Above Tijeras	2/8/2021	1200	444.2	08330830
Above Tijeras	3/3/2021	1200	492.5	08330830
Above Tijeras	3/10/2021	1200	552.1	08330830
Above Tijeras	5/18/2021	0930	1,215.3	08330830
Above Tijeras	6/23/2021	1100	340.7	08330830
Above Tijeras	8/16/2021	1030	267.3	08330830
Below Tijeras	4/7/2020	1300	459.5	08330830
Below Tijeras	4/27/2020	1200	490.4	08330830
Below Tijeras	5/13/2020	1100	515.3	08330830
Below Tijeras	6/10/2020	1120	510.8	08330830

Table 2. Daily mean discharge values used for all events at five sites on the Rio Grande in Albuquerque, New Mexico.—Continued

[Data from U.S. Geological Survey (USGS, 2021). Dates shown as month/day/year. Sample times shown in 24-hour format. All sample times are local. ft³/s, cubic foot per second; Blvd, boulevard]

Short name	Sample date	Sample time	Daily mean discharge (ft ³ /s)	USGS site number
Below Tijeras	7/23/2020	1000	297.0	08330830
Below Tijeras	8/31/2020	0910	273.4	08330830
Below Tijeras	2/8/2021	0950	444.2	08330830
Below Tijeras	3/3/2021	0920	492.5	08330830
Below Tijeras	3/10/2021	0900	552.1	08330830
Below Tijeras	5/18/2021	1100	1,215.3	08330830
Below Tijeras	6/23/2021	0840	340.7	08330830
Below Tijeras	8/16/2021	1100	267.3	08330830
Valle de Oro	4/7/2020	1400	459.5	08330830
Valle de Oro	4/27/2020	1400	490.4	08330830
Valle de Oro	5/13/2020	1300	515.3	08330830
Valle de Oro	6/10/2020	1310	510.8	08330830
Valle de Oro	7/23/2020	1120	297.0	08330830
Valle de Oro	8/31/2020	1020	273.4	08330830
Valle de Oro	2/8/2021	1130	444.2	08330830
Valle de Oro	3/3/2021	1100	492.5	08330830
Valle de Oro	3/10/2021	1010	552.1	08330830
Valle de Oro	5/18/2021	1210	1,215.3	08330830
Valle de Oro	6/23/2021	1010	340.7	08330830
Valle de Oro	8/16/2021	1130	267.3	08330830

Real-Time PCR System (Thermo Fisher Scientific, Waltham, Massachusetts) on the basis of conditions reported in the references for the following MST markers:

- human-associated *Bacteroides* marker (human marker, HF183/BacR287) (Green and others, 2014),
- canine-associated *Bacteroides* marker (canine marker, BacCan) (Kildare and others, 2007), and
- waterfowl-associated *Helicobacter* marker (waterfowl marker, GFD) (Green and others, 2012).

MST Quality Control

Quality control analyses and procedures are vital to the production of reliable microbial source tracking data. In addition to field blanks collected onsite, several types of blank samples were created during the analytical process:

- filter blank—sterile, buffered water filtered at the same time as the samples at the OWML,

- extraction blank—sterile vial containing only acid-washed beads was DNA-extracted along with each batch of samples, and

- qPCR blanks (no template controls)—sterile, molecular-grade water was used instead of sample DNA extract as qPCR template.

All samples were analyzed by qPCR in duplicate; final results represent an average of duplicate qPCR reactions. Potential matrix effects on the qPCR were evaluated by analyzing a portion of each sample that was spiked with a known quantity of target DNA. If matrix inhibition was observed, sample DNA extracts were diluted accordingly and then processed by qPCR.

Each qPCR run included a seven-point standard curve run in duplicate (serial dilutions of known quantities of target DNA). MST marker concentrations for unknown samples were interpolated from the standard curves by converting qPCR output (cycle threshold values).

MST Data Qualifications

A limit of detection was established for each MST qPCR assay. The limit of detection was the lowest concentration detected with 95-percent confidence. If detections were found in the blank samples, the limit of blanks was calculated. If the limit of blanks had a higher concentration than the limit of detection, the limit of blanks was used as the detection limit. Results for samples that were below either limit were reported as less than (<) the detection limit.

Low-level quantification can be unreliable. To avoid misinterpretation of unreliable results, a limit of quantification was established for each assay. The limit of quantification was established by using the average cycle threshold value of the limit of detection and subtracting two times the standard deviation of those cycle threshold values. The result for a sample that detected the target DNA and was above the limit of detection but below the limit of quantification was reported with an “E” to denote that it was an estimated value. Along with the “E,” there may also be a value qualifier. The “b” value qualifier meant that the reported concentration was extrapolated below the limit of quantification but is above the limit of detection. The “~” value qualifier meant that the duplicate qPCR results do not agree.

Escherichia coli Bacteria and Microbial Source Tracking Markers (Human-, Canine-, and Waterfowl-Specific Sources) in Bed Material

River-bottom sediment samples were analyzed for concentrations of *E. coli* bacteria and MST markers (human-, canine-, and waterfowl-specific sources) at the OWML by the methods described in the section “Microbial Source Tracking Markers in Water.” Processing steps for sediment samples (Francy and Darner, 1998) were required before analysis. Briefly, a 30-g aliquot of composited sediment from the three subsamples was placed into a bottle containing 300 mL of sterile phosphate buffer; a second aliquot of composited sediment was reserved to determine percent dry weight. The bottle was placed on a wrist-action shaker for 45 minutes, then removed; suspended materials were allowed to settle for 30 seconds, and the liquid phase was decanted for analysis (filtration for MST marker analysis and analysis by colilert). After *E. coli* and MST analyses were completed, calculations were made to convert MST marker (copies) and *E. coli* (MPN) results to copies or MPN/g_{DW}.

Suspended Sediment in Water

Suspended sediment concentration in the water column was measured by the USGS NMWSC Sediment Laboratory following the Quality-Assurance Plan for the Analysis of Fluvial Sediment (Stiles, 2006). Once received, samples were stored in a cool, dark location, allowed to settle for 2 weeks, and analyzed within 60 days. Depending on the amount of

sand or clay in the sample, either the filtration method or the evaporation method described in ASTM D3977–97 (American Society for Testing and Materials, 2019) was used for analysis.

If the evaporation method was used, the sample was washed in an evaporation dish, dried, and weighed. If the dissolved-solids concentration was greater than 2,300 mg/L and the concentration of sediment was less than 200 mg/L, then a dissolved-solids correction needed to be applied. If the filtration method was used, the sample was poured into a crucible, vacuum pressured, and forced through a filter. The crucible was then dried, cooled, and weighed. No mathematical adjustments were needed if the filtration method was used. For both the evaporation and filtration method, concentrations were calculated using the Sediment Laboratory Environmental Data System (Stiles, 2006).

Particle Size in Bed Material

Sand-fine analysis was performed on bed material samples at the NMWSC Sediment Laboratory following the Quality-Assurance Plan for the Analysis of Fluvial Sediment by the NMWSC Sediment Laboratory (Stiles, 2006). The fine analysis refers to the percentage of particles that pass through a 0.062-millimeter (mm) sieve, and the sand analysis refers to the percentage of particles remaining on that sieve. Following the recommendations of Guy (1969) and Tyler Industrial Products (1976), the dry sieve method was used for this analysis with a W.S. Tyler Ro-Tap machine. First, the sand and gravel were separated from the silt-clay fraction by wet sieving. Next, the sand and gravel were dried and weighed. The sample was shaken in the W.S. Tyler Ro-Tap machine for 20 minutes, and then the size-fraction weights that remained in each sieve were entered into the Sediment Laboratory Environmental Data System (Stiles, 2006). If the sample weighed more than 400 g, the sample was split into two samples for analysis on the Ro-Tap machine.

Data Analysis Methods

The replicate pair variability was analyzed in different ways depending on the analyte. In describing the “two-range model,” Mueller and others (2015) state that over low concentrations, standard deviation (SD) is generally uniform, but at higher concentrations, relative standard deviation (RSD) is more uniform than SD. Paretti and others (2018) reported extensively on quality control and assurance methods for bacteria sampling methods and with a large amount of *E. coli* replicates, were able to analyze the variability in sampling methods using the bias-corrected log-log regression model (Mueller and others, 2015). Because of the lower number of replicate pairs and relatively higher concentrations for all parameters, RSD was used instead of SD to study variability for all analytes,

$$\text{RSD} = 100 \times \left(\frac{\text{Standard Deviation } (C_{\text{environmental}}, C_{\text{replicate}})}{\text{Mean } (C_{\text{environmental}}, C_{\text{replicate}})} \right) \quad (1)$$

where

$C_{\text{environmental}}$ is the environmental sample concentration, and
 $C_{\text{replicate}}$ is the replicate sample concentration.

For *E. coli* and suspended sediment, which had some lower concentrations, SD was also calculated. Results for many analytes were censored (that is, less than the reporting level) in one or both replicate samples, which excluded them from replicate analysis.

Instantaneous or mean daily loading rates, also known as flux, for *E. coli* and MST markers were calculated using a simple approach (Meals and others, 2013) of multiplying concentration times discharge and were reported in units of MPN or copies per day,

$$\text{Flux} = K \times Q \times C \quad (2)$$

where

K is the unit conversion factor (2.45×10^7 when calculating mean daily flux in most probable number per day [MPN/day] or copies per day [copies/day], which accounts for conversions of cubic feet to milliliters [1 cubic foot equals 28,316.8 mL] and seconds to days [86,400 seconds equal 1 day]),

Q is discharge (in cubic feet per second), and
 C is concentration (MPN or copies per 100 mL).

The mean daily flux is the instantaneous rate at which the load passes a point in the river. This flux is not equal to the load, which is the mass of a substance passing through a specific point in the river. The load is calculated over a specified timeframe with frequent monitoring. Because of the infrequency of the sampling and targeting of dry season, lower-flow events, accurate loads cannot be calculated.

To categorically compare fluxes of different microbiological constituents at a single site and across multiple sites, the mean daily fluxes were divided into five bins (Bushon and others, 2017). For each constituent, regardless of flow conditions, all results were combined, ranked, and binned. All nondetects were assigned to the 0 bin. Bins 1–4 contained an equal number of sample results per bin, with the highest fluxes placed in bin 4. Table 3 lists the ranges of daily fluxes per bin for each constituent. Once the bins were established, each sample result for a given constituent was assigned a bin number that was then separately averaged for each site for dry and wet season samples. Average bin scores can be used to compare the fluxes of a single constituent across sites and to compare the fluxes of different constituents at a single site.

Statistical analyses were conducted in the R statistical computing environment (R Core Team, 2022). Because a high percentage of MST marker data were censored values, summary statistics were calculated for censored data using the “censtats” function in the Nondetects and Data Analysis (NADA) software package, and the summary statistics using

Table 3. Range of mean daily fluxes per bin for *Escherichia coli* and microbial source tracking markers.

Constituent	Flux		
	Bin	Minimum	Maximum
<i>Escherichia coli</i> (billion most probable number)	1	75	580
	2	581	980
	3	981	1,761
	4	1,762	93,000
Human marker (billion copies)	1	9,200	59,000
	2	63,000	185,000
	3	186,000	543,000
	4	544,000	4,200,000
Canine marker (billion copies)	1	15,000	52,000
	2	58,000	92,000
	3	99,000	160,000
	4	161,000	340,000
Waterfowl marker (billion copies)	1	22,000	38,000
	2	46,000	115,000
	3	116,000	190,000
	4	200,000	550,000

the regression on order statistics method were reported for this study. Boxplots were created by using the “cenboxplot” function from the NADA software package (Lee, 2020) in the R statistical computing environment (R Core Team, 2022). Boxplots for analytes with censored data were grouped by sample type; percentiles were estimated by using the robust regression on order statistics method (Helsel, 2012). Outlier data points on boxplots were defined for this study as greater than 1.5 times the interquartile range. To evaluate correlations, Pearson’s r and Kendall’s tau were calculated for uncensored data, and only tau was calculated for censored data. All *E. coli* concentration data were log₁₀ transformed before conducting the Pearson’s r calculations because the bacteria data were not normally distributed. Strong linear correlations of $r = 0.9$ or above typically correspond to tau values of about 0.7 or above (Helsel and others, 2020). For uncensored data, the base R function “cor.test” was used to determine correlations using both Pearson’s r and Kendall’s tau, and for censored data the “cenken” function was used from the NADA package to determine Kendall’s tau (Lee, 2020). When evaluating differences between the datasets, Kruskal-Wallis rank sum test was used for uncensored data using base R function “kruskal.test,” and when a probability value (p-value) was less than 0.05, the differences were found to be significant. If significant differences were found, then to determine if three-point sampling locations within sites differed from others for the uncensored data, Dunn’s test using the Benjamini & Hochberg correction method was used using the R package “PMCMRplus” and its “kwAllPairsDunnTest” function (Pohlert, 2021). To evaluate differences between three-point sampling locations within sites for uncensored data, the “cen1way” function from the NADA2 package was used (Julian and Helsel, 2021), which performs the Peto-Peto nonparametric test of differences.

Data-Quality Assurance and Assessment

Quality control and assurance samples were collected in the field to ensure the field sample collection methods and field environment did not introduce contamination into the sampling process and to determine the variability in the sample collection process and laboratory methods.

Field blanks were collected at each a field site by pouring sterile phosphate buffer water with magnesium chloride directly into a sterilized 1-L poly sample collection bottle. The field blanks were analyzed to assess the equipment and field activities for any contamination potentially introduced into the samples during the cleaning and sampling process. Six field

blanks were collected and analyzed for *E. coli* and MST markers. One source solution blank was analyzed for *E. coli* and was collected in the laboratory by directly pouring the buffer water into the sterilized graduated cylinders used for measuring out the sample for processing. All field and source blanks had no detections of *E. coli* or MST markers.

Six replicate water samples and three replicate bed material samples (table 4) were collected sequentially. Sequential replicates assessed the variability of samples resulting from field activities as well as laboratory analysis. Five replicate water samples from the east bank of Above Tijeras were collected on April 27, 2020, June 10, 2020, July 23, 2020, March 3, 2021, and June 23, 2021, and analyzed for *E. coli*, MST markers, and suspended sediment. A water replicate from the east bank location at Bridge Blvd was collected on May 18, 2021, and analyzed for *E. coli* and suspended sediment only. Bed material replicate samples from the east bank location at Above Tijeras were collected on June 10, 2020, March 3, 2021, and June 23, 2021, and analyzed for *E. coli* and MST markers.

On the basis of the two-range model, low-range SDs were calculated for *E. coli* at concentrations below 150 MPN/100 mL and for suspended sediment concentrations below 100 mg/L. High-range RSDs were calculated for all other analytes. For the replicate water sample results, the average RSDs were less than 20 percent for most analytes, except for the human marker (44.1 percent) and the canine marker (28.3 percent). The average SD was 3.3 MPN/100 mL for low-range *E. coli* replicates and 10.1 mg/L for low-range suspended sediment concentration replicates. Bed material samples were mostly below the reporting level, but for the *E. coli* and human marker replicate pairs where RSDs could be calculated, the averages were 15.9 MPN/g_{DW} SD for *E. coli* replicates and 24.5 percent RSD for the human marker.

