

Prepared in cooperation with the Tennessee Duck River Development Agency

Water-Quality Data and *Escherichia coli* Predictions for Selected Karst Catchments of the Upper Duck River Watershed in Central Tennessee, 2007–10



Data Series 1003

U.S. Department of the Interior U.S. Geological Survey

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By Jennifer Murphy, James Farmer, and Alice Layton

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SALLY JEWELL, Secretary

U.S. Geological Survey

Suzette M. Kimball, Director

U.S. Geological Survey, Reston, Virginia: 2016

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Conversion Factors

U.S. customary units to International System of Units

Multiply	Ву	To obtain
	Length	
inch	2.54×10 ⁴	micrometer
inch	25.4	millimeter
foot (ft)	0.3048	meter (m)
mile (mi)	1.609	kilometer (km)
	Area	
square mile (mi ²)	2.590	square kilometer (km ²)
	Flow rate	
cubic foot per second (ft ³ /s)	0.02832	cubic meter per second (m ³ /s)
	Rainfall rate	
inch per year (in/yr)	25.4	millimeter per year (mm/yr)

Temperature in degrees Celsius (°C) may be converted to degrees Fahrenheit (°F) as follows: °F = $(1.8 \times °C) + 32$.

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

 $^{\circ}C = (^{\circ}F - 32) / 1.8.$

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 (μ S/cm at 25 °C).

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

Deoxyribonucleic acid (DNA) concentrations are given in nanograms per microliter (ng/µL).

Bacteria concentrations are given in most probable number of bacteria per 100 milliliters (MPN/100mL).

Bacteria loads are given in million bacteria (1×10⁶).

Abbreviations

AICc	Akaike information criterion
AMLE	adjusted maximum likelihood estimation
ASTM	American Society for Testing and Materials
DNA	deoxyribonucleic acid
LRL	laboratory reporting level
LT-MDL	long-term method detection limit
NWISWeb	National Water Information System Web interface
NWQL	National Water-Quality Laboratory
PCR	polymerase chain reaction
pmol	picomole
rRNA	ribosomal ribonucleic acid
TDEC	Tennessee Department of Environment and Conservation
USGS	U.S. Geological Survey
VIF	variance inflation factor

Water-Quality Data and *Escherichia coli* Predictions for Selected Karst Catchments of the Upper Duck River Watershed in Central Tennessee, 2007–10

By Jennifer Murphy¹, James Farmer², and Alice Layton³

Abstract

The U.S. Geological Survey, in cooperation with the Tennessee Duck River Development Agency, monitored water quality at several locations in the upper Duck River watershed between October 2007 and September 2010. Discrete water samples collected at 24 sites in the watershed were analyzed for water quality, and Escherichia coli (E. coli) and enterococci concentrations. Additional analyses, including the determination of anthropogenic-organic compounds, bacterial concentration of resuspended sediment, and bacterial-source tracking, were performed at a subset of sites. Continuous monitoring of streamflow, turbidity, and specific conductance was conducted at seven sites; a subset of sites also was monitored for water temperature and dissolved oxygen concentration. Multiple-regression models were developed to predict instantaneous E. coli concentrations and loads at sites with continuous monitoring. This data collection effort, along with the E. coli models and predictions, support analyses of the relations among land use, bacteria source and transport, and basin hydrology in the upper Duck River watershed.

Introduction

The Duck River is the principal source of drinking water for several communities in the upper Duck River watershed in central Tennessee (fig. 1; Knight and Kingsbury, 2007). The Tennessee Department of Environment and Conservation (TDEC) lists several streams in the watershed as impaired, however, because of the presence of pathogens or elevated concentrations of bacteria such as *Escherichia coli* (*E. coli*) (Tennessee Department of Environment and Conservation, 2006). Understanding the sources and pathways of these contaminants in the watershed is complicated by the underlying carbonate bedrock and karst landscape. Short groundwater residence times, rapid movement of recharge through solution openings in the bedrock, and efficient connection between surface and groundwater (Knight and Kingsbury, 2007) make identifying specific transport pathways difficult. Furthermore, pathogen and bacteria sources in the watershed have been increasing and changing over time. Population in the vicinity of the study area (Bedford County, fig. 1) increased by 50 percent, from about 28,000 people in 1980 to about 42,000 people in 2005 (U.S. Census Bureau, 1995, 2012), which probably led to an increase in the number and density of septic systems in the area. In addition, the number of cattle in Bedford County increased by about 20 percent, and the number of meat chickens raised per year increased by more than 700 percent between 1987 and 2007 (U.S. Department of Agriculture, 1987, tables 11 and 14; and U.S. Department of Agriculture, 2007, tables 11 and 13). An important first step to protecting the watershed as a drinking-water resource is the collection of water-quality data. To address this need, the U.S. Geological Survey (USGS), in cooperation with the Tennessee Duck River Development Agency, monitored water quality at several locations in the watershed between October 2007 and September 2010.

Purpose and Scope

The purpose of this report is to describe water-quality data collected along the upper Duck River and selected tributaries during 2007–10, and present this information in a format that can be used to support analysis of temporal and spatial patterns of water quality in relation to land use, hydrology, and bacteria sources. Additionally, this report presents regression models developed to predict instantaneous *E. coli* loads and concentrations at several locations in the upper Duck River watershed. This report includes descriptions of methods used for water- and sediment-sample collection and analysis, continuous monitoring of streamflow and selected water-quality characteristics, bacteria-source tracking, and regression modeling of *E. coli* concentrations and loads for sites located in the watershed.

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The data described in this report are available in Farmer and others (2016). For sites with continuous monitoring, the instantaneous unit-values for discharge and water-quality characteristics are available online at USGS National Water Information System Web interface (NWISWeb; http:// waterdata.usgs.gov/tn/nwis/).