Known-source fecal samples were collected and analyzed to verify the analytical performance of the MST markers used in the study. Analysis of all known source samples resulted in detections of the associated MST marker (table 5). Analysis of wastewater influent from the WWTP resulted in detection of the canine marker in addition to the human marker, and the analysis of the waterfowl marker also included detection of the canine marker. This was not unexpected because fecal bacteria may be transferred between host species living in close contact, resulting in co-occurrence of host-associated MST markers in a sample (Field and Samadpour, 2007; Stewart and others, 2007; Roslev and Bukh, 2011). Concentrations of MST markers in cross-reacting samples were lower than in target samples except in the case of the second Canada goose sample, as shown in table 5.

Table 4. Replicate samples and their calculated standard deviations and relative standard deviations for seven samples collected on the Rio Grande in the Albuquerque, New Mexico, area during 2020–21.

[Dates shown as month/day/year. SD, standard deviation; RSD, relative standard deviation; *E. coli*, *Escherichia coli*; MPN/100 mL, most probable number per 100 milliliters; NC, not calculated; mg/L, milligram per liter; copies/100 mL, copies per 100 milliliters; E, estimated value; <, less than; --, no value; MPN/g_{DW}, most probable number per gram of dry weight sediment; copies/g_{DW}, copies per gram of dry weight sediment; Blvd, boulevard]

Descriptor	Sample medium								
	Water					Bed material			
	Analyte					Analyte			
	<i>E. coli</i> (MPN/100 mL)	Suspended sediment concentration (mg/L)	Human marker (copies/ 100 mL)	Canine marker (copies/ 100 mL)	Waterfowl marker (copies/ 100 mL)	<i>E. coli</i> (MPN/g _{DW})	Human marker (copies g _{DW})	Canine marker (copies/ g _{DW})	Waterfowl marker (copies/ g _{DW})
Above Tijeras, East Bank, 4/27/2020									
Environmental	160	120	200,000	E6,100	<1,800	NR	NR	NR	NR
Replicate	170	119	220,000	11,000	<2,200	NR	NR	NR	NR
SD	NC	NC	NC	NC	--	NR	NR	NR	NR
RSD	4.3	0.6	6.7	40.5	--	NR	NR	NR	NR
Above Tijeras, East Bank, 6/10/2020									
Environmental	160	49	26,000	<3,100	<11,000	59	E560	<660	<2,300
Replicate	150	55	46,000	<3,100	<2,200	E14	E710	<660	<450
SD	NC	4.2	NC	--	--	31.8	NC	--	--
RSD	4.6	NC	39.3	--	--	NC	16.7	--	--
Above Tijeras, East Bank, 7/23/2020									
Environmental	390	107	E21,000	<11,000	<1,500	NR	NR	NR	NR
Replicate	390	113	47,000	<4,500	<1,500	NR	NR	NR	NR
SD	NC	NC	NC	--	--	NR	NR	NR	NR
RSD	0	3.9	54.1	--	--	NR	NR	NR	NR
Above Tijeras, East Bank, 3/3/2021									
Environmental	60	59	160,000	15,000	E3,600	<1	E610	<600	<290
Replicate	60	93	150,000	13,000	E3,000	0.67	920	<170	<81
SD	0	24	NC	NC	NC	--	NC	--	--
RSD	NC	NC	4.6	10.1	12.9	--	28.7	--	--
Bridge Blvd, East Bank, 5/18/21									
Environmental	34	156	--	--	--	NR	NR	NR	NR
Replicate	26	165	--	--	--	NR	NR	NR	NR
SD	6.3	NC	--	--	--	NR	NR	NR	NR

Table 4. Replicate samples and their calculated standard deviations and relative standard deviations for seven samples collected on the Rio Grande in the Albuquerque, New Mexico, area during 2020–21.—Continued

[Dates shown as month/day/year. SD, standard deviation; RSD, relative standard deviation; *E. coli*, *Escherichia coli*; MPN/100 mL, most probable number per 100 milliliters; NC, not calculated; mg/L, milligram per liter; copies/100 mL, copies per 100 milliliters; E, estimated value; <, less than; --, no value; MPN/g_{DW}, most probable number per gram of dry weight sediment; copies/g_{DW}, copies per gram of dry weight sediment; Blvd, boulevard]

Descriptor	Sample medium								
	Water					Bed material			
	Analyte					Analyte			
	<i>E. coli</i> (MPN/100 mL)	Suspended sediment concentration (mg/L)	Human marker (copies/ 100 mL)	Canine marker (copies/ 100 mL)	Waterfowl marker (copies/ 100 mL)	<i>E. coli</i> (MPN/g _{DW})	Human marker (copies g _{DW})	Canine marker (copies/ g _{DW})	Waterfowl marker (copies/ g _{DW})
Bridge Blvd, East Bank, 5/18/21—Continued									
RSD	NC	4	--	--	--	NR	NR	NR	NR
Above Tijeras, East Bank, 6/23/2021									
Environmental	240	71	E2,700	E4,600	<630	4.1	E240	<190	<93
Replicate	180	74	27,000	E2,800	<630	4.1	E160	<180	<88
SD	NC	2.1	NC	NC	--	0	NC	--	--
RSD	19	NC	115.7	34.4	--	NC	28.3	--	--

Table 5. Reported values of microbial source tracking (MST) markers from known-source samples collected near Albuquerque, New Mexico, for MST.

[Dates shown as month/day/year. Values in bold typeface indicate samples where the concentration was above the limit of detection (LOD). The LOD for a given sample is represented by less than (<) a concentration when a nondetect occurred. The LOD varies per assay and is defined as the lowest concentration detected with 95-percent confidence]

Sample date	Known source	Human marker	Canine marker	Waterfowl marker
Copies per gram dry weight				
3/3/2021	Canada goose	<62,000	<110,000	17,000,000
3/3/2021	Canada goose	<33,000	1,200,000	540,000
5/18/2021	Dog	<34,000	2,000,000,000,000	<29,000
Copies per 100 milliliters				
12/16/2020	Southside Water Reclamation Plant	480,000,000	34,000,000	<22,000

Characterization of *Escherichia coli* Microbial Source Tracking Markers and Other Parameters in Water and Bed Material

The following discussion provides an overview of the findings in water and bed material. Physical parameters, suspended sediment concentrations, *E. coli*, and MST markers were evaluated and summarized. *E. coli* concentrations in water were compared to water-quality standards. The fluxes for *E. coli* and MST markers in water were calculated for all sites and locations. Bed material *E. coli* and MST marker concentrations were also evaluated, and particle size was reviewed to determine if there was fine material providing an opportunity for bacteria regrowth. Water and bed material data were then evaluated to determine if there were any temporal (dry versus wet season) and spatial (sample site and location) differences. Lastly, MST marker concentrations were ranked to determine the dominant sources of fecal bacteria at the different sites.

Physical Parameters and Suspended Sediment Characterization in Water

Physical parameters, including pH, specific conductance, dissolved oxygen, and turbidity, were evaluated at each sample site and three-point sampling location (fig. 6). There was minimal fluctuation in values between each of the three-point sampling locations at the Bridge Blvd and Rio Bravo sites but also between the Bridge Blvd and Rio Bravo sampling groups. As expected, the effluent outfall affected several of the physical parameters after it entered from the east bank upstream from the Above Tijeras site and continued downstream to the Valle de Oro site. Specific conductance increased from a median of

333 microsiemens per centimeter at 25 degrees Celsius ($\mu\text{S}/\text{cm}$ at 25 °C) at the west bank to 587 $\mu\text{S}/\text{cm}$ at the east bank at Above Tijeras, whereas the median ranged from 331 to 334 $\mu\text{S}/\text{cm}$ at Bridge Blvd and Rio Bravo upstream from the effluent outfall. pH decreased at the east bank at Above Tijeras to a median of 7.5 as compared to the median from the upstream sites at Bridge Blvd and Rio Bravo, which ranged from 8.2 to 8.3. Median dissolved oxygen concentrations ranged from 8.1 to 8.3 mg/L upstream at Bridge Blvd and Rio Bravo versus the east bank of Above Tijeras where the median dissolved oxygen concentration was 7.8 mg/L. The median turbidity ranged from 53 to 64 formazin nephelometric units (FNU) at Bridge Blvd and Rio Bravo, and the median turbidity at east bank at Above Tijeras was 39 FNU. The influence of the effluent on the east bank samples continued to be visible at the downstream sites Below Tijeras and Valle de Oro, with medians of 49 and 75 FNU, respectively, and lower than the other three-point sampling locations at those sites.

Suspended sediment concentrations (fig. 7A) were similar at Bridge Blvd and Rio Bravo and at the west and center locations at Above Tijeras, with median values ranging from 138 to 177 mg/L. At the east bank at Above Tijeras, suspended sediment concentrations decreased to a median value of 103 mg/L. Suspended sediment concentrations increased along the west bank at Below Tijeras, with a median of 185 mg/L and a maximum value of 2,990 mg/L. Valle de Oro suspended sediment concentrations showed more consistent mixing in the cross section, ranging from median values of 136 to 198 mg/L.

Escherichia coli Characterization in Water

E. coli concentrations vary between the dry season and the rest of the year. Table 6 contains summary statistics for *E. coli*, including mean, median, maximum, and minimum values of all the data and only the dry season data. Figure 8A–E shows time series plots of *E. coli* and MST marker

20 Assessing *E. coli* and Microbial Source Tracking Markers in the Rio Grande in the South Valley

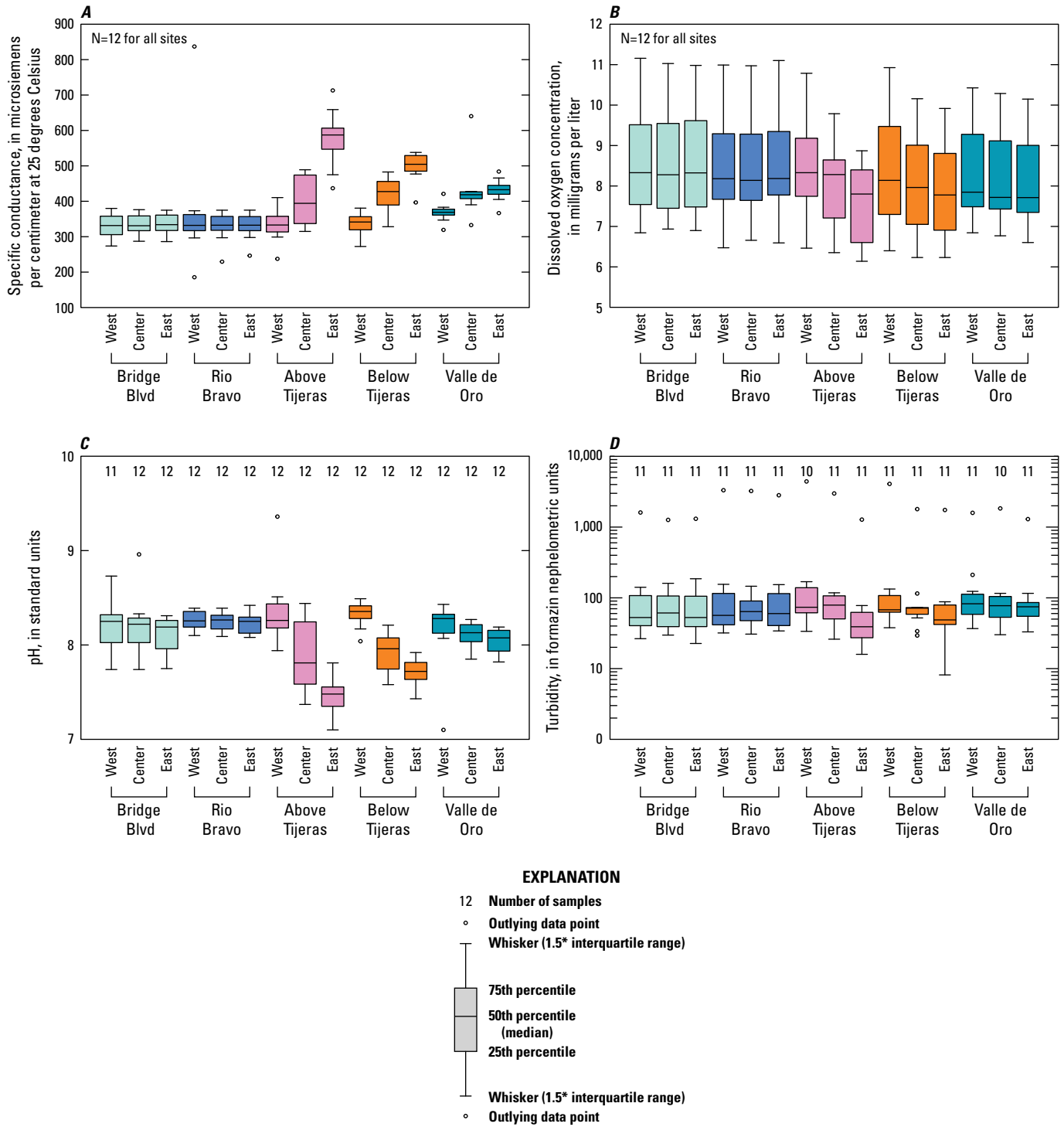


Figure 6. Boxplots showing physical parameters including *A*, specific conductance, *B*, dissolved oxygen, *C*, pH, and *D*, turbidity from all sampled events at five sites on the Rio Grande in Albuquerque, New Mexico.

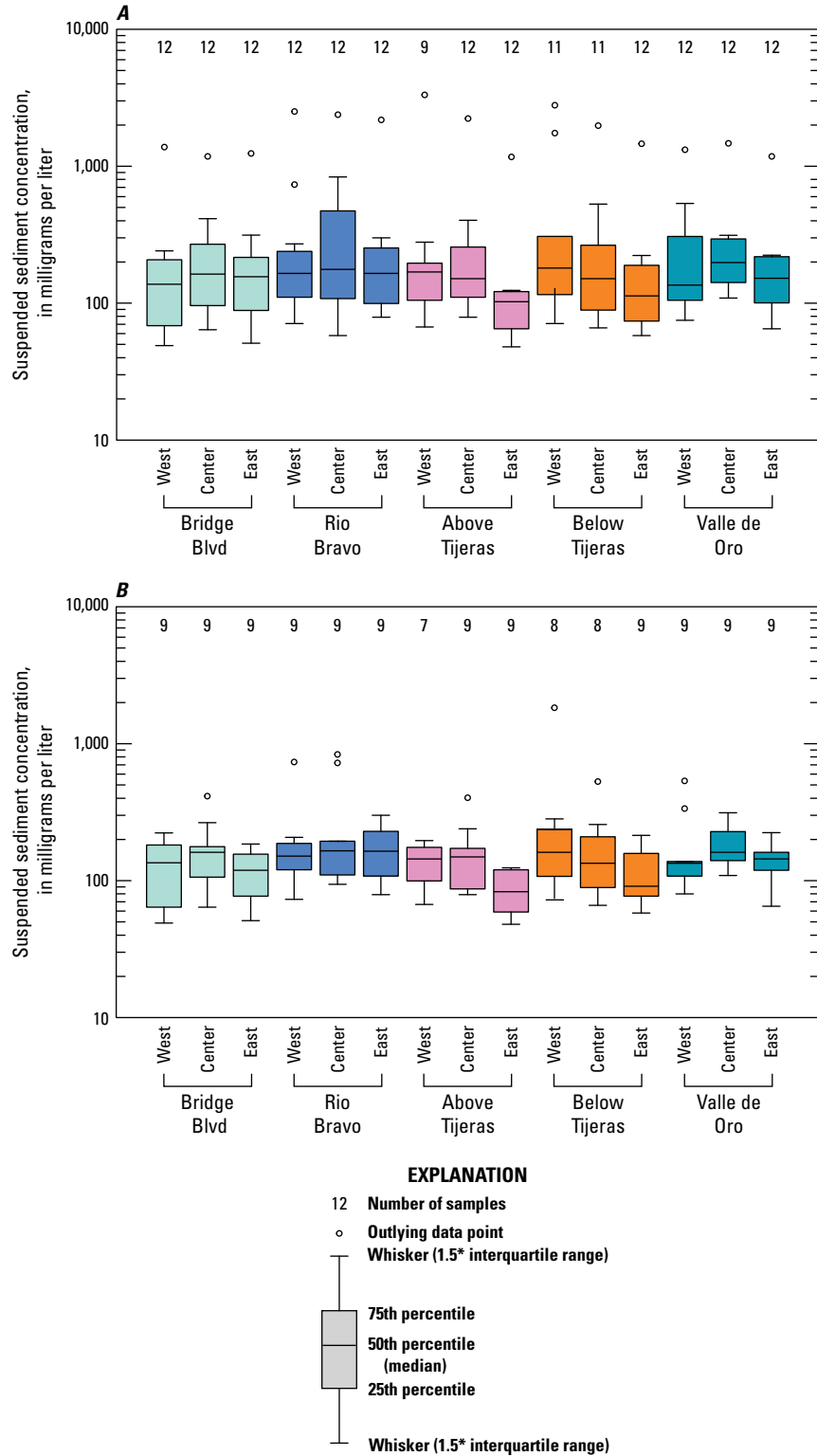


Figure 7. Boxplots showing suspended sediment concentrations from *A*, all sampled events and, *B*, dry season only events at five sites on the Rio Grande in Albuquerque, New Mexico.

concentrations for each site and sampling location. Figures 9 and 10 show boxplots for all data collected at each site and sampling location and data collected during the dry season only, respectively. Median *E. coli* values ranged from 42 to 160 MPN/100 mL for all samples, and from 32 to 120 MPN/100 mL for dry season samples (table 6). The west sample location had the highest concentrations collected at Above Tijeras (during the dry season, fig. 10) and Below Tijeras (all data, fig. 9), which were an order of magnitude higher in concentration than those of other samples. These higher values during the dry season could be indicative of an increase of fecal contamination on the west side of the river during the event.

Correlation coefficients were computed to determine how well *E. coli* concentrations correlated with discharge, suspended sediment concentration, and turbidity (table 7). Relations between *E. coli* and the other constituents were not strong (Pearson's $r < 0.9$ and Kendall's tau < 0.7), which is likely because the majority of these samples were collected in the dry season, when *E. coli* concentrations aren't likely to be due to increased flow, such as during storm events with greater discharge. The strongest correlation is between *E. coli* in water and turbidity.

Escherichia coli Mean Daily Fluxes

Mean daily fluxes were calculated for each *E. coli* sample collected from the three-point sampling events (fig. 11, table 8). The TMDL for *E. coli* for the study reach ranges from 5.27×10^{12} cfu/100 mL/day in high flow conditions (2,960 million gallons per day or 4,580 ft³/s) to 1.89×10^{11} cfu/100 mL/day in low flow conditions (106 million gallons per day or 164 ft³/s; NMEDSWQB, 2019b). Because there were limited *E. coli* sampling data, true loads could not be calculated, and fluxes (MPN/day) are the rates that have been calculated. Therefore, the TMDL and the fluxes cannot be directly compared.

Fluxes were consistently under 1.0×10^4 billion MPN/day for the bulk of the study except for one event (fig. 11), which was a large storm event. That last storm event, which was in August 2021, had the greatest fluxes ranging from 5.89×10^3 billion MPN/day to 9.25×10^4 billion MPN/day.

Comparison of *Escherichia coli* to Water Quality Standards

Two water-quality standards were used for comparison to the data collected during this study. The NMWQS 20.6.4.105 and 20.6.4.900 New Mexico Administrative Code (New Mexico Water Quality Control Commission, 2010) for *E. coli* for a single sample is 410 cfu/100 mL for the study area reach. Additionally, Isleta Pueblo has a standard for *E. coli* for a single sample of 88 cfu/100 mL for primary contact ceremonial and recreational use. All sites and locations had *E. coli* concentrations exceeding both standards. In total, 72 samples

exceeded the Isleta Pueblo standard of 88 cfu/100 mL, and 19 samples exceeded the NMWQS of 410 cfu/100 mL. In samples collected during only the dry season, 41 samples exceeded the Isleta Pueblo standard, and 1 sample exceeded the NMWQS.

Below Tijeras had the most exceedances with 11 sampling events having 1 or more sample locations exceeding the Isleta standard. Two events exceeded the NMWQS, both during the wet season. The exceedances ranged from 96 to 14,000 MPN/100 mL, and the east bank had the most frequent exceedances for all 11 sampling events. For the dry season samples, Above Tijeras was the site having the one sample exceeding the NMWQS, with a value of 1,400 MPN/100 mL, and the Below Tijeras remained the site with the highest number of exceedances over the Isleta Pueblo standard.

Characterization of *Escherichia coli* and Particle Size Analysis in Bed Material

Samples for bed material analyzed for *E. coli* and particle size analysis were only collected during the dry season. The mean values for *E. coli* at all the sites ranged from 0.5 to 10 MPN/g_{DW} (Table 6). Unlike the dry-season *E. coli* concentrations in water where the maximum value was observed at the Above Tijeras West location, the maximum concentration for *E. coli* in bed material was observed at Above Tijeras East with 59 MPN/g_{DW}. There is little variation of *E. coli* in bed material between the sites (fig. 12). Six percent of bed material samples were below the detection limit. Above Tijeras showed the most variation between the west and east locations.

Approximately 78 percent of the samples (n=72) had no fine particles (USGS, 2021). Bridge Blvd and Rio Bravo showed no fine material, but the downstream sites had a small percentage (4 percent or less) of fine material. Particle sizes in sediment have been identified as explaining some of the *E. coli* variability in rivers. Pachepsky and Shelton (2011) found more studies reporting an increase in *E. coli* concentrations when there was an increased amount of clay and silt bed material present. Coarse sediments may not provide as much protection from the sunlight or protozoan grazing for *E. coli* to survive as would finer sediments (Pachepsky and Shelton, 2011). Because *E. coli* concentrations in bed material were detected at low concentrations and the bed material is primarily a sandy substrate during the dry season, it is likely that the substrate is not a suitable habitat that would allow for bacterial growth during the dry season. Sampling particle size in both bed sediment and suspended sediment and *E. coli* concentrations in bed material during or directly after a storm would provide insight about whether more fine-grained material is present in bed material that could contribute to a higher *E. coli* concentration in the water.

This sand/fine particle ratio was compared to historical bed sediment data collected at the Albuquerque gage (USGS site no. 08330000) to understand if this finding is similar to historical data in the river. The gage is upstream from the

Table 6. Summary statistics for *Escherichia coli* (*E. coli*) in water for all samples, for samples collected during the dry season only, and for *E. coli* in bed material during the dry season.

[Data from U.S. Geological Survey (USGS, 2021). MPN/100 mL, most probable number per 100 milliliters; MPN/g_{DW}, most probable number per gram dry weight; Blvd, boulevard]

Site name	Summary statistics for <i>E. coli</i> concentrations in water (MPN/100 mL) for all data				Summary statistics for <i>E. coli</i> concentrations in water (MPN/100 mL) for dry season				Summary statistics for <i>E. coli</i> concentrations in bed material (MPN/g _{DW}) for dry season			
	Mean	Median	Minimum	Maximum	Mean	Median	Minimum	Maximum	Mean	Median	Minimum	Maximum
Bridge Blvd West	160	69	18	740	58	54	18	130	¹ 2	¹ 1	¹ 0.1	¹ 10
Bridge Blvd Center	150	62	15	740	67	58	15	200	¹ 2	¹ 1	¹ 0.1	¹ 10
Bridge Blvd East	390	88	6	2,900	80	64	6	200	¹ 2	¹ 1	¹ 0.1	¹ 10
Rio Bravo West	370	54	12	3,400	68	42	12	220	¹ 0.5	¹ 0.8	¹ 0.1	¹ 21
Rio Bravo Center	240	45	15	1,900	52	43	15	180	¹ 0.5	¹ 0.8	¹ 0.1	¹ 21
Rio Bravo East	500	42	6	5,200	47	32	6	170	¹ 0.5	¹ 0.8	¹ 0.1	¹ 21
Above Tijeras West	540	81	6	4,100	200	47	6	1,400	0.6	0.4	0.1	1.1
Above Tijeras Center	810	59	24	8,700	61	50	24	140	0.7	0.3	0.1	4
Above Tijeras East	380	150	58	2,900	120	87	58	240	10	3	0.6	59
Below Tijeras West	1,300	77	35	14,000	73	48	35	170	0.8	0.8	0.1	2
Below Tijeras Center	410	100	33	3,600	93	84	33	240	1	0.7	0.1	2
Below Tijeras East	370	160	64	2,400	150	120	64	290	3	1	0.1	9
Valle de Oro West	350	50	12	3,400	58	46	12	110	¹ 3	¹ 2	¹ 1	¹ 16
Valle de Oro Center	760	69	36	8,200	81	61	36	170	¹ 3	¹ 2	¹ 1	¹ 16
Valle de Oro East	750	100	56	7,700	100	89	56	230	¹ 3	¹ 2	¹ 1	¹ 16

¹Composite sample from the east, west, and center sampling locations.

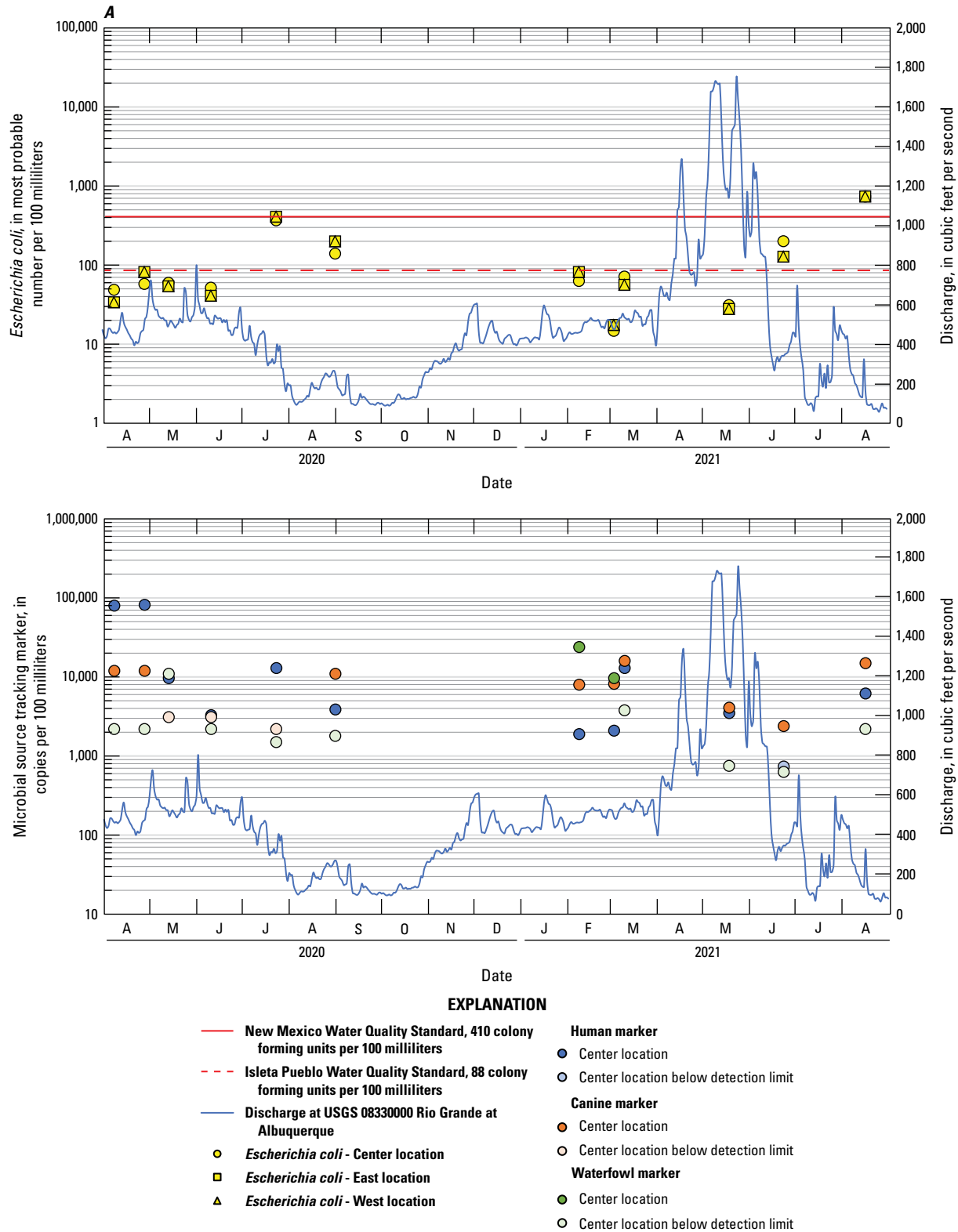


Figure 8. *Escherichia coli* and microbial source tracking marker time series plots showing data collected from all events at the A, Bridge Blvd, B, Rio Bravo, C, Above Tijeras, D, Below Tijeras, and E, Valle de Oro sites.