Description of Study Area

The upper Duck River watershed study area includes segments of the main channel of the upper Duck River and selected tributaries in the vicinity of Shelbyville, Bedford County, Tennessee (fig. 1). The study area is situated primarily within the Inner Nashville Basin Level IV Ecoregion (Griffith and others, 1997) and is underlain by Ordovician and Mississippian carbonate rock (Knight and Kingsbury, 2007). The area is characterized by hilly terrain with local relief between 60 and 500 feet (ft) and land cover consisting of about 30 percent forest and 50 percent grassland, with developed land and row crops each representing less than 20 percent (Homer and others 2015). Climate for the area is temperate with warm temperatures and moderate-to-high humidity. Climate normals (1982-2010) indicate average annual precipitation in Shelbyville, Tenn., is 56 inches per year (in/yr), delivered relatively evenly throughout the year. Average annual temperature is 59 °F, ranging from an average temperature of 40 °F in the winter to 77 °F in the summer (Arguez and others, 2012).

The area is karstic and characterized by sinkholes, springs, and relatively thin soils, commonly less than 5 ft thick above bedrock (McCroskey, 2003). Groundwater flows primarily through solution openings which develop along bedding planes and joints in the carbonate rock because of physical and chemical weathering. The size and number of solution openings in the Ordovician- and Mississippian-age aquifer decrease with depth, and groundwater flows primarily within 300 ft of the land surface (Brahana and Bradley, 1986).

Drainage areas of the 24 sites range from 0.2 to 481 square miles (mi²; table 1). The sites draining the largest areas are along the main channel of the Duck River. All other sites have drainage areas less than 36 mi², many of which are much smaller. Upstream of the two main-channel Duck River sites there is a low-head dam located at Shelbyville, Tenn., and approximately 27 miles (mi) upstream of Shelbyville is Normandy Lake, a reservoir consisting of a 110-ft-high dam and 17-mi-long impoundment. The reservoir is operated by Tennessee Valley Authority for water-supply purposes, flood damage reduction, and recreational use.

Water-Quality Data Collection

The USGS and the Tennessee Duck River Development Agency collected water-quality samples at 24 sites in the upper Duck River watershed between October 2007 and September 2010 (table 1). Water-quality samples for nutrients, bacteria, and selected inorganic constituents were collected at all sites, and additional analyses, such as bacterial analyses of resuspended sediment, bacterial-source tracking, and determination of anthropogenic organic compounds, were completed at four to eight sites depending on the analysis. Seven sites were continuously monitored for streamflow and other water-quality characteristics.

Field Water Quality, Suspended Sediment, and Bacteria

Water-quality samples collected between the fall of 2007 and spring of 2010 at 24 sites in the upper Duck River watershed were analyzed for field water quality and suspended sediment. Depending on the site and constituent (or property), samples were collected 1 to 42 times during this period (table 2). Measurements of water temperature, specific conductance, and turbidity were taken in the field using a calibrated, handheld, YSI multiparameter sonde. Grab samples were collected from the center of stream above the deepest part of the channel, and water samples were split for specific analyses. Alkalinity and total hardness concentrations were determined by titration in the field. A portable spectrophotometer (Hach DR2800; Hach Company, 2007) also was used in the field to determine concentrations of ammonia (Hach method 10205), boron (Hach method 10061), bromine (Hach method 8016), chloride (Hach method 8113), nitrate (Hach method 10206), nitrite (Hach method 10207) and phosphorus (Hach method 10209). Samples denoted with an "E" remark code indicate the value fell outside of the detection range and were reported as "estimated." Suspended sediment concentration was determined at the USGS Kentucky Water Science Center Sediment Laboratory in Louisville, Kentucky using American Society for Testing and Materials (ASTM) method number D3977-97 (2002).

Water samples also were analyzed for E. coli and enterococci (such as *Escherichia faecium* and *Escherichia faecalis*) concentrations using a defined substrate test method in the laboratory at the USGS Lower Mississippi-Gulf Water Science Center, Nashville, Tenn. For this study, Colilert or Enterolert test kits were used for *E. coli* or enterococci, respectively, along with Quanti-Tray and Quanti-Tray-2000 well trays (IDEXX Laboratories, Inc.). Several tenfold dilution series were made of each sample, which extended the maximum detection range from 2,400 most probable number of bacteria per 100 milliliters (for undiluted wells on the Quanti-Tray-2000) to 240,000 MPN/100 mL. Some of the analyses indicated concentrations higher than the maximum detection range for undiluted or diluted well-trays, and for these cases, concentrations were reported as greater than (>) the maximum detection level. It is USGS standard operating procedure to report fecal indicator bacteria results with two significant figures, as has been done for the data described in this report. For field water-quality, suspended sediment and bacteria results, see SampledWaterQuality Results.xlsx in Farmer and others (2016).

Map identifier (fig. 1)	Station name	USGS station number	Drainage area (mi²)	Latitude (decimal degrees)	Longitude (decimal degrees)
DR1	Duck River at Shelbyville, TN	03597860	425	35.4829	86.4626
DR2	Duck River near Shelbyville, TN	03598000	481	35.4804	86.4992
FC11	Fall Creek near Deason, TN	03598173	16.4	35.5837	86.4880
FC13	Fall Creek below Hurricane Creek near Elbethel, TN	03598177	35.6	35.5644	86.5167
FC9	Benford Creek near Shelbyville, TN	03598169	5.6	35.5717	86.4442
HC10	Hurricane Creek near Elbethel, TN	03598165	12.9	35.5576	86.4983
PC2	Parch Corn Creek at Martin Road near Elbethel, TN	0359816545	2.8	35.5617	86.4855
PC2sp	Unnamed spring near Parch Corn Creek at Frank Martin Road	353344086291101	Unknown	35.5622	86.4864
A1	Alexander Creek at Sudsberry Road near Longview	035981960	2.9	35.6498	86.5063
A3	Alexander Creek near Sanders Cemetery	035981977	17.1	35.6167	86.5336
BigSp	Big Spring near North Fork Creek near Midland Road	035981962	Unknown	35.6228	86.4739
CrkSp	Carrick Spring at Minkslide Road near Shelbyville, TN	0359815605	3.2	35.5547	86.4136
FC10	Fall Creek at Midland Road near Deason, TN	0359816970	14.0	35.5837	86.4872
FC7	Hutton Creek at U.S. Hwy 231 near Deason, TN	0359816920	2.5	35.5951	86.4378
HC2	Hurricane Creek at Minkslide Road near Shelbyville, TN	03598156	0.2	35.5551	86.4125
HC3	Coops Branch at Fairfield Pike near Shelbyville, TN	0359815750	0.8	35.5434	86.4139
HC4	Coops Branch trib at Fairfield Pike near Shelbyville, TN	0359815780	0.4	35.5406	86.4183
HC5	Coops Branch below Minkslide Road near Shelbyville, TN	03598158	1.8	35.5501	86.4278
HC6	Hurricane Creek at Airport Road near Shelbyville, TN	03598157	5.4	35.5542	86.4328
NF3	North Fork Creek at Anderton Road	035981955	5.3	35.6265	86.4644
NF6	North Fork at Big Spring Road	035981965	10.8	35.6226	86.4994
NF7	North Fork Creek at Old Nashville Dirt Road	035981967	17.5	35.5986	86.5378
PC1	Parch Corn Creek at Midland Road near Shelbyville, TN	0359816525	1.4	35.5598	86.4669
PC3	Parch Corn Creek near Hurricane Creek near Elbethel, TN	0359816555	16.8	35.5615	86.5017

Table 1. Site information and summary of data collected.

[USGS, U.S. Geological Survey; mi2, square mile; WQ, water quality; -, data not collected]

Map identifier (fig. 1)	Station name	WQ sampling	Bacterial analysis of resuspended sediment	Bacterial- source tracking	Anthropogenic- organic compounds	Continuous monitoring
DR1	Duck River at Shelbyville, TN	Х		Х		Х
DR2	Duck River near Shelbyville, TN	Х		Х		Х
FC11	Fall Creek near Deason, TN	Х	Х	Х		Х
FC13	Fall Creek below Hurricane Creek near Elbethel, TN	Х	Х	Х		Х
FC9	Benford Creek near Shelbyville, TN	Х	Х	Х	Х	Х
HC10	Hurricane Creek near Elbethel, TN	Х	Х	Х	Х	Х
PC2	Parch Corn Creek at Martin Road near Elbethel, TN	Х	Х	Х	Х	Х
PC2sp	Unnamed spring near Parch Corn Creek at Frank Martin Road	Х	Х	Х	Х	
A1	Alexander Creek at Sudsberry Road near Longview	Х	_	_	_	_
A3	Alexander Creek near Sanders Cemetery	Х	_	-	_	-
BigSp	Big Spring near North Fork Creek near Midland Road	Х	_	-	_	-
CrkSp	Carrick Spring at Minkslide Road near Shelbyville, TN	Х	-	-	-	_
FC10	Fall Creek at Midland Road near Deason, TN	Х	-	-	-	-
FC7	Hutton Creek at U.S. Hwy 231 near Deason, TN	Х	-	-	-	-
HC2	Hurricane Creek at Minkslide Road near Shelbyville, TN	Х	-	-	-	-
HC3	Coops Branch at Fairfield Pike near Shelbyville, TN	Х	-	-	-	-
HC4	Coops Branch trib at Fairfield Pike near Shelbyville, TN	Х	_	-	-	-
HC5	Coops Branch below Minkslide Road near Shelbyville, TN	Х	_	_	_	-
HC6	Hurricane Creek at Airport Road near Shelbyville, TN	Х	-	_	_	-
NF3	North Fork Creek at Anderton Road	Х	_	_	_	_
NF6	North Fork at Big Spring Road	Х	-	_	-	-
NF7	North Fork Creek at Old Nashville Dirt Road	Х	-	-	-	-
PC1	Parch Corn Creek at Midland Road near Shelbyville, TN	Х	-	_	-	-
PC3	Parch Corn Creek near Hurricane Creek near Elbethel, TN	Х	-	-	-	-

 Table 1.
 Site information and summary of data collected. —Continued

 [USGS, U.S. Geological Survey; mi², square mile; WQ, water quality; –, data not collected]

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Table 2.

[Dates are shown as month/day/year. °C, degrees Celsius; μS/cm, microsiemens per centimeter at 25°C; FNU, formazin nephelometric units; mg/L, milligrams per liter; CaCO₃, calcium carbonate; N, nitrogen; P, phosphorus; MPN, most probable number; –, not analyzed]

es)	Boron (mg/L)	ю	4	12	6	11	10	11	5	ю	3	2	3	3	1	2	1	1	2	1	2	3	3	2	2
estimated valu	Hardness (mg/L as CaCO ₃)	28	28	39	38	38	38	42	32	3	3	3	3	3	1	3	2	2	3	2	3	3	3	4	3
of censored or	Alkalinity (mg/L as CaC0 ₃)	10	10	14	14	13	13	15	11	I	I	I	I	I	I	I	I	I	I	I	I	I	I	Ι	I
nples (number	Suspended sediment (mg/L)	9	9	9	7	7	9	9	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I	I
r-quality sar	Turbidity (FNU)	26	27	36	37	36	35	38	25	5	5	5	4	5	1	4	4	4	5	5	5	5	5	9	4
number of water	Specific conductance (µS/cm)	30	30	40	40	39	39	41	22	5	5	9	4	9	1	4	5	5	9	4	5	5	5	7	9
Total r	Water temperature (°C)	I	I	4	4	4	3	3	2	4	4	4	4	4	1	4	3	3	4	4	4	4	4	4	4
1004	Last sample (date)	5/4/2010	5/4/2010	6/9/2009	6/9/2009	6/9/2009	6/9/2009	6/9/2009	6/9/2009	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	6/17/2008	2/9/2009	6/17/2008
.	rırsı sample (date)	10/27/2008	10/23/2008	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	2/12/2008	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007	10/23/2007
	USGS station number	03597860	03598000	03598173	03598177	03598169	03598165	0359816545	353344086291101	035981960	035981977	035981962	0359815605	0359816970	0359816920	03598156	0359815750	0359815780	03598158	03598157	035981955	035981965	035981967	0359816525	0359816555
Mon	indep identifier (fig. 1)	DR1	DR2	FC11	FC13	FC9	HC10	PC2	PC2sp	A1	A3	BigSp	CrkSp	FC10	FC7	HC2	HC3	HC4	HC5	HC6	NF3	NF6	NF7	PC1	PC3