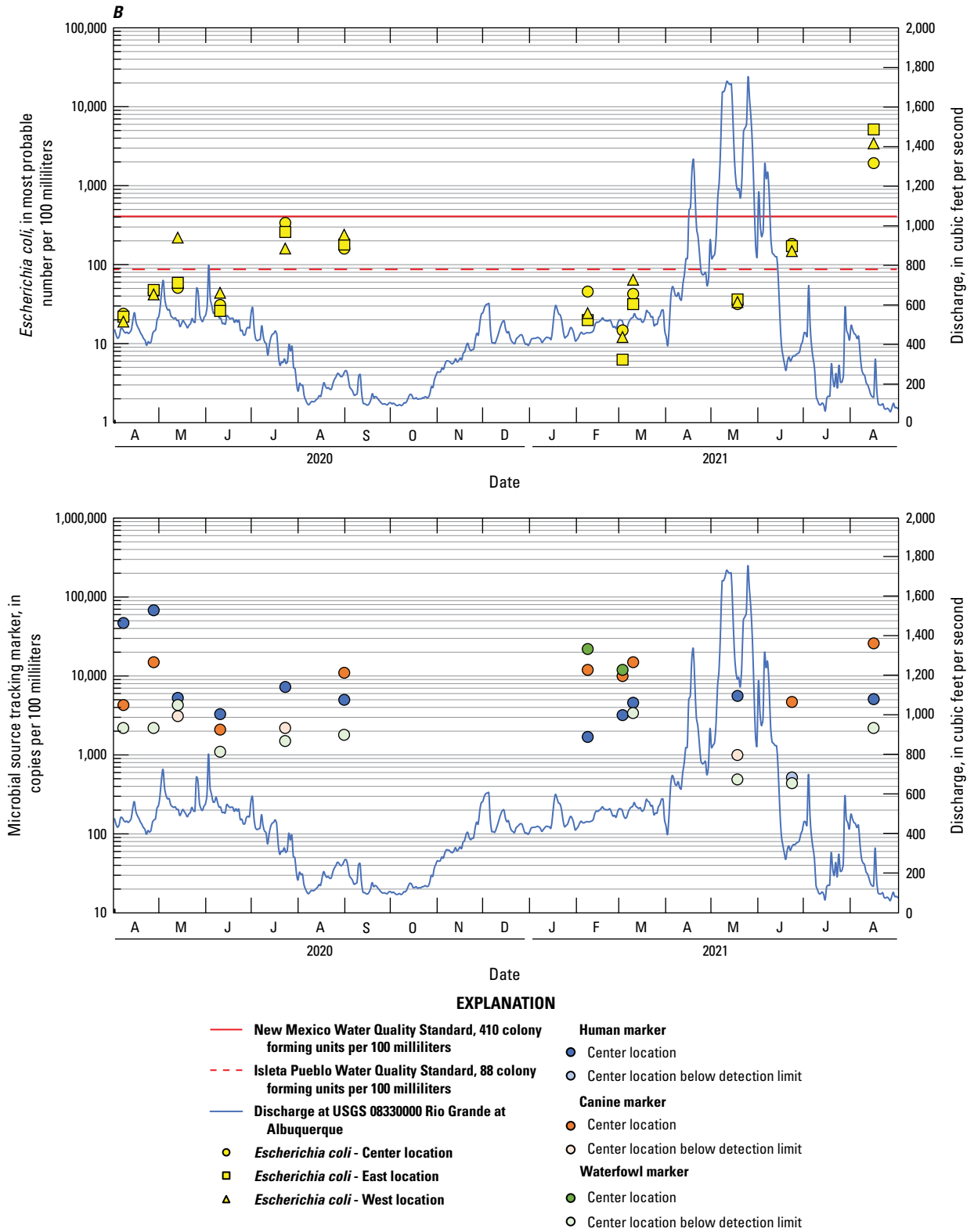


Figure 8.—Continued

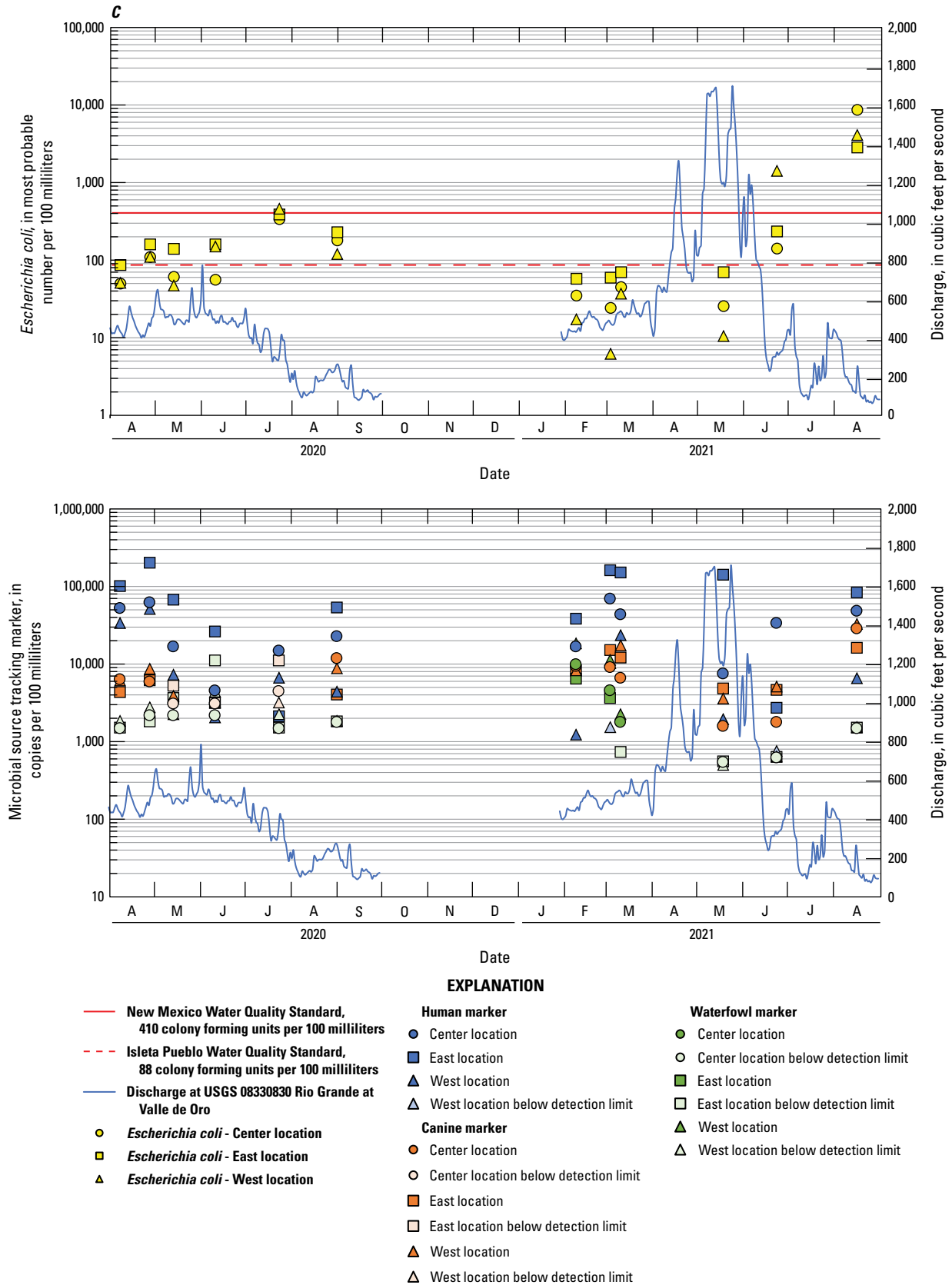


Figure 8.—Continued

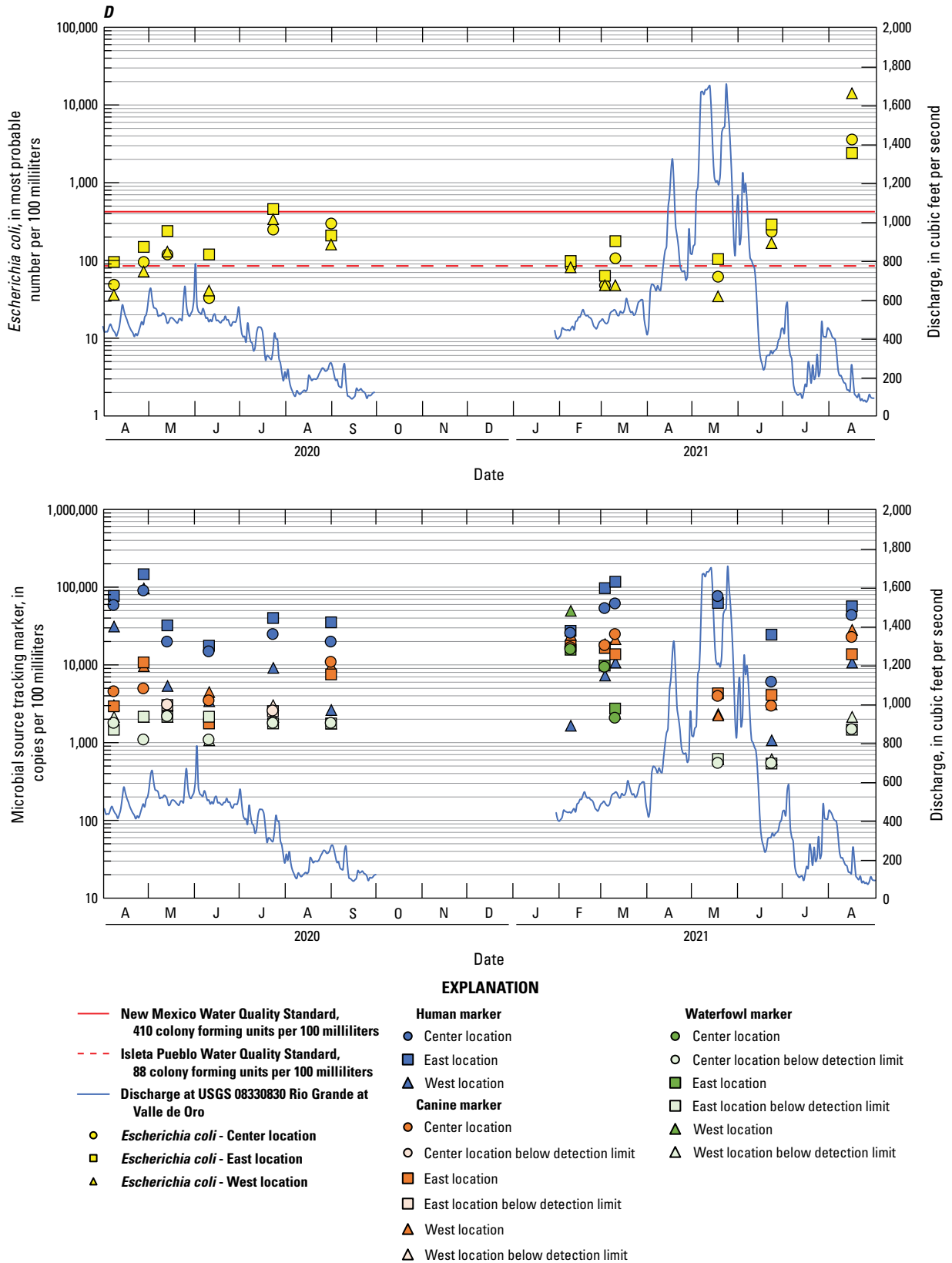


Figure 8.—Continued

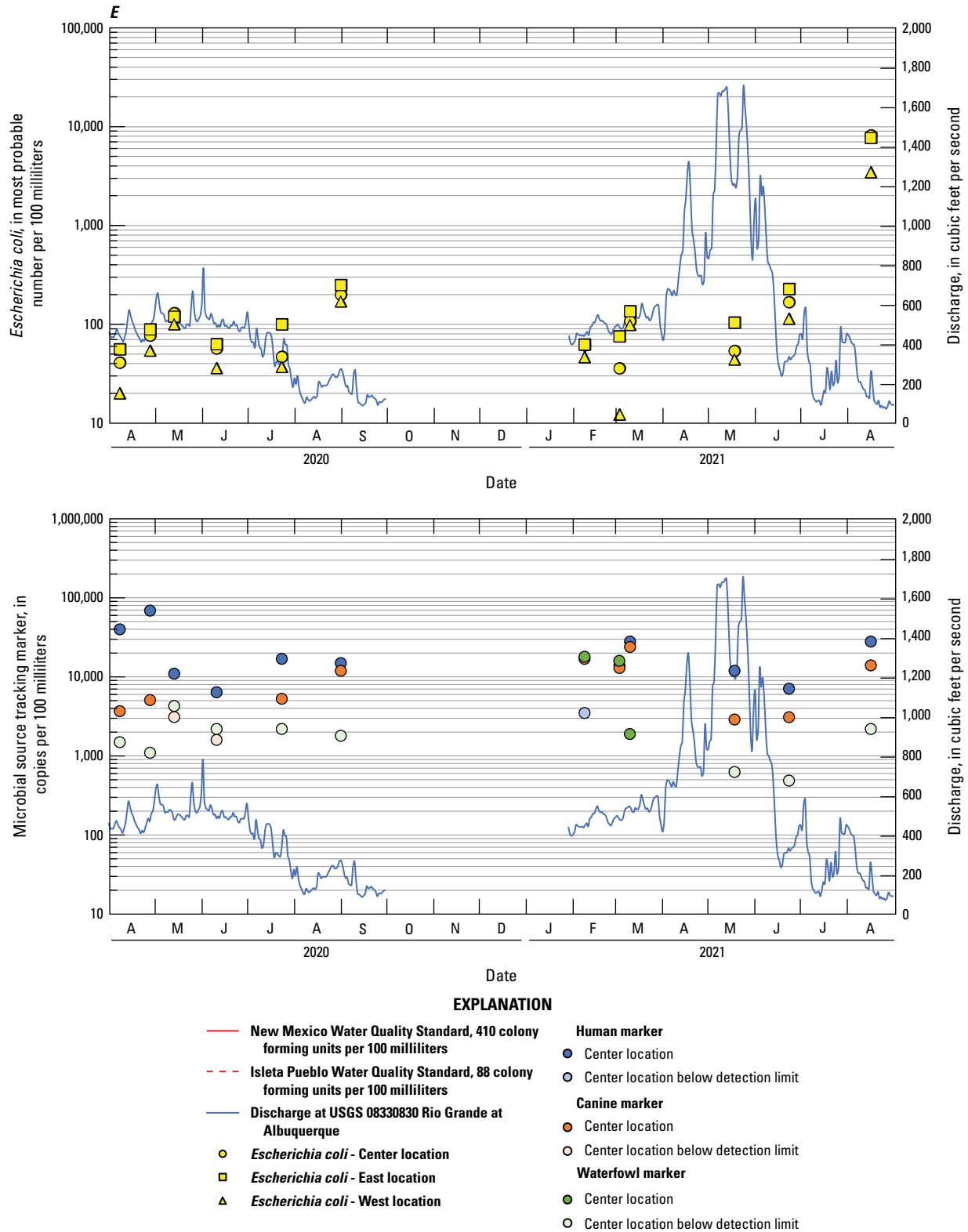


Figure 8.—Continued

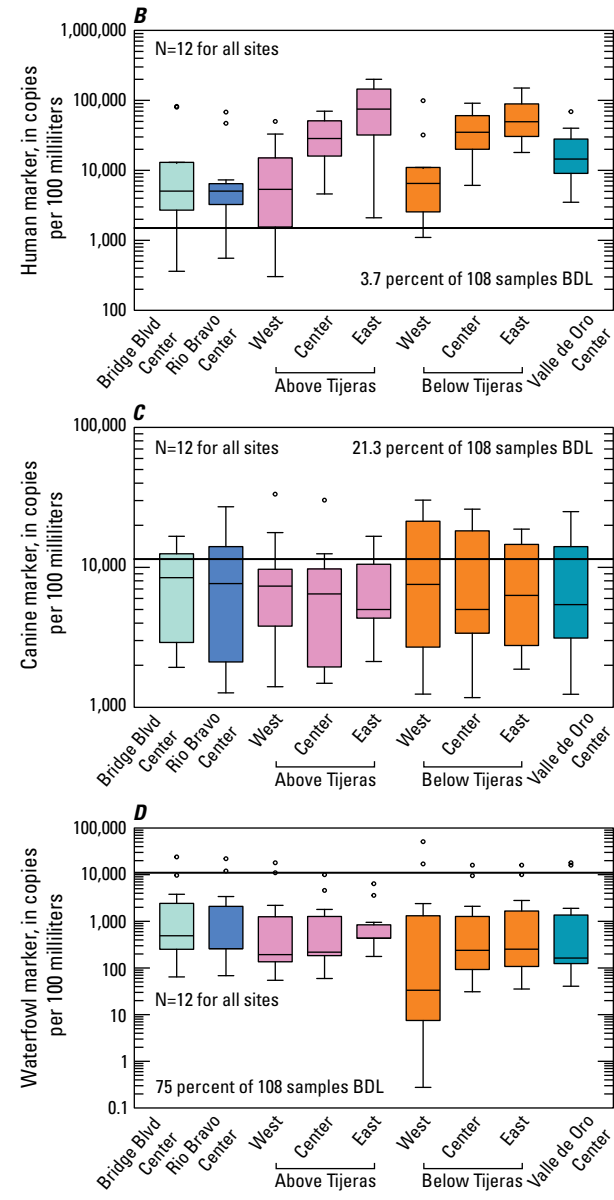
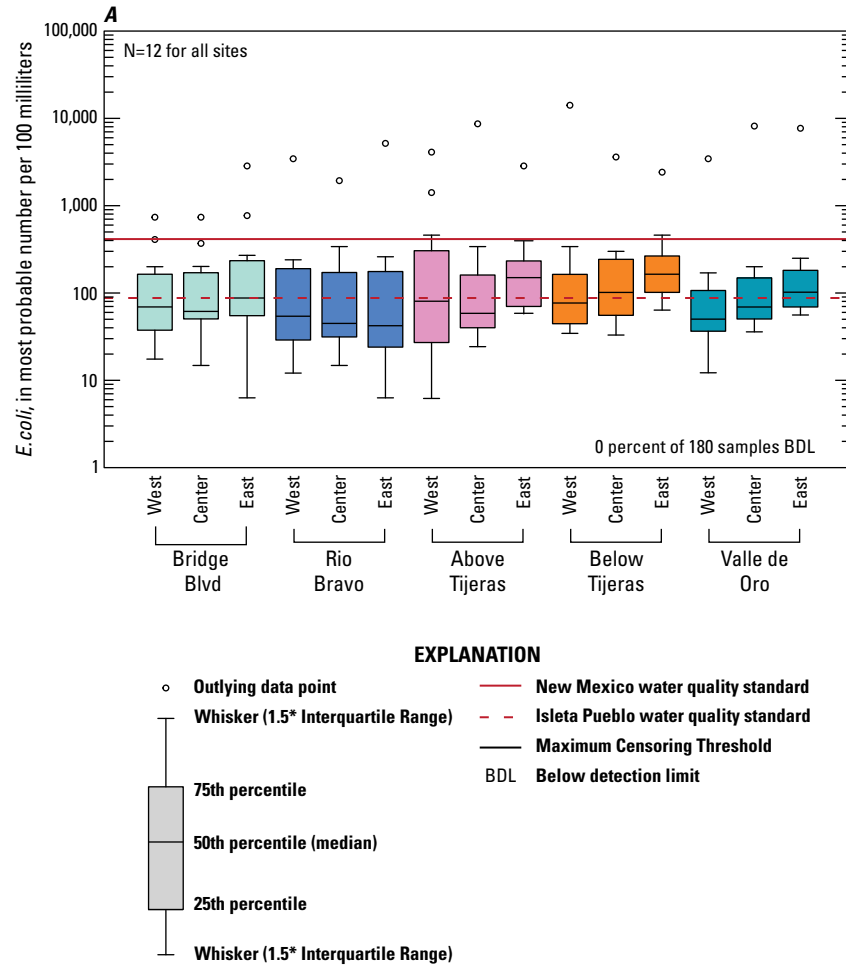


Figure 9. *Escherichia coli* (*E. coli*) and microbial source tracking marker boxplots showing data collected from all sampled events at five sites on the Rio Grande in Albuquerque, New Mexico.

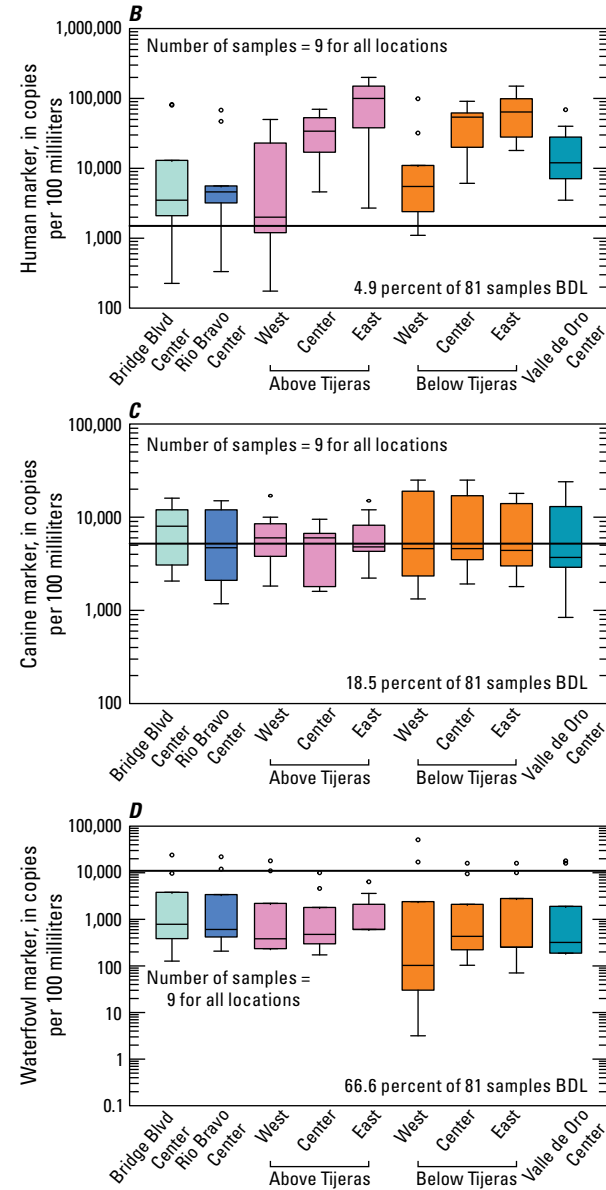
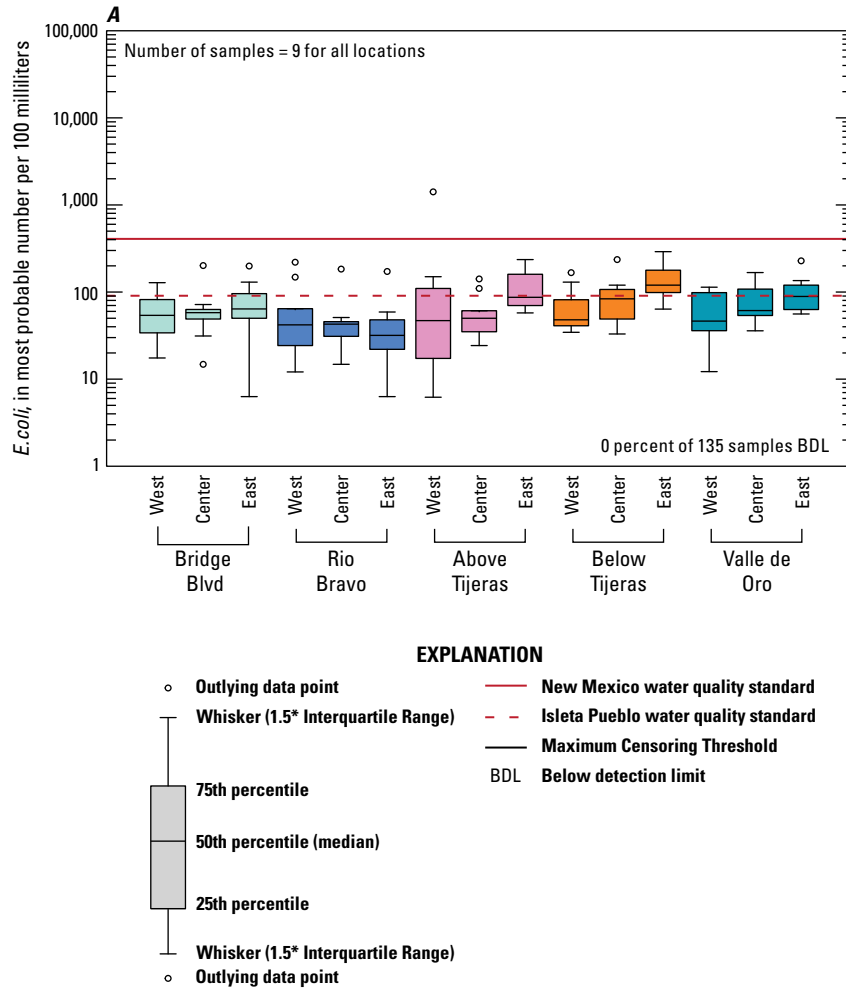
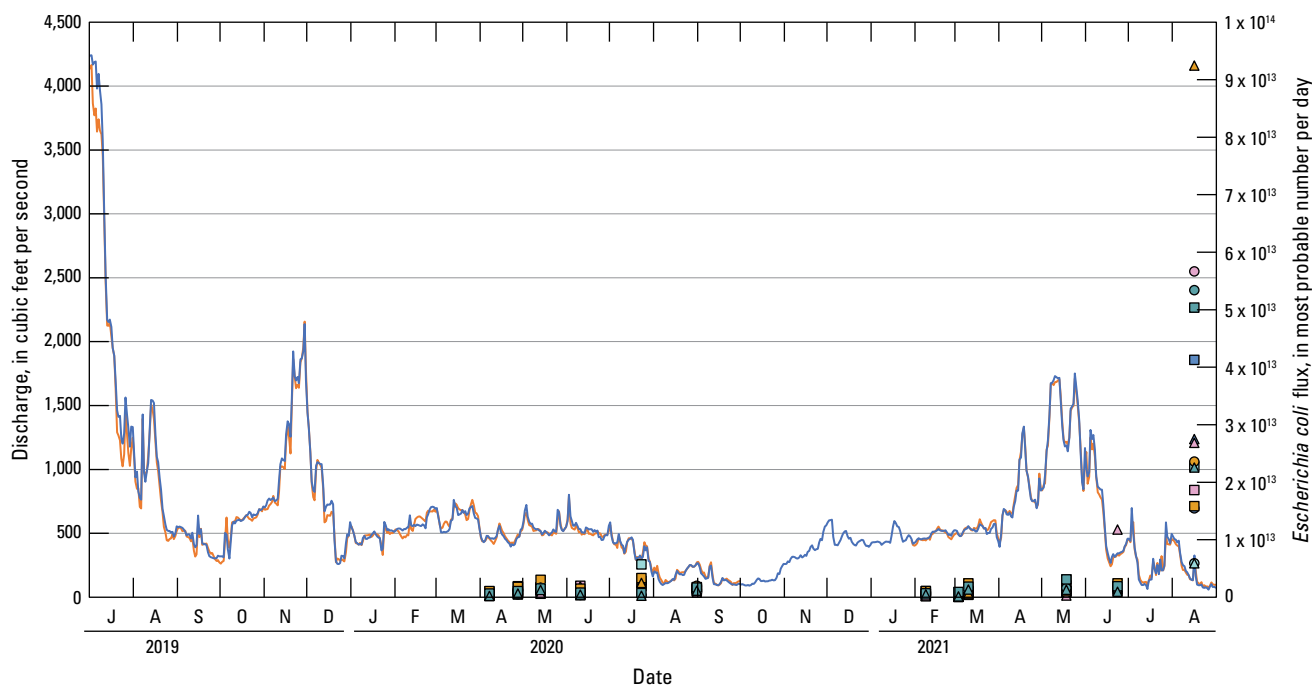


Figure 10. *Escherichia coli* (*E. coli*) and microbial source tracking marker boxplots showing data collected from dry season events at five sites on the Rio Grande in Albuquerque, New Mexico.

Table 7. Correlation statistics calculated for all data, including *Escherichia coli* (*E. coli*) in water and bed material against discharge, turbidity, and suspended sediment concentration.

[NC, not calculated]

Descriptor	Discharge		Turbidity		Suspended sediment concentration		<i>E. coli</i> in water		Human marker	
	Pearson's <i>r</i>	Kendall's tau	Pearson's <i>r</i>	Kendall's tau	Pearson's <i>r</i>	Kendall's tau	Pearson's <i>r</i>	Kendall's tau	Pearson's <i>r</i>	Kendall's tau
<i>E. coli</i> , in water	-0.44	-0.41	0.72	0.26	0.70	0.16	NC	NC	NC	0.05