6 Water-Quality Data and *Escherichia coli* Predictions for Selected Karst Catchments of the Upper Duck River

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 Table 2. Date range and sample counts of field water quality, suspended sediment and bacteria data.
 Communication

 [Dates are shown as month/day/year. °C, degrees Celsius; µS/cm, microsiemens per centimeter at 25°C; FNU, formazin nephelometric units; mg/L, milligrams per liter, CaCO,, calcium carbonate; N, nitrogen; P, phosphorus; MPN, most probable number; -, not analyzed]

	(- (0 (((•			
Man			Total n	umber of water	-quality sample	s (number of co	ensored or esti	imated values)	
identifier (fig. 1)	USGS station number	Bromine (mg/L)	Chloride (mg/L)	Nitrate (mg/L as N)	Nitrite (mg/L as N)	Phosphorus (mg/L as P)	Ammonia (mg/L as N)	<i>Escherichia</i> <i>coli</i> (MPN/100mL)	Enterococci (MPN/100mL)
DR1	03597860	-	6	11	11 (5)	10(1)	11	32	30
DR2	03598000	1	6	8	11 (4)	10	11	32 (1)	30
FC11	03598173	6	19	19	17 (5)	18	17	40 (2)	35(2)
FC13	03598177	7	18	18(1)	16 (6)	17 (5)	15	40(1)	35(2)
FC9	03598169	8	18(1)	18 (2)	16(2)	17(1)	15	39 (4)	34
HC10	03598165	7	19	19 (3)	16 (5)	17 (2)	15	39 (2)	34
PC2	0359816545	8	18	18(1)	18 (5)	16(4)	17	41	38(1)
PC2sp	353344086291101	4	12	12	12 (6)	8 (1)	11	32	30(2)
A1	035981960	2	5	5	3	4	3	6 (1)	I
A3	035981977	2	5	5	3	4	2	5 (1)	I
BigSp	035981962	2	9	9	3	9	2	5	Ι
CrkSp	0359815605	2	4	4	3 (1)	4	2	4 (1)	I
FC10	0359816970	2	9	9	2	9	2	6 (3)	I
FC7	0359816920	-		-1	1	1	1	1	I
HC2	03598156	2	4	4(1)	3	4	2	4	I
HC3	0359815750	2	5	4	2	5	1	5	I
HC4	0359815780	2	9	9	2	9	1	4	I
HC5	03598158	2	9	6(1)	3 (1)	9	1	6	I
HC6	03598157	2	5	5	2	5	2	5	I
NF3	035981955	2	5	5	3	4	2	5 (1)	I
NF6	035981965	1	5	5	3	5	2	5 (2)	I
NF7	035981967	2	9	9	3	9	3	6 (1)	I
PC1	0359816525	2	9	6(1)	3 (1)	5 (1)	2	7	I
PC3	0359816555	2	9	6(1)	3 (1)	5 (1)	3	6 (2)	I

Quality-assurance procedures were implemented for constituents analyzed in the field using the Hach DR2800 spectrophotometer. Across all constituents determined using the spectrophotometer, a total of 115 duplicate samples were collected and analyzed. The percent error between each sample and its respective duplicate was calculated using equation 1:

$$\left(\frac{S-D}{mean(S+D)}\right)*100,\qquad(1)$$

where

- *S* is the concentration of the measured sample, and
- *D* is the concentration of the duplicate sample.

These percent errors (equation 1) were then averaged across all sites by water-quality constituent. The mean error was typically within 10 percent for most water-quality constituents, with the exception of boron, turbidity, and *E. coli* (MPN/100 mL), for which the mean percent errors were 43, 17, and 30 percent, respectively.

Standards were also processed in the field (n = 38), and errors were calculated using equation 2:

$$\left(\frac{T-S_{st}}{T}\right)*100, \qquad (2)$$

 Table 3.
 Additional analyses data summary.

[Dates shown as month/day/year; -, not analyzed]

where

- *T* is the reported concentration of the standard, and
- S_{st} is the measured concentration of the standard.

The errors between the measured and reported concentration of the standard were typically within 10 percent with the exception of ammonia where the measured and reported concentrations were different by more than 20 percent for two pairs of analyses. When appropriate, field equipment was assessed and adjusted, and samples were reanalyzed or recollected to resolve errors observed in field determinations.

Bacterial Data from Resuspended Sediment

E. coli and enterococci concentrations were measured in water samples collected at six sites before and after artificial resuspension of bed sediment (table 3). At most of these sites, samples were collected at low flow twice during the spring of 2009. To manually resuspend the bed sediment, the sampler kicked the streambed. Water samples were collected immediately before and after resuspension of the sediment. *E. coli* and enterococci concentrations (most probable number per 100 milliliters) were determined using enzyme substrate assays as described above. For the results of the sediment resuspension tests, see **SedimentBac_Results.xlsx** in Farmer and others (2016).

Map identifier (fig. 1)	USGS station number	Bacterial determinations of resuspended sediment (sampling dates)	Bacterial-source tracking (number of samples collected between 10/23/2008 and 5/4/2010)	Anthropogenic organic compounds (sampling dates)
DR1	03597860	-	16	_
DR2	03598000	-	16	_
FC11	03598173	3/23/2009 6/9/2009	17	-
FC13	03598177	3/23/2009 6/9/2009	17	-
FC9	03598169	3/23/2009 6/9/2009	16	10/27/2008 6/2/0209
HC10	03598165	3/23/2009 6/9/2009	16	6/2/2009
PC2	0359816545	3/23/2009 6/9/2009	16	10/27/2008 6/2/2009
PC2sp	353344086291101	6/9/2009	16	10/27/2008 6/2/2009

Bacterial Source Tracking

For eight sites, molecular bacterial source-tracking techniques were applied to samples collected between the October 10, 2008, and May 4, 2010 (table 3). Approximately 16 samples were collected per site, and the majority of samples were collected prior to the spring of 2009. Bacterialsource tracking is a suite of methods used to identify sources of fecal contamination in water. In this study, Bacteroides spp. were used as indicator microbes because these organisms are found in relatively high proportion in fecal bacterial populations, have high specificity to the host animal (likely related to differences in the digestive track), and demonstrate limited growth outside the host animal (Layton and others, 2006). In particular, Bacteroides spp. 16S ribosomal ribonucleic acid (rRNA) genes were targeted to identify the proportions of human and bovine feces in water. Water samples were analyzed for bacteria using standard membrane filtrations and U.S. Environmental Protection Agency-approved defined substrate culturable methods (U.S. Environmental Protection Agency, 2000; American Public Health Association and others, 2004).

Standard membrane filters were used to filter Bacteroides spp. and E.coli from water samples. Water samples were filtered using 3.0-, 0.45-, and 0.22-micrometer (µm) nitrocellulose filters (Millipore, Inc.). The filters were used for molecular counts of *Bacteroides* spp. and *E.coli*. After filtration, water filters were stored at -80 °C until deoxyribonucleic acid (DNA) extraction. For DNA extraction, 1/4 or 1/2 of the filter was sliced with a sterile razor blade into approximately 2-millimeter (mm) strips, placed into the lysing matrix tube, and analyzed using a FastDNA SPIN Kit for soil (Qbiogene, Inc., n.d.). Subsequent homogenization and nucleic acid extractions were performed according to the FastDNA SPIN protocols (Qbiogene, Inc., n.d.). Following DNA extraction, DNA concentrations were determined and samples were diluted to achieve an approximately 10-nanogram-per-microliter $(ng/\mu l)$ concentration.

Quantitative polymerase chain reaction (PCR) was performed using primers and probes as described in Layton and others (2006) and Knappett and others (2011). Each 25-µL PCR assay consisted of 2.5 µL of sample, 12.5 µL of PCR mix, 5 picomoles (pmol) of the forward primer and reverse primers, and 15 pmol of the probe. Annealing temperatures were 60 °C for the Bacteroides spp. assays and 55 °C for the E. coli rRNA assay. PCR standards consisted of cloned rRNA genes for each assay. All samples were run in triplicate, and for each sample, a fourth PCR assay containing 2.5 µL of sample and 2.5×10^5 copies of a plasmid standard was also run to monitor PCR inhibition. For each assay type, a composite standard curve was used to calculate the number of copies per PCR reaction across the complete dataset as previously described (Bell and others, 2009). Sample data were converted to copies per 100 milliliters of water (copies/100mL)

based on the DNA dilution factor, the volume of DNA extract, the fraction of the filter extracted, and the amount of water filtered. Further information about bacterial-source tracking procedures is provided in Bell and others (2009) and Layton and others (2006).

The bacterial-source tracking assays used two humanspecific genetic markers, the HuBac and HF183, and one bovine-specific marker, BoBac. These markers were identified according to previously described methods for HuBac (Layton and others, 2006), HF183, and BoBac (Surbeck, 2009). The two human *Bacteroides* spp. fecal source tracking assays, HuBac and HF183, were evaluated separately because HuBac and HF183 target different human *Bacteroides* spp. and thus may differ in abundance and host specificity. Source-tracking assays also were completed for total *Bacteroides* spp. (all *Bacteroides* spp. contained in the samples which can include animal sources other than humans and cattle) and *E. coli*. For the results of the bacterial-source tracking analyses see **BacSource_Results.xlsx** in Farmer and others (2016).

Anthropogenic Organic Compounds

Water samples were collected at four sites during 2008–09 for analysis of 69 anthropogenic organic compounds (tables 3 and 4). Water samples were collected as grab samples during low flow and were not filtered prior to analysis. The anthropogenic organic compounds include surfactants, food additives, fragrances, antioxidants, flame retardants, plasticizers, industrial solvents, disinfectants, fecal sterols, polycyclic aromatic hydrocarbons, and high-use domestic pesticides (table 4). Most of these compounds are unregulated in drinking water, and their potential health effects on humans and aquatic wildlife are not well understood. These compounds can serve as an indicator of wastewater contributions to the stream (Heberer, 2002; Ternes and others, 2002).

Anthropogenic organic compounds were analyzed at the USGS National Water-Quality Laboratory (NWQL) in Denver, Colorado (Zaugg and others, 2006). The NWQL uses two detection limits when reporting concentration values. A lower detection limit, referred to as the long-term method detection limit (LT-MDL), controls false positives (reporting a compound as present when it is not in the sample). An upper detection limit, referred to as the laboratory reporting level (LRL), controls false negatives (reporting a compound as not present when it is in the sample) (Childress and others, 1999). If a value falls between the LT-MDL and the LRL, the value is reported as an estimated value (coded as E) and indicates with less than 99-percent confidence that the compound is present in the sample. If a value was not detected at the lower detection limit, then the value is reported as less than (<) the upper detection limit (Childress and others, 1999). For uncensored anthropogenic organic compound results (detects only), see AnthroOrganic Results.xlsx in Farmer and others (2016).

Table 4. List of anthropogenic organic compounds	analyzed
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[USGS, U.S. Geological Survey; CAS, Chemical Abstract Service; µg/L, micrograms per liter; aka, also known as; -, not applicable