<i>E. coli</i> , in bed material	NC	NC	-0.14	-0.05	-0.13	-0.06	0.27	0.24	NC	0.04



EXPLANATION

<p>Discharge</p> <ul style="list-style-type: none"> — 08330000 — 08330830 	<p><i>Escherichia coli</i> flux location</p> <ul style="list-style-type: none"> ○ Bridge Blvd Center ● Rio Bravo Center ○ Above Tijeras Center ● Below Tijeras Center ● Valle de Oro Center □ Bridge Blvd East ■ Rio Bravo East □ Above Tijeras East ■ Below Tijeras East ■ Valle de Oro East △ Bridge Blvd West ▲ Rio Bravo West △ Above Tijeras West ▲ Below Tijeras West ▲ Valle de Oro West
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Figure 11. Mean daily fluxes for *Escherichia coli* and corresponding discharge during the study period.

sampling sites and was used for comparative purposes, because bed sediment used for particle-size analysis has been collected for more than 20 years and provides a good description of the bed particle size distribution. The bed sediment data collected from 2019 to 2020 (USGS, 2021) for 0.125-, 0.25-, 0.5-, 1-, 2-, 4-, and 8-mm particle size

analysis were analyzed, and the first sediment particle size to reach 50 percent of the sample was considered the average size. For the entire date range, the average particle size during these years only ranged from 0.5 to 1 mm. As defined by the Wentworth Scale, 0.5 to 1 mm is considered “coarse sand” (Wentworth, 1922).

Table 8. *Escherichia coli* (*E. coli*) fluxes for each site and event at the three-point sampling locations.

[Data from U.S. Geological Survey (USGS, 2021). Dates shown as month/day/year. Mean daily flux values are shown in E notation. MPN/d, most probable number per day; Blvd, boulevard]

Site name	Sample date	<i>E. coli</i> mean daily flux (billion MPN/d)		
		East location	Center location	West location
Bridge Blvd	4/7/2020	5.70E+02	5.58E+02	3.87E+02
Bridge Blvd	4/27/2020	6.87E+02	6.64E+02	9.38E+02
Bridge Blvd	5/13/2020	1.66E+03	7.65E+02	6.88E+02
Bridge Blvd	6/10/2020	8.34E+02	6.77E+02	5.34E+02
Bridge Blvd	7/23/2020	5.73E+03	2.75E+03	3.05E+03
Bridge Blvd	8/31/2020	1.76E+03	9.13E+02	1.30E+03
Bridge Blvd	2/8/2021	1.07E+03	7.07E+02	9.19E+02
Bridge Blvd	3/3/2021	7.57E+01	1.78E+02	2.10E+02
Bridge Blvd	3/10/2021	1.08E+03	9.78E+02	7.69E+02
Bridge Blvd	5/18/2021	1.00E+03	9.10E+02	8.11E+02
Bridge Blvd	6/23/2021	1.67E+03	1.69E+03	1.08E+03
Bridge Blvd	8/16/2021	2.28E+04	5.89E+03	5.89E+03
Rio Bravo	4/7/2020	2.51E+02	2.73E+02	2.17E+02
Rio Bravo	4/27/2020	5.49E+02	5.03E+02	4.81E+02
Rio Bravo	5/13/2020	7.52E+02	6.50E+02	2.80E+03
Rio Bravo	6/10/2020	3.39E+02	4.04E+02	5.73E+02
Rio Bravo	7/23/2020	1.93E+03	2.53E+03	1.19E+03
Rio Bravo	8/31/2020	1.17E+03	1.04E+03	1.57E+03
Rio Bravo	2/8/2021	2.23E+02	5.12E+02	2.72E+02
Rio Bravo	3/3/2021	7.57E+01	1.78E+02	1.45E+02
Rio Bravo	3/10/2021	4.33E+02	5.82E+02	8.76E+02
Rio Bravo	5/18/2021	1.06E+03	9.24E+02	9.77E+02
Rio Bravo	6/23/2021	1.45E+03	1.54E+03	1.25E+03
Rio Bravo	8/16/2021	4.13E+04	1.54E+04	2.75E+04
Above Tijeras	4/7/2020	9.78E+02	5.62E+02	5.73E+02
Above Tijeras	4/27/2020	1.92E+03	1.32E+03	1.32E+03
Above Tijeras	5/13/2020	1.76E+03	7.69E+02	5.92E+02
Above Tijeras	6/10/2020	2.00E+03	7.00E+02	1.87E+03
Above Tijeras	7/23/2020	2.83E+03	2.47E+03	3.34E+03
Above Tijeras	8/31/2020	1.54E+03	1.20E+03	8.03E+02
Above Tijeras	2/8/2021	6.26E+02	3.80E+02	1.88E+02
Above Tijeras	3/3/2021	7.21E+02	2.93E+02	7.47E+01
Above Tijeras	3/10/2021	9.50E+02	6.08E+02	4.98E+02
Above Tijeras	5/18/2021	2.08E+03	7.61E+02	3.12E+02
Above Tijeras	6/23/2021	1.97E+03	1.18E+03	1.18E+04
Above Tijeras	8/16/2021	1.86E+04	5.67E+04	2.69E+04
Below Tijeras	4/7/2020	1.08E+03	5.51E+02	4.05E+02
Below Tijeras	4/27/2020	1.80E+03	1.15E+03	8.64E+02
Below Tijeras	5/13/2020	3.03E+03	1.51E+03	1.64E+03

Table 8. *Escherichia coli* (*E. coli*) fluxes for each site and event at the three-point sampling locations. —Continued

[Data from U.S. Geological Survey (USGS, 2021). Dates shown as month/day/year. Mean daily flux values are shown in E notation. MPN/d, most probable number per day; Blvd, boulevard]

Site name	Sample date	<i>E. coli</i> mean daily flux (billion MPN/d)		
		East location	Center location	West location
Below Tijeras	6/10/2020	1.50E+03	4.12E+02	5.12E+02
Below Tijeras	7/23/2020	3.34E+03	1.82E+03	2.47E+03
Below Tijeras	8/31/2020	1.40E+03	2.01E+03	1.07E+03
Below Tijeras	2/8/2021	1.07E+03	9.08E+02	8.87E+02
Below Tijeras	3/3/2021	7.68E+02	5.77E+02	5.78E+02
Below Tijeras	3/10/2021	2.41E+03	1.45E+03	6.48E+02
Below Tijeras	5/18/2021	3.11E+03	1.84E+03	1.03E+03
Below Tijeras	6/23/2021	2.42E+03	1.97E+03	1.39E+03
Below Tijeras	8/16/2021	1.58E+04	2.36E+04	9.25E+04
Valle de Oro	4/7/2020	6.30E+02	4.61E+02	2.25E+02
Valle de Oro	4/27/2020	1.07E+03	9.24E+02	6.48E+02
Valle de Oro	5/13/2020	1.51E+03	1.64E+03	1.26E+03
Valle de Oro	6/10/2020	7.87E+02	7.12E+02	4.50E+02
Valle de Oro	7/23/2020	7.27E+02	3.41E+02	2.69E+02
Valle de Oro	8/31/2020	1.67E+03	1.34E+03	1.14E+03
Valle de Oro	2/8/2021	6.78E+02	6.66E+02	5.04E+02
Valle de Oro	3/3/2021	9.09E+02	4.33E+02	1.47E+02
Valle de Oro	3/10/2021	1.83E+03	1.46E+03	1.33E+03
Valle de Oro	5/18/2021	3.10E+03	1.60E+03	1.31E+03
Valle de Oro	6/23/2021	1.90E+03	1.40E+03	9.48E+02
Valle de Oro	8/16/2021	5.04E+04	5.34E+04	2.26E+04

E. coli concentrations in bed material had little to no correlation between *E. coli* in water, turbidity, and suspended sediment concentration on the basis of Pearson’s *r* and (or) Kendall’s tau (table 7). Of the two, Kendall’s tau is a more appropriate indicator of correlation for these parameters because the relation between *E. coli* in bed material and *E. coli* in water, turbidity, and suspended sediment concentration is not linear. The highest correlation observed was between *E. coli* in bed material and *E. coli* in water, with a Kendall’s tau of 0.24; however, it is well below the value of 0.7 and so does not indicate a strong correlation.

Microbial Source Tracking in Water and Bed Material

The human marker was detected in all but 3.7 percent of the 108 water samples collected (figs. 8–10). The human marker concentrations in water vary by location, increasing downstream from the effluent outfall (observed mainly

in center and east locations of the Above Tijeras and Below Tijeras sites) and do not vary by season. Median values in water samples ranged from 5,100 to 75,000 copies/100 mL for all data collected (fig. 9, table 9), and 2,000 to 100,000 copies/100 mL for concentrations during the dry season (fig. 10). The eastern sample location at Above Tijeras had the highest concentrations of up to 200,000 copies/100 mL, and that sample had a corresponding *E. coli* concentration in water of 160 MPN/100 mL.

No statistically significant correlation was found between *E. coli* concentrations in water and the human marker concentrations in water for this study, which was focused on dry season sampling (table 7). Although elevated human marker concentrations could indicate increases in fecal contamination, that was not observed during this sampling. The qPCR methods used in this study do not distinguish between viable and nonviable *Bacteroides* bacteria from which the human marker originates, and it has been found that dead bacteria can be detected by the method up to 4 weeks after it has been killed (Nielsen and others, 2007). Therefore, a high human

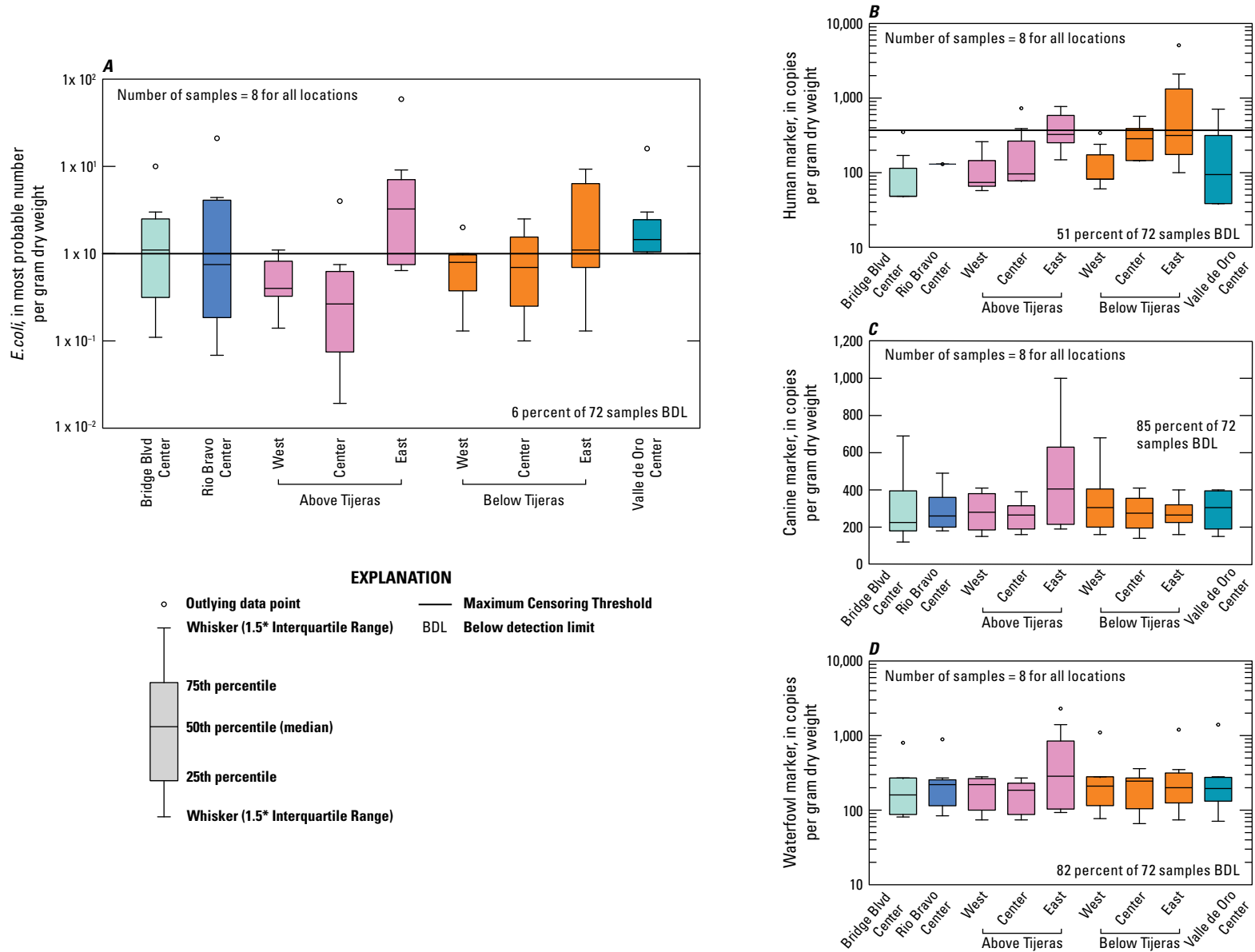


Figure 12. *Escherichia coli* (*E. coli*) and microbial source tracking markers in bed material boxplots showing data collected from all sampled events at five sites on the Rio Grande in Albuquerque, New Mexico.

Table 9. Summary statistics for human microbial source tracking markers in water for all samples and dry season samples and bed material samples from the dry season.

[Data from U.S. Geological Survey (USGS, 2021). copies/100 mL, most probable number per 100 milliliters; copies/g_{DW}, copies per gram dry weight; Blvd, boulevard; <, less than]

Site name and location	Summary statistics for human marker concentrations in water (copies/100 mL) for all data				Summary statistics for human marker concentrations in water (copies/100 mL) for dry season				Summary statistics for human marker concentrations in bed material (copies/g _{DW}) for dry season			
	Mean	Median	Minimum	Maximum	Mean	Median	Minimum	Maximum	Mean	Median	Minimum	Maximum
Bridge Blvd Center	34,000	17,000	<740	82,000	22,000	3,500	<740	82,000	100	48	<42	350
Rio Bravo Center	13,000	5,100	<520	68,000	15,000	4,600	<520	68,000	130	130	<100	200
Above Tijeras West	11,000	5,400	<740	50,000	13,000	2,000	<740	50,000	110	74	<95	260
Above Tijeras Center	33,000	29,000	4,600	70,000	34,000	34,000	4,600	70,000	210	96	96	730
Above Tijeras East	85,000	75,000	2,100	200,000	98,000	100,000	2,700	200,000	400	330	<150	770
Below Tijeras West	16,000	6,500	1,100	99,000	18,000	5,500	1,100	99,000	130	82	<72	340
Below Tijeras Center	42,000	35,000	6,100	91,000	46,000	54,000	6,100	91,000	300	290	<78	570
Below Tijeras East	63,000	50,000	18,000	150,000	68,000	64,000	18,000	150,000	1,100	310	100	5,100
Valle de Oro Center	21,000	15,000	3,500	69,000	21,000	12,000	3,500	69,000	210	94	<85	710

marker concentration does not always equate with a high viable *E. coli* concentration, as was the case downstream from the effluent outfall, where a large amount of nonviable human-associated markers were found with smaller concentrations of *E. coli*.

For bed material samples, human marker median values ranged from 48 to 330 copies per gram dry weight (copies/g_{DW}) (fig. 12, table 9). The maximum human marker observed in the bed material was 5,100 copies/g_{DW} at Below Tijeras East, with an associated *E. coli* concentration of 4 MPN/g_{DW}. There was also little correlation found between the *E. coli* concentrations in bed material and human markers in water (table 7).

The canine markers in water do not appear to vary by location and slightly decrease during the dry season (figs. 9 and 10; table 10). Of the 108 samples collected, 21.3 percent were below the detection limit. Median concentrations ranged from 4,800 to 7,400 copies/100 mL for all data collected (fig. 9) and ranged from 3,700 to 8,000 copies/100 mL during the dry season (fig. 10). The west sample locations at Above Tijeras and Below Tijeras had the highest concentrations (32,000 and 29,000 copies/100 mL, respectively) and corresponding *E. coli* concentrations of 4,100 and 14,000 MPN/100 mL, respectively.

Summary statistics could not be calculated for the canine marker in bed material samples using the NADA package, because more than 80 percent of the data were censored (Lee, 2020). Maximum and minimum values in samples where the canine marker was above the detection limit were variable. The concentrations ranged from 180 to 680 copies/g_{DW}. The maximum concentration of 680 copies/g_{DW} was observed at Below Tijeras West and had a corresponding *E. coli* concentration in bed material of 0.9 MPN/g_{DW} (USGS, 2021).

The waterfowl markers in water do not appear to vary by location and slightly increase during the dry season, during the months when the migratory bird population increases (figs. 8 and 9). Seventy-five percent of the 108 samples were below the detection limit. Median values ranged from 33 to 480 copies/100 mL for all data collected and increased to 100–780 copies/100 mL during the dry season (Table 11). The western sample locations at Below Tijeras had the highest concentration (51,000 copies/100 mL) and a corresponding *E. coli* concentration in water of 82 MPN/100 mL.

The waterfowl marker was not detected in 82 percent of bed material samples, and summary statistics could not be calculated using the NADA package (Lee, 2020). Maximum and minimum concentrations of the waterfowl marker that were above the detection limit were 1,100 and 130 copies/g_{DW}, respectively. Out of this range, the maximum concentration was the only value reported that was not qualified as an estimate with “b” (extrapolated below the limit of quantification but above the limit of detection). This sample, having a concentration of 1,100 copies/g_{DW}, was collected at the Below Tijeras site from the west bank location in February 2021 and had a corresponding *E. coli* concentration for this sample of 0.9 MPN/g_{DW} (USGS, 2021).

Mean daily fluxes were calculated for each MST marker in water collected from the three-point sampling events (table 12). These fluxes were then ranked and placed in bins and those bins were used to compare fluxes of different microbiological constituents at a single site and across multiple sites. The flux ranks are discussed below, and scores were used to compare results across sites.