Compound	USGS parameter code	CAS registry number	Laboratory reporting level (µg/L)
Metalaxyl	4254	57837-19-1	0.2
2-Methylnaphthalene	30194	91-57-6	0.2
Dichlorvos	30218	62-73-7	0.2
Bromacil	30234	314-40-9	0.8
Bromoform	32104	75-25-2	0.2
Anthracene	34220	120-12-7	0.2
Benzo[a]pyrene	34247	50-32-8	0.2
Diethyl phthalate	34336	84-66-2	0.2
Fluoranthene	34376	206-44-0	0.2
Isophorone	34408	78-59-1	0.2
Phenanthrene	34461	85-01-8	0.2
Pyrene	34469	129-00-0	0.2
Tetrachloroethylene	34475	127-18-4	0.4
1,4-Dichlorobenzene	34571	106-46-7	0.2
Phenol	34694	108-95-2	0.2
Naphthalene	34696	91-20-3	0.2
Chlorpyrifos	38932	2921-88-2	0.2
Pentachlorophenol	39032	87-86-5	0.8
Prometon	39056	1610-18-0	0.2
bis(2-Ethylhexyl) phthalate	39100	117-81-7	2.0
Diazinon	39570	333-41-5	0.2
Atrazine	39630	1912-24-9	0.2
Carbaryl	39750	63-25-2	0.2
3-tert-Butyl-4-hydroxy anisole (BHA)	61702	25013-16-5	0.2
4-Nonylphenol diethoxylate, (sum of all iso- mers) aka NP2EO	61703	_	3.2
4-Nonylphenol monoethoxylate, (sum of all isomers) aka NP1EO	61704	-	1.6
Tris(dichlorisopropyl)phosphate	61707	13674-87-8	0.2
Triclosan	61708	3380-34-5	0.2
5-Methyl-1H-benzotriazole	61944	136-85-6	1.6
Cotinine	61945	486-56-6	0.8
N,N-diethyl-meta-toluamide (DEET)	61947	134-62-3	0.2
beta-Stigmastanol	61948	19466-47-8	1.7
4-tert-Octylphenol monoethoxylate, aka OP1EO	62485	-	1.0
4-tert-Octylphenol diethoxylate, aka OP2EO	62486	_	0.5

 Table 4.
 List of anthropogenic organic compounds analyzed.—Continued

IUSGS U.S.	Geological Survey: CA	S Chemical Abstract Ser	vice: ug/L micrograms	s per liter: aka_also	known as: – not applicable
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Compound	USGS parameter code	CAS registry number	Laboratory reporting level (µg/L)
2,6-Dimethylnaphthalene	62805	581-42-0	0.2
3-beta-Coprostanol	62806	360-68-9	1.6
3-Methyl-1(H)-indole (Skatole)	62807	83-34-1	0.2
4-Cumylphenol	62808	599-64-4	0.2
4-n-Octylphenol	62809	1806-26-4	0.2
4-tert-Octylphenol	62810	140-66-9	0.4
Acetophenone	62811	98-86-2	0.4
Acetyl hexamethyl tetrahydronaphthalene (AHTN)	62812	21145-77-7	0.2
9,10-Anthraquinone	62813	84-65-1	0.2
Benzophenone	62814	119-61-9	0.2
beta-Sitosterol	62815	83-46-5	1.6
Bisphenol A	62816	80-05-7	0.4
Camphor	62817	76-22-2	0.2
Cholesterol	62818	57-88-5	1.6
d-Limonene	62819	5989-27-5	0.2
Hexahydrohexamethylcyclopentabenzopyran (HHCB)	62823	1222-05-5	0.2
Indole	62824	120-72-9	0.2
Isoborneol	62825	124-76-5	0.2
Isoquinoline	62826	119-65-3	0.2
Menthol	62827	89-78-1	0.2
Methyl salicylate	62828	119-36-8	0.2
para-Nonylphenol (total) (branched)	62829	84852-15-3	1.6
Tris(2-butoxyethyl)phosphate	62830	78-51-3	0.2
Tris(2-chloroethyl)phosphate	62831	115-96-8	0.2
Tributyl phosphate	62832	126-73-8	0.2
Triethyl citrate (ethyl citrate)	62833	77-93-0	0.2
Triphenyl phosphate	62834	115-86-6	0.2
3,4-Dichlorophenyl isocyanate	63145	102-36-3	1.6
2,2',4,4'-Tetrabromodiphenylether (PBDE 47)	63147	5436-43-1	0.3
p-Cresol	77146	106-44-5	0.2
Isopropylbenzene	77223	98-82-8	0.2
Carbazole	77571	86-74-8	0.2
Caffeine	81436	58-08-2	0.2
1-Methylnaphthalene	81696	90-12-0	0.2
Metolachlor	82612	51218-45-2	0.2

Continuous Data

Continuous stage, turbidity, and specific conductance data were collected between water years 2008 and 2010 at seven sites (table 1 and 5). Additionally, water temperature was measured at all sites except 03598169 (FC9), and dissolvedoxygen concentrations were measured at 03597860 (DR1). The period of record varied at each site but was typically about 6 months at the tributary sites and about 2 years at the main channel sites (table 5). Measurement intervals ranged from 15 to 60 minutes depending on site and characteristic measured. Continuous measurements of stream stage were collected at all seven sites using a submersible pressure transducer. Discharge was measured periodically across a range of flow conditions, and a stage-discharge rating curve was developed for each site using standard USGS protocols (Turnipseed and Sauer, 2010; Rantz and others, 1982). The rating curves were applied to the continuous gage height record to determine a continuous discharge record at each site. At 03597860 (DR1), discharge measurements were not made when gage height was greater than 11.31 ft; therefore, no discharge values are available for gage heights above this level. A YSI multi-parameter sonde (YSI 6136) was used to collect instantaneous measurements of turbidity, water temperature, specific conductance, and dissolved oxygen. Unit values for these water-quality characteristics and discharge can be retrieved for each site from USGS NWISWeb (http://waterdata.usgs.gov/tn/nwis/nwis).

Escherichia coli Concentration and Load Predictions

To predict instantaneous E. coli concentration and load, linear regression models were developed using measured E. coli concentrations (MPN/100mL) and continuously monitored characteristics, such as discharge, turbidity, or specific conductance. If continuously monitored turbidity data were not available for an E. coli sample, the turbidity measured using a handheld probe at the time of sample collection was used. The implementation of LOADEST (Runkel and others, 2004; Runkel, 2013) for the R statistical software program (rloadest, Lorenz and others, 2013) was used to develop a regression model for each of the sites having continuously monitored data. Model coefficients were estimated using Adjusted Maximum Likelihood Estimation (AMLE). The AMLE provides maximum likelihood estimates of regression model coefficients, corrects for bias both in the model coefficients and model estimates, and can be useful when data are censored (Runkel and others, 2004).

Model development proceeded in three steps. First, based on the bivariate relationships between *E. coli* (log-transformed and untransformed) and possible explanatory continuous variables (including turbidity, flow, specific conductance, water temperature, dissolved oxygen, and time) a preliminary set of explanatory variables were selected for each site. Second, in rloadest, initial models for instantaneous *E. coli* load were

Table 5. Date range of continuously monitored water-quality characteristics for this study.