Table 10. Summary statistics for canine microbial source tracking markers in water for all samples and for samples collected during the dry season only.

[Data from U.S. Geological Survey (USGS, 2021). copies/100 mL, most probable number per 100 milliliters; Blvd, boulevard;<, less than]

Site name and location	Summary statistics for canine marker concentrations in water (copies/100 mL) for all data				Summary statistics for canine marker concentrations in water (copies/100 mL) for dry season			
	Mean	Median	Minimum	Maximum	Mean	Median	Minimum	Maximum
Bridge Blvd Center	8,600	5,700	<2,200	16,000	7,500	8,000	2,400	16,000
Rio Bravo Center	8,800	7,400	<1,000	26,000	7,300	4,700	<1,000	15,000
Above Tijeras West	8,800	7,100	<3,100	32,000	7,100	6,000	<3,100	17,000
Above Tijeras Center	7,300	6,200	<1,600	29,000	5,000	6,000	<1,600	9,500
Above Tijeras East	7,200	4,800	<3,100	16,000	6,800	4,800	<3,100	15,000
Below Tijeras West	11,000	7,300	2,300	29,000	10,000	4,600	2,300	25,000
Below Tijeras Center	9,700	4,800	<2,600	25,000	9,100	4,600	3,000	25,000
Below Tijeras East	8,300	6,100	1,800	18,000	8,400	4,400	1,800	18,000
Valle de Oro Center	8,600	5,200	<1,600	24,000	7,900	3,700	<1,600	24,000

Table 11. Summary statistics for waterfowl microbial source tracking markers in water for all samples and the samples collected during the dry season only.

[Data from U.S. Geological Survey (USGS, 2021). copies/100 mL, most probable number per 100 milliliters; Blvd, boulevard; <, less than]

Site name and location	Summary statistics for waterfowl marker concentrations in water (copies/100 mL) for all data				Summary statistics for waterfowl marker concentrations in water (copies/100 mL) for dry season			
	Mean	Median	Minimum	Maximum	Mean	Median	Minimum	Maximum
Bridge Blvd Center	3,100	480	<630	24,000	4,500	780	<630	24,000
Rio Bravo Center	3,300	260	<440	22,000	4,500	600	<440	22,000
Above Tijeras West	2,700	190	<490	18,000	3,700	380	<490	18,000
Above Tijeras Center	1,600	220	<550	10,000	2,100	480	<550	10,000
Above Tijeras East	1,300	440	<550	11,000	1,700	610	<550	11,000
Below Tijeras West	5,900	33	<630	51,000	7,900	100	<630	51,000
Below Tijeras Center	2,400	240	<550	16,000	3,200	430	<550	16,000
Below Tijeras East	2,600	250	<550	16,000	3,400	250	<550	16,000
Valle de Oro Center	3,200	160	<490	18,000	4,100	320	<490	18,000

Spatial and Temporal Evaluation

Data were evaluated to determine if there were any temporal (dry versus wet season) and spatial (sample site and location) differences using the Kruskal-Wallis and Peto-Peto test statistics (table 13). These statistical tests look for differences between centers of independent groups (Helsel, 2012; Helsel and others, 2020). The most notable seasonal differences were observed in *E. coli* concentrations in water, suspended sediment concentrations, dissolved oxygen, and turbidity, which is consistent with the wet season samples collected after storm events, which can contribute *E. coli* and sediment to the system. Seasonal variation was not evaluated in bed material because all bed material samples were collected in the dry season. When evaluating the most statistically significant differences between sampling locations (each three-point sampling location at a sample site was grouped and evaluated), specific conductance, pH, and the human marker had the most statistically significant differences, most likely caused by the influence of the effluent along the eastern bank of the river downstream from the effluent outfall (fig. 4).

The pH was significantly lower, and the specific conductance was significantly higher at several sites and locations downstream from the effluent outfall (fig. 6). The pH and specific conductance at Above Tijeras East were statistically significantly different from all of the other locations (pH p-values ranged from 0.0003 to 0.0128 and specific conductance p-values ranged from 0.0002 to 0.0078). This statistically significant difference continued downstream but began to dissipate through the cross sections at the Below Tijeras and Valle de Oro sites (fig. 6).

The human marker increases significantly at the effluent outfall (fig. 9). Above Tijeras East and Center differed significantly from the upstream sites and western sites downstream,

with p-values ranging from 0.0051 to 0.0311. Although this difference was notable, there was no statistically significant increase of *E. coli* input into the system at the effluent outfall. As noted in the previous section, the analytical method does not distinguish between viable and nonviable bacteria (Nielsen and others, 2007). *E. coli* concentrations are a measurement of viable bacteria, but there was no statistically significant increase of *E. coli* concentrations in water noted throughout the study area since the study largely sampled during the dry season. Because the *E. coli* concentrations throughout the study reach did not have statistically significant differences, this would indicate that the effluent outfall was not a significant contribution of *E. coli* in the study area, and therefore the source of bacteria could be originating upstream from these sites.

The *E. coli* concentrations in bed material were statistically significantly greater at Above Tijeras East (p-values = 0.048), as compared to the other locations at Above Tijeras (fig. 12). The concentrations at Valle de Oro Center were significantly greater than those at the Above Tijeras Center and Above Tijeras West locations (p-values = 0.048) and those at Below Tijeras West (p-value = 0.048).

Strength of Bacteria and Potential Sources

Mean daily fluxes were calculated for *E. coli* and MST markers in water (tables 8 and 12). These fluxes were used to compare different microbiological constituents across multiple sites. The fluxes were ranked by site and placed into four equally sized bins with flux ranges for each bin listed in table 3 and the average bin scores listed in table 14 and figure 13. All results below the detection limit were placed into the 0 bin. The average bin scores listed in table 14 and

Table 12. Microbial source tracking marker mean daily flux for each site and event at the three-point sampling location.

[Dates shown as month/day/year. Mean daily flux values are shown in E notation. Blvd, boulevard; BDL, below detection limit]

Site name	Sample date	Sample location	Mean daily flux (billion copies per day)		
			Human marker	Canine marker	Waterfowl marker
Bridge Blvd	4/7/2020	Center	9.12E+05	1.37E+05	BDL
Bridge Blvd	4/27/2020	Center	9.38E+05	1.37E+05	BDL
Bridge Blvd	5/13/2020	Center	1.24E+05	BDL	BDL
Bridge Blvd	6/10/2020	Center	4.30E+04	BDL	BDL
Bridge Blvd	7/23/2020	Center	9.67E+04	BDL	BDL
Bridge Blvd	8/31/2020	Center	2.54E+04	7.18E+04	BDL
Bridge Blvd	2/8/2021	Center	2.13E+04	8.97E+04	2.69E+05
Bridge Blvd	3/3/2021	Center	2.52E+04	9.85E+04	1.17E+05
Bridge Blvd	3/10/2021	Center	1.77E+05	2.18E+05	5.17E+04
Bridge Blvd	5/18/2021	Center	1.02E+05	1.19E+05	2.18E+04
Bridge Blvd	6/23/2021	Center	BDL	2.02E+04	BDL
Bridge Blvd	8/16/2021	Center	4.95E+04	1.20E+05	BDL
Rio Bravo	4/7/2020	Center	5.36E+05	4.90E+04	BDL
Rio Bravo	4/27/2020	Center	7.78E+05	1.72E+05	BDL
Rio Bravo	5/13/2020	Center	6.75E+04	BDL	BDL
Rio Bravo	6/10/2020	Center	4.30E+04	2.74E+04	BDL
Rio Bravo	7/23/2020	Center	5.43E+04	BDL	BDL
Rio Bravo	8/31/2020	Center	3.26E+04	7.18E+04	BDL
Rio Bravo	2/8/2021	Center	1.91E+04	1.35E+05	2.47E+05
Rio Bravo	3/3/2021	Center	3.84E+04	1.20E+05	1.44E+05
Rio Bravo	3/10/2021	Center	6.26E+04	2.04E+05	4.63E+04
Rio Bravo	5/18/2021	Center	1.63E+05	BDL	BDL
Rio Bravo	6/23/2021	Center	BDL	3.95E+04	BDL
Rio Bravo	8/16/2021	Center	4.07E+04	2.07E+05	BDL
Above Tijeras	4/7/2020	Center	5.96E+05	7.19E+04	BDL
Above Tijeras	5/13/2020	Center	2.14E+05	BDL	BDL
Above Tijeras	6/10/2020	Center	5.75E+04	BDL	BDL
Above Tijeras	7/23/2020	Center	1.09E+05	BDL	BDL
Above Tijeras	8/31/2020	Center	1.54E+05	8.03E+04	BDL
Above Tijeras	2/8/2021	Center	1.85E+05	1.03E+05	1.09E+05
Above Tijeras	3/3/2021	Center	8.43E+05	1.11E+05	5.54E+04
Above Tijeras	3/10/2021	Center	5.94E+05	9.05E+04	2.43E+04
Above Tijeras	5/18/2021	Center	2.26E+05	4.76E+04	BDL
Above Tijeras	6/23/2021	Center	2.83E+05	1.50E+04	BDL
Above Tijeras	8/16/2021	Center	3.20E+05	1.90E+05	BDL
Above Tijeras	4/7/2020	East	1.12E+06	4.83E+04	BDL
Above Tijeras	4/27/2020	East	2.40E+06	7.32E+04	BDL
Above Tijeras	5/13/2020	East	8.45E+05	6.56E+04	BDL
Above Tijeras	6/10/2020	East	1.53E+04	BDL	BDL

Table 12. Microbial source tracking marker mean daily flux for each site and event at the three-point sampling location.—Continued

[Dates shown as month/day/year. Mean daily flux values are shown in E notation. Blvd, boulevard; BDL, below detection limit]

Site name	Sample date	Sample location	Mean daily flux (billion copies per day)		
			Human marker	Canine marker	Waterfowl marker
Above Tijeras	7/23/2020	East	3.55E+05	BDL	BDL
Above Tijeras	8/31/2020	East	3.55E+05	2.68E+04	BDL
Above Tijeras	2/8/2021	East	4.13E+05	8.91E+04	6.95E+04
Above Tijeras	3/3/2021	East	1.93E+06	1.81E+05	4.34E+04
Above Tijeras	3/10/2021	East	2.03E+06	1.62E+05	BDL
Above Tijeras	5/18/2021	East	4.16E+06	1.43E+05	BDL
Above Tijeras	6/23/2021	East	2.25E+04	3.83E+04	BDL
Above Tijeras	8/16/2021	East	5.43E+05	1.05E+05	BDL
Above Tijeras	4/7/2020	West	3.71E+05	6.75E+04	BDL
Above Tijeras	4/27/2020	West	6.00E+05	1.02E+05	BDL
Above Tijeras	5/13/2020	West	8.95E+04	4.79E+04	BDL
Above Tijeras	6/10/2020	West	2.50E+04	BDL	BDL
Above Tijeras	7/23/2020	West	4.72E+04	BDL	BDL
Above Tijeras	8/31/2020	West	2.88E+04	5.75E+04	BDL
Above Tijeras	2/8/2021	West	1.30E+04	8.80E+04	1.96E+05
Above Tijeras	3/3/2021	West	BDL	1.20E+05	1.33E+05
Above Tijeras	3/10/2021	West	3.11E+05	2.30E+05	2.97E+04
Above Tijeras	5/18/2021	West	5.65E+04	1.04E+05	BDL
Above Tijeras	6/23/2021	West	BDL	4.17E+04	BDL
Above Tijeras	8/16/2021	West	4.19E+04	2.09E+05	BDL
Below Tijeras	4/7/2020	Center	6.63E+05	5.17E+04	BDL
Below Tijeras	4/27/2020	Center	1.09E+06	6.00E+04	BDL
Below Tijeras	5/13/2020	Center	2.52E+05	BDL	BDL
Below Tijeras	6/10/2020	Center	1.87E+05	4.37E+04	BDL
Below Tijeras	7/23/2020	Center	1.82E+05	BDL	BDL
Below Tijeras	8/31/2020	Center	1.34E+05	7.36E+04	BDL
Below Tijeras	2/8/2021	Center	2.83E+05	1.85E+05	1.74E+05
Below Tijeras	3/3/2021	Center	6.51E+05	2.17E+05	1.14E+05
Below Tijeras	3/10/2021	Center	8.37E+05	3.38E+05	2.84E+04
Below Tijeras	5/18/2021	Center	2.29E+06	1.19E+05	BDL
Below Tijeras	6/23/2021	Center	5.08E+04	2.50E+04	BDL
Below Tijeras	8/16/2021	Center	2.88E+05	1.50E+05	BDL
Below Tijeras	4/7/2020	East	8.88E+05	3.37E+04	BDL
Below Tijeras	4/27/2020	East	1.80E+06	1.32E+05	BDL
Below Tijeras	5/13/2020	East	4.16E+05	BDL	BDL
Below Tijeras	6/10/2020	East	2.25E+05	2.25E+04	BDL
Below Tijeras	7/23/2020	East	2.98E+05	BDL	BDL
Below Tijeras	8/31/2020	East	2.41E+05	5.15E+04	BDL
Below Tijeras	2/8/2021	East	3.04E+05	1.96E+05	1.74E+05

Table 12. Microbial source tracking marker mean daily flux for each site and event at the three-point sampling location.—Continued

[Dates shown as month/day/year. Mean daily flux values are shown in E notation. Blvd, boulevard; BDL, below detection limit]

Site name	Sample date	Sample location	Mean daily flux (billion copies per day)		
			Human marker	Canine marker	Waterfowl marker
Below Tijeras	3/3/2021	East	1.19E+06	2.05E+05	1.20E+05
Below Tijeras	3/10/2021	East	1.62E+06	1.89E+05	3.78E+04
Below Tijeras	5/18/2021	East	1.90E+06	1.31E+05	BDL
Below Tijeras	6/23/2021	East	2.08E+05	3.50E+04	BDL
Below Tijeras	8/16/2021	East	3.79E+05	9.16E+04	BDL
Below Tijeras	4/7/2020	West	3.60E+05	BDL	BDL
Below Tijeras	4/27/2020	West	1.19E+06	1.19E+05	BDL
Below Tijeras	5/13/2020	West	6.93E+04	BDL	BDL
Below Tijeras	6/10/2020	West	4.37E+04	5.75E+04	BDL
Below Tijeras	7/23/2020	West	6.83E+04	BDL	BDL
Below Tijeras	8/31/2020	West	1.81E+04	6.69E+04	BDL
Below Tijeras	2/8/2021	West	1.85E+04	2.72E+05	5.54E+05
Below Tijeras	3/3/2021	West	9.04E+04	2.29E+05	2.05E+05
Below Tijeras	3/10/2021	West	1.49E+05	2.97E+05	3.24E+04
Below Tijeras	5/18/2021	West	7.14E+04	6.84E+04	BDL
Below Tijeras	6/23/2021	West	9.17E+03	2.67E+04	BDL
Below Tijeras	8/16/2021	West	7.19E+04	1.90E+05	BDL
Valle de Oro	4/7/2020	Center	4.50E+05	4.16E+04	BDL
Valle de Oro	4/27/2020	Center	8.28E+05	6.12E+04	BDL
Valle de Oro	5/13/2020	Center	1.39E+05	BDL	BDL
Valle de Oro	6/10/2020	Center	8.00E+04	BDL	BDL
Valle de Oro	7/23/2020	Center	1.24E+05	3.85E+04	BDL
Valle de Oro	8/31/2020	Center	1.00E+05	8.03E+04	BDL
Valle de Oro	2/8/2021	Center	3.80E+04	1.85E+05	1.96E+05
Valle de Oro	3/3/2021	Center	1.69E+05	1.57E+05	1.93E+05
Valle de Oro	3/10/2021	Center	3.78E+05	3.24E+05	2.57E+04
Valle de Oro	5/18/2021	Center	3.57E+05	8.62E+04	BDL
Valle de Oro	6/23/2021	Center	5.92E+04	2.58E+04	BDL
Valle de Oro	8/16/2021	Center	1.83E+05	9.16E+04	BDL

illustrated in [figure 13](#) can be used to compare the fluxes of a single constituent across sites and to compare the fluxes of different constituents at a single site.

The results of bin rankings indicate that for all samples, the *E. coli* mean daily fluxes were greatest at the Below Tijeras, Above Tijeras, and Valle de Oro sites ([fig. 13](#)). Average bin scores for the human marker were also highest at these sites ([table 14](#)), implying they are near a source of human fecal contamination. The average bin scores for the canine marker were highest at Bridge Blvd and Below

Tijeras. Within each site and among the three MST markers, the canine marker had the highest bin score at Bridge Blvd and Rio Bravo, and the human marker had the highest bin score at Above Tijeras, Below Tijeras, and Valle de Oro. The waterfowl marker was detected less frequently than the human and canine markers during both seasons, and bins scores were lower for all samples and the dry season. During the dry season, the average bin scores for *E. coli* and the human markers were highest at the same sites as those for all samples. The canine markers had the highest bin scores at Bridge Blvd and

Table 13. Kruskal-Wallis and Peto-Peto statistical test results for temporal and spatial differences.

[Statistically significant probability values (p-values; less than 0.05) are bolded. Small p-values are shown in E notation. *E. coli*, *Escherichia coli*; NC, not calculated]

Variable	Season p-value	Sample location p-value
Kruskal-Wallis statistical test		
<i>E. coli</i> , in water	2.20E-16	0.19
Suspended sediment concentrations	7.35E-06	0.26
Specific conductance	0.861	2.20E-16
pH	0.5946	2.85E-13
Dissolved oxygen	2.20E-16	0.86
Turbidity	1.06E-07	0.59
Peto-Peto statistical test		
Human marker, in water	0.563	8.09E-09
Canine marker, in water	0.103	0.998
Waterfowl, in water	NC	0.997
<i>E. coli</i> , in bed material	NC	0.0198

Table 14. Bin scores for *Escherichia coli* (*E. coli*) and microbial source tracking markers at all sites for all samples and dry season samples.

[Blvd, boulevard]

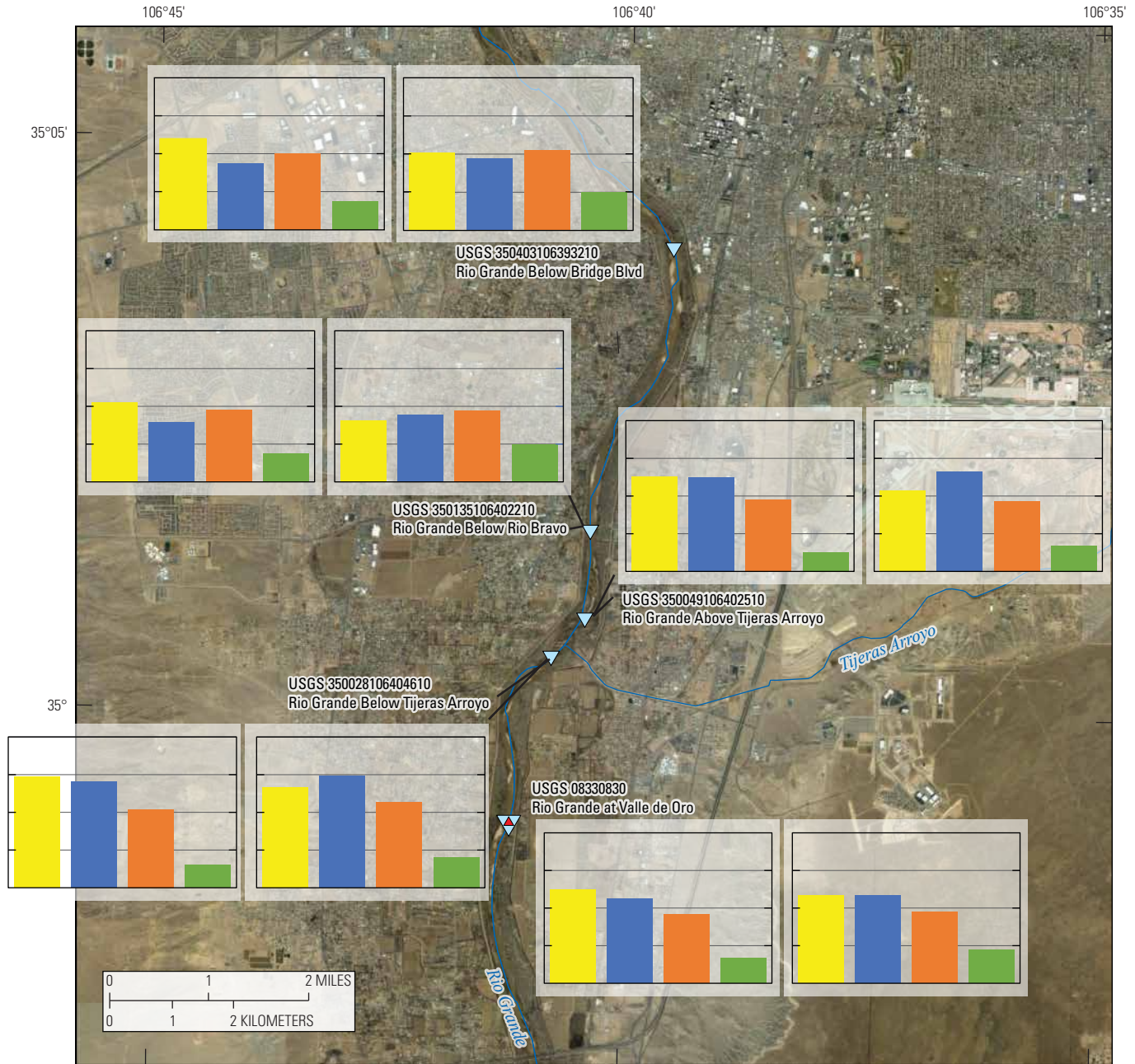
Site name	Average bin score			
	<i>E. coli</i>	Human marker	Canine marker	Waterfowl marker
All samples				
Bridge Blvd	2.39	1.75	2.00	0.83
Rio Bravo	2.11	1.58	1.92	0.75
Above Tijeras	2.64	2.53	1.89	0.47
Below Tijeras	2.92	2.81	2.11	0.61
Valle de Oro	2.44	2.33	1.83	0.67
Dry season samples				
Bridge Blvd	2.00	1.89	2.11	1.11
Rio Bravo	1.63	1.78	1.89	1.00
Above Tijeras	2.33	2.74	1.93	0.63
Below Tijeras	2.63	2.96	2.26	0.81
Valle de Oro	2.33	2.44	1.89	0.89

Below Tijeras. The waterfowl marker was found more often during the dry season than the wet season, and bin scores were highest at Bridge Blvd and Rio Bravo. Within each site and among the three MST markers, the canine marker remained the highest bin score at Bridge Blvd and Rio Bravo, and the human marker had the highest bin score at Above Tijeras, Below Tijeras, and Valle de Oro.

The top potential sources of fecal bacteria are human and canine (table 15) based on a combination of known potential sources of fecal bacteria, percentage of detections

of the source markers in both water and bed sediment samples, and analysis of the average bin scores for daily mean flux (table 15). Given that the analysis of known-source fecal sample from the wastewater influent from the WWTP resulted in detection of the canine marker in addition to the human marker, this could indicate that the canine marker could at least in part be from wastewater influence. This occurrence is not unexpected because fecal bacteria may be transferred between host species living in close contact (Field and Samadpour, 2007; Stewart and others, 2007; Roslev and

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Base from U.S. Geological Survey National Map 3D Elevation Program (3DEP)
 Universal Transverse Mercator, zone 13
 North American Datum of 1983

Land use from Dewitz and others, 2021

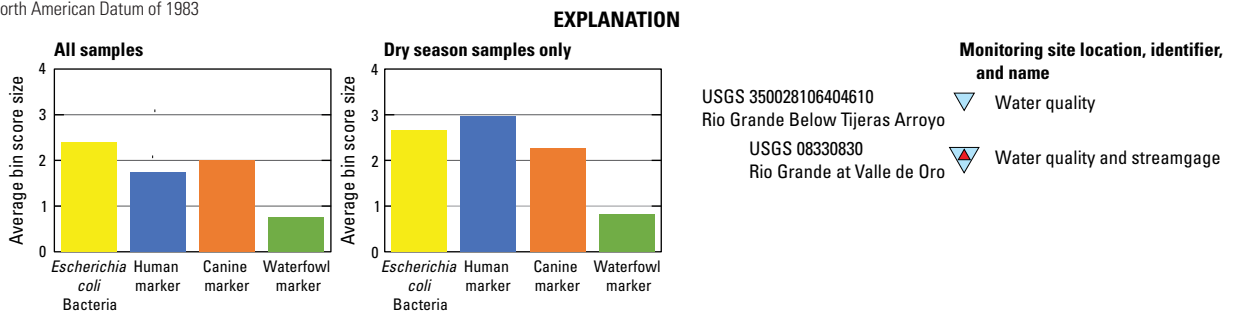


Figure 13. Average bin scores for *Escherichia coli* and microbial source tracking markers from all events and dry season events at five sites on the Rio Grande in Albuquerque, New Mexico.

Table 15. Summary of potential sources, *Escherichia coli* (*E. coli*) and microbial source tracking marker data from water samples at each site and location, and top source ranking.

[MPN/100 mL, most probable number per 100 milliliters; NS, not sampled; copies/100 mL, copies per 100 milliliters; Blvd, boulevard; %, percent]

Site name and location	Potential sources near site	Median <i>E. coli</i> (MPN/100 mL)	Human marker median (copies/100 mL)	Human marker percent nondetects	Canine marker median (copies/100 mL)	Canine marker percent nondetects	Waterfowl median (copies/100 mL)	Waterfowl marker percent nondetects	Highest bin ranking marker
Bridge Blvd West	Migratory bird flyway, open space trails, septic use	69	NS	NS	NS	NS	NS	NS	NS
Bridge Blvd Center	Same as above	62	17,000	8%	5,700	25%	480	67%	Canine
Bridge Blvd East	Same as above, plus dog park, pumping station	88	NS	NS	NS	NS	NS	NS	NS
Rio Bravo West	Septic use, migratory bird flyway, open space trails	54	NS	NS	NS	NS	NS	NS	NS
Rio Bravo Center	Same as above	45	5,100	8%	7,400	25%	260	75%	Canine
Rio Bravo East	Same as above	42	NS	NS	NS	NS	NS	NS	NS
Above Tijeras West	Septic use, migratory bird flyway, open space trails	81	5,400	17%	7,100	17%	190	75%	Human
Above Tijeras Center	Same as above	59	29,000	0%	6,200	25%	220	75%	Human
Above Tijeras East	Same as above, plus effluent outfall	150	75,000	0%	4,800	25%	440	83%	Human
Below Tijeras West	Confluence of Tijeras Arroyo, septic use, migratory bird flyway, open space trails	77	6,500	0%	7,300	25%	33	75%	Human
Below Tijeras Center	Same as above	100	35,000	0%	4,800	17%	240	75%	Human
Below Tijeras East	Same as above, plus effluent outfall	160	50,000	0%	6,100	17%	250	75%	Human
Valle de Oro West	Confluence of Tijeras Arroyo, septic use, migratory bird flyway, open space trails	50	NS	NS	NS	NS	NS	NS	NS
Valle de Oro Center	Same as above	69	15,000	0%	5,200	17%	160	75%	Human
Valle de Oro East	Same as above, plus effluent outfall	100	NS	NS	NS	NS	NS	NS	NS

Bukh 2011). With the presence of many different potential sources (fig. 4), the high incidence of septic use (LaMotte, 2018) and general park and trail use (City of Albuquerque Parks and Recreation Department Open Space Division, 2014) may be nonpoint sources contributing to the reach during the dry season because there is little correlation between *E. coli* concentrations and discharge. Also, the lack of statistically significant spatial differences in *E. coli* concentrations between sampling locations may also indicate that there are no significant contributions of *E. coli* within the study area and contributions could be coming from one or more sources upstream from the study area.

Summary

The Rio Grande, in southern Albuquerque, New Mexico, is a Clean Water Act Section 303(d) Category 5 impaired reach for *Escherichia coli* (*E. coli*). The reach is 5 miles in length, extending from Tijeras Arroyo to the Isleta Pueblo boundary. An evaluation of *E. coli* and microbial source trackers (human-, canine-, and waterfowl-specific sources) was conducted by the U.S. Geological Survey to determine the extent and source of fecal bacteria within the impaired reach of the Rio Grande during the dry season (November through June) in 2020 and 2021.

Samples collected during both the dry season and the wet season provided data over a range of flow conditions and provide insight about the extent and source of fecal bacteria. Because fecal microorganisms may readily attach to sediments, riverbed material samples were also collected. During the dry season, *E. coli* concentrations in water were primarily detected below the New Mexico Surface Water Quality Standard of 410 colony forming units per 100 milliliters and mostly human and canine sources were detected. However, approximately 40 percent of the water samples exceeded the Isleta Pueblo water-quality standard of 88 colony forming units per 100 milliliters.

E. coli concentrations in bed material were detected at low concentrations, and the bed material was a sandy substrate with little fine-grained material, likely not providing a suitable habitat for bacteria growth. Mostly human markers were detected in the bed material, with over 80 percent of the canine and waterfowl markers being below the detection limits.

Statistically significant spatial and temporal differences were found in water-quality samples for *E. coli* (seasonal) and the human marker concentrations (downstream from potential sources). Specific conductance and pH also exhibited statistically significant differences downstream from the effluent outfall, indicating that the effluent travels alongside the eastern bank of the Rio Grande for some distance. Given the lack of correlation with discharge and the human marker being most prevalent in the study area, the source of *E. coli* in the dry season is likely a persistent nonpoint source, such as leaking septic systems or illegal dumping in the area. The lack of

statistically significant spatial differences in *E. coli* concentrations between sampling locations may also indicate that there are no significant contributions of *E. coli* within the study area and that contributions could be originating upstream from the study area.

After calculating the mean daily fluxes, ranking them, and placing them into bins, the average bin scores were used to compare the fluxes of a single constituent across sites and to compare the fluxes of different constituents at a single site. Human and canine sources were the strongest signals observed in the study reach.

With the presence of so many nonpoint sources of fecal bacteria, leaking septic systems, illegal dumping, and general park and trail use may be contributing the most *E. coli* bacteria to the reach during the dry season. The results from this study will help decision makers determine the efficacy of their best management practices and guide new practices to improve water quality in the study reach.

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