[Unit-value continuous data can be retrieved from the U.S. Geological Survey (USGS) National Water Information System Web interface (NWISWeb) (http://waterdata.usgs.gov/nwis) for these sites. Certain sites and characteristics may have period of records that extend beyond the range presented here. Dates are shown as month/day/year. –, not collected. Units of continuous data: discharge (cubic feet per second), turbidity (formazin nephelometric units), specific conductance (microsiemens per centimeter at 25 degrees Celsius), water temperature (degrees Celsius), dissolved oxygen (milligrams per liter)]

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USGS station number (Map identifier)	Discharge	Turbidity	Specific conductance	Water temperature	Dissolved oxygen
03597860 (DR1)	10/1/2007– 9/30/2010	11/26/2008– 9/30/2010	10/1/2007– 9/30/2010	10/1/2007– 9/30/2010	10/1/2007– 9/30/2010
03598000 (DR2)	10/1/2007– 9/30/2009	3/5/2009– 9/30/2009	3/4/2009– 9/30/2009	3/4/2009– 9/30/2009	-
03598173 (FC11)	12/10/2008– 6/30/2009	12/10/2008– 6/30/2009	12/10/2008– 6/30/2009	12/10/2008– 6/30/2009	-
03598177 (FC13)	12/10/2008– 6/21/2009	12/10/2008– 6/30/2009	12/11/2008– 6/30/2009	12/10/2008– 6/30/2009	-
03598169 (FC9)	12/5/2008– 6/30/2009	12/5/2008– 6/16/2009	12/5/2008– 6/15/2009	-	-
03598165 (HC10)	12/5/2008– 6/30/2009	12/5/2008– 6/17/2009	12/5/2008– 6/17/2009	12/5/2008– 6/17/2009	-
0359816545 (PC2)	11/26/2008– 6/30/2009	11/26/2008– 6/23/2009	12/1/2008– 6/22/2009	12/1/2008– 6/22/2009	-

calibrated using these explanatory variables and the best working model was selected based on model diagnostics, residual plots, the corrected Akaike Information Criterion (AICc), and additional bias statistics comparing the observed and estimated loads. Third, explanatory variables were added or removed from the "working" model in an effort to optimize model diagnostics and AICc; a final model was selected from these iterations. The bias statistics include the load bias, B_p , in percent (equation 3)

$$B_{p} = 100 \left(\frac{\sum_{k=1}^{N} (\hat{L} - L)}{\sum_{k=1}^{N} L} \right),$$
(3)

where

- \hat{L} is estimated load,
- *L* is observed load, and
- *N* is the number of observations in the calibration dataset;

the partial load ratio (PLR, equation 4)

$$PLR = \frac{\sum_{k=1}^{N} \hat{L}}{\sum_{k=1}^{N} L};$$

(4)

and the Nash-Sutcliffe Efficiency Index (E, equation 5)

$$E = 1 - \frac{\sum_{k=1}^{N} \left(L - \hat{L}\right)^2}{\sum_{k=1}^{N} \left(L - \bar{L}\right)^2} , \qquad (5)$$

where

 \overline{L} is the mean of the observed loads.

The goal was to optimize the working model so that B_p was never greater than 25 percent, *PLR* was between 0.5 and 2, and *E* was between 0 and 1 (Runkel and others, 2004).

All models were built using the logarithm of instantaneous *E. coli* load as the response variable, and the explanatory variables varied by site (table 6). For an explanatory variable to be included in a model, it had to lower the AICc and be statistically significant. Once the final *E. coli* load model was selected, the model was also parameterized using *E. coli* concentrations (MPN/100mL) given the same explanatory variables. To assess bias for *E. coli* concentration estimates, the

bias statistics described above were also used though observed and estimated bacteria concentration were substituted in place of load.

A few of the sites required special considerations during model development. For site 03597860 (DR1), because of the configurations of the channel, discharge was measured only when the gage height was below 11.31 ft. However, samples were collected when the water level was above this height, and therefore, some E. coli concentrations at this site had no associated flow. For these samples, flow was assigned the maximum observed discharge value for the study period (597 cubic feet per second $[ft^3/s]$). Thus, for site 03597860 (DR1), the estimated loads represent the total load when flow is less than 597 ft³/s but only a partial load when flow exceeds 597 ft³/s. At this site, the discharge record was also truncated at 597 ft³/s prior to predicting *E. coli* loads and concentrations. In addition, sites 03598000 (DR2) and 03598169 (FC9) had a few right-censored E. coli values (n=1 and n=2, respectively), and because these samples represented a small proportion of the data, these values were assigned to the censoring level prior to model development.

All final models used flow and turbidity terms, and some included seasonal, quadratic flow, quadratic turbidity, or specific conductance terms (table 6). Even though flow, turbidity and specific conductance were moderately correlated at these sites, variance inflation factors (VIF) indicate limited multicol-linearity (VIFs all < 4, except 03598000 (DR2) where VIFs were 4.2 and 5.3 for the natural log of turbidity and natural log of flow, respectively) and only small increases in the standard errors of the model coefficients. Furthermore, because these models are being used for prediction and not inference, correlated variables are not necessarily problematic. No final model was selected for site 03598165 (HC10) because of inadequate model diagnostics.

For all sites, the *E. coli* concentration models have smaller \mathbb{R}^2 values than the load models, indicating the *E. coli* load models have greater explanatory power (table 6). The bias of estimated loads and concentrations was generally less than 15 percent and always less than 20 percent. For several of the models, when the regression equation was parameterized using *E. coli* concentration, the flow term was no longer statistically significant (table 6). Because the *E. coli* load and concentration models were calibrated using data that represent only a short period of time (at most 2 years), the models presented in table 6 should not be used for predictions outside of the calibration period.

For the predicted instantaneous *E. coli* loads and concentrations, along with associated upper and lower 95-percent prediction intervals, see **PredictedEcoli_03597860.xlsx**, **PredictedEcoli_03598000.xlsx**, **PredictedEcoli_03598173. xlsx**, **PredictedEcoli_03598177.xlsx**, **PredictedEcoli_03598169. xlsx**, and **PredictedEcoli_0359816545.xlsx** in Farmer and others (2016).

Map	NSGS	Calibration		Bacidual	R-constad	Load or	Partial	Nash-Sutcliffe
identifier (fig. 1)	· station number	sample count	Model formula	variance	(percent)	concentration bias (percent)	load ratio	efficiency index (E)
			Load models					
¹ DR1	03597860	30	ln(L) = -1.308 + 1.247*ln(Turb) + 2.575*ln(Q) + 2.957*ln(Q) + 2.957*ln(Q)2 + 2.174*ln(SpC)	0.48	95.0	14.350	1.14	0.85
DR2	03598000	27	ln(L) = 8.735 + 1.427*ln(Turb) + 0.203*ln(Turb)2 + 0.964*sin(Time) + 0.402*cos(Time) + 1.077*ln(Q)	0.61	96.4	13.81	1.14	66.0
FC11	03598173	31	$\ln(L) = 7.9631 + 1.0661*\ln(Turb) + 0.9775*\ln(Q)$	0.96	89.1	17.21	1.17	0.74
FC13	03598177	32	$\ln(L) = 6.3277 + 0.8962*\ln(Turb) + 1.3130*\ln(Q)$	0.66	94.8	18.30	1.18	0.86
FC9	03598169	32	ln(L) = 12.4913 + 2.2993*ln(Turb) + -0.3349*ln(Turb)2 + 0.7858*ln(Q)	0.66	96.2	10.78	1.11	0.86
PC2	0359816545	33	$\ln(L) = 7.7974 + 1.1492*\ln(Turb) + 0.8007*\ln(Q)$	1.02	85.2	2.82	1.03	0.84
			Concentration models					
DR1	03597860	30	ln(C) = -10.311 + 1.247*ln(Turb) + 1.575*ln(Q) + 2.957*ln(Q) 2.957*ln(Q) + 2.174*ln(SpC)	0.48	93.0	14.34	1.14	0.85
² DR2	03598000	27	ln(C) = 5.537 + 1.426*ln(Turb) + 0.203*ln(Turb)2 + 0.964*sin(Time) + 0.402*cos(Time) + 0.077*ln(Q)	0.61	91.0	9.78	1.10	66.0
² FC11	03598173	31	$\ln(C) = 4.76583 + 1.06614*\ln(Turb) + -0.02246*\ln(Q)$	0.96	62.5	-8.82	0.91	0.24
FC13	03598177	32	$\ln(C) = 3.1304 + 0.8962*\ln(Turb) + 0.3130*\ln(Q)$	0.66	81.5	14.60	1.15	0.63
² FC9	03598169	32	ln(C) = 9.2940 + 2.2993*ln(Turb) + -0.3349*ln(Turb)2 + -0.2142*ln(Q)	0.66	89.7	-9.92	0.90	0.82
² PC2	0359816545	33	ln(C) = 4.6001 + 1.1492*ln(Turb) + -0.1993*ln(Q)	1.02	46.1	4.54	1.05	0.54
i Model i	for DR1 is for parti-	al loads when di	scharge is greater than 597 cubic feet per second.					

 $^{2}\mathrm{Flow}$ term (ln(Q)) in concentration model is not statistically significant at this site.

 Table 6. Escherichia coli load and concentration models.

14 Water-Quality Data and *Escherichia coli* Predictions for Selected Karst Catchments of the Upper Duck River

All sample data, results, and predictions described in this report are available as Microsoft Excel files (version 2013) and can be downloaded from Farmer and others (2016). Each workbook file contains a single dataset. Table 7 shows the available data and to determine the type and amount of data available for each site, see tables 1, 2, 3 and 5 in the main text of this report or the metadata provided with the data. Continuous data are available at USGS NWISWeb (http://waterdata.usgs.gov/tn/nwis/nwis).

Summary

This report describes the study area, sample collection, and processing methods for water-quality data from 24 sites located in the upper Duck River watershed. Data tables contain the processed water-quality data at all sites, including field water quality, suspended sediment concentration and bacteria concentration. Results from additional analyses at a subset of sites, including determination of bacteria concentration in resuspended sediment, bacterial source tracking, and determination of anthropogenic organic compounds; and predictions of instantaneous Escherichia coli concentrations and load at selected sites are available in Farmer and others (2016). Protecting the watershed as a drinking-water resource begins with monitoring water quality, and the data and methods presented in this report support analyses of the relations among land use, bacteria source and transport, and basin hydrology in the watershed.

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File name	Description
Field w	vater quality, suspended sediment, and bacteria
SampledWaterQuality_Results.xlsx	Results of field water quality, suspended sediment and bacteria for all 24 sites
	Additional analyses
AnthroOrganic_Results.xlsx	Anthropogenic organic determinations for four sites
SedimentBac_Results.xlsx	Bacterial analyses of resuspended sediment at six sites
BacSource_Results.xlsx	Bacterial-source tracking results for eight sites
	Regression equations
RegModels.xlsx	Regression equations used for predicting instantaneous loads and concentrations
Predicted instantaneous loads and concentration	ns, including 95- percent prediction intervals, and associated discharge for selected sites
Predicted_03597860.xlsx	Predicted values for site 03597860, map identifier DR1
Predicted_03598000.xlsx	Predicted values for site 03598000, map identifier DR2
Predicted_03598173.xlsx	Predicted values for site 03598173, map identifier FC11
Predicted_03598177.xlsx	Predicted values for site 03598177, map identifier FC13
Predicted_03598169.xlsx	Predicted values for site 03598169, map identifier FC9
Predicted_0359816545.xlsx	Predicted values for site 0359816545, map identifier PC2

Table 7. Data in Farmer and others (2016) data release.

Farmer, J., Layton, A., and Murphy, J., 2016, Water-quality datasets and *E. coli* predictions for selected streams in the upper Duck River Watershed, central Tennessee, 2007–10: U.S. Geological Survey [ScienceBase] data release, accessed February 12, 2016, at http://dx.doi.org/10.5066/F7445JKC.

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