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OKLAHOMA DEPARTMENT OF ENVIRONMENTAL QUALITY AND
U.S. ENVIRONMENTAL PROTECTION AGENCY

Reconnaissance of Surface-Water Quality and Possible Sources of Nutrients and Bacteria in the Turkey Creek Watershed, Northwest Oklahoma, 2002–2003

Scientific Investigations Report 2004–5039




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

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By Carol J. Becker

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U.S. Department of the Interior
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Conversion Factors and Datum

Multiply	By	To obtain
	Length	
inch (in.)	2.54	centimeter (cm)
foot (ft)	0.3048	meter (m)
mile (mi)	1.609	kilometer (km)
	Area	
square mile (mi^2)	2.590	square kilometer (km^2)
	Volume	
gallon (gal)	3.785	liter (L)
	Flow rate	
cubic foot per second (ft^3/s)	0.02832	cubic meter per second (m^3/s)
	Mass	
ton, short (2,000 lb)	0.9072	metric ton

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

°F = (1.8 × °C) + 32

Vertical coordinate information is referenced to the North American Vertical Datum of 1988 (NAVD 88).

Horizontal coordinate information is referenced to the North American Datum of 1988 (NAVD 88).

Altitude, as used in this report, refers to distance above the vertical datum.

Specific conductance is given in microsiemens per centimeter at 25 degrees Celsius (µS/cm at 25˚C).

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

Reconnaissance of Surface-Water Quality and Possible Sources of Nutrients and Bacteria in the Turkey Creek Watershed, Northwest Oklahoma, 2002-2003

By Carol J. Becker

Abstract

The U.S. Geological Survey in cooperation with the Oklahoma Department of Environmental Quality and the U.S. Environmental Protection Agency investigated the distribution of surface-water quality and possible sources of nutrients and *Escherichia coli* bacteria to surface water in Turkey Creek, which flows about 70 miles through mostly rural agricultural areas in northwest Oklahoma. Results show that discharge on the main stem of Turkey Creek increased during low-flow conditions from an average of 5.4 cubic feet per second at the upper most site to 39 cubic feet per second at the lower most site in the watershed, indicating that Turkey Creek gains water from ground-water discharge. A portion of the increase in stream discharge may be from discharges of treated effluent from city sewage lagoons. However, the volume and frequency of discharges are unknown.

Surface-water-quality samples show that specific conductance ranged from 1,180 to 1,740 microsiemens per centimeter at 25 degrees Celsius during low-flow conditions and in general, decreased downstream with site 1 or site 2 having the largest measurement and site 5 having the lowest. The pH values were slightly alkaline and ranged from 6.8 to 8.5 with a median of 8.2. Dissolved oxygen ranged from 9.3 to 15.9 milligrams per liter in samples collected in the months of November, February, and March and ranged from 5.3 to 13.9 milligrams per liter in samples collected in the months of June, July, and August.

Surface-water-quality samples show that the median concentrations of nitrite plus nitrate as nitrogen (1.16 milligrams per liter) and total phosphorus (0.275 milligram per liter) are larger than the average median concentrations of 0.35 and 0.083 milligram per liter, respectively, calculated from water-quality sites in Oklahoma and part of Arkansas (excluding sites in the Ozark Highland and the Ouachita Mountains ecoregions) having similar stream orders and stream slopes. Concentrations of nitrite plus nitrate as nitrogen increased slightly in the winter months and decreased in the summer months, whereas, concentrations of total phosphorus and orthophosphate as phosphorus tended to increase during the summer months and decrease in the winter months. During high-flow conditions total phosphorus increased 7.7 times above the average concentration of 0.261 milligram per liter in low-flow samples. Orthophosphate concentrations increased 3.5 to 4 times during high-flow conditions.

Almost all low-flow samples showed $\delta^{15}N$ values between 4 and 10 parts per thousand, above the range for atmospheric nitrogen and synthetic fertilizer and below the range for animal waste. These samples may represent a mixture of nitrate from these two sources and other sources enriched with $\delta^{15}N$, such as soils and plants.

Results of the bacterial source tracking indicated that the two source groups having the greatest number of ribopattern matches with surface-water isolates were the cattle group, 53 isolates or 23.5 percent, and the human group, 41 isolates or 18.2 percent. Fewer surface-water isolates matched the deer and horse groups, 8.0 percent and 3.5 percent, respectively. About 43 percent or 96 surface-water isolates were not matched to any source group.

Introduction

Turkey Creek is located in northwest Oklahoma. From the headwaters in Alfalfa County, Oklahoma, Turkey Creek flows about 70 miles through mostly rural agricultural areas until it reaches the Cimarron River (fig. 1). Turkey Creek is one of many water bodies in Oklahoma, and across the Nation, that is stressed by an increase in human development, agriculture, and stricter controls on water quality. Two water-quality issues of concern for Turkey Creek have been the nutrients—nitrogen and phosphorus—and coliform bacteria. Elevated concentrations of nitrogen and phosphorus compounds were measured by the Oklahoma Conservation Commission (OCC) during ambient water-quality monitoring at six sites on Turkey Creek and tributaries from November 1997 to August 1998 (Oklahoma Conservation Commission, written commun., 2000). Concentrations of nitrite plus nitrate as nitrogen, total nitrogen, and total phosphorus were larger than most other streams in Oklahoma having similar hydrologic characteristics (Haggard and others, 2003). Densities of fecal and strep coliform bacteria above the Oklahoma standard of 200 colonies (per 100 milliliters) for primary contact recreation also were frequently measured by the OCC. Fecal coliform and fecal streptococci bacteria are used as indicators of fecal contamination in water and are monitored for the possible presence of disease-causing pathogens.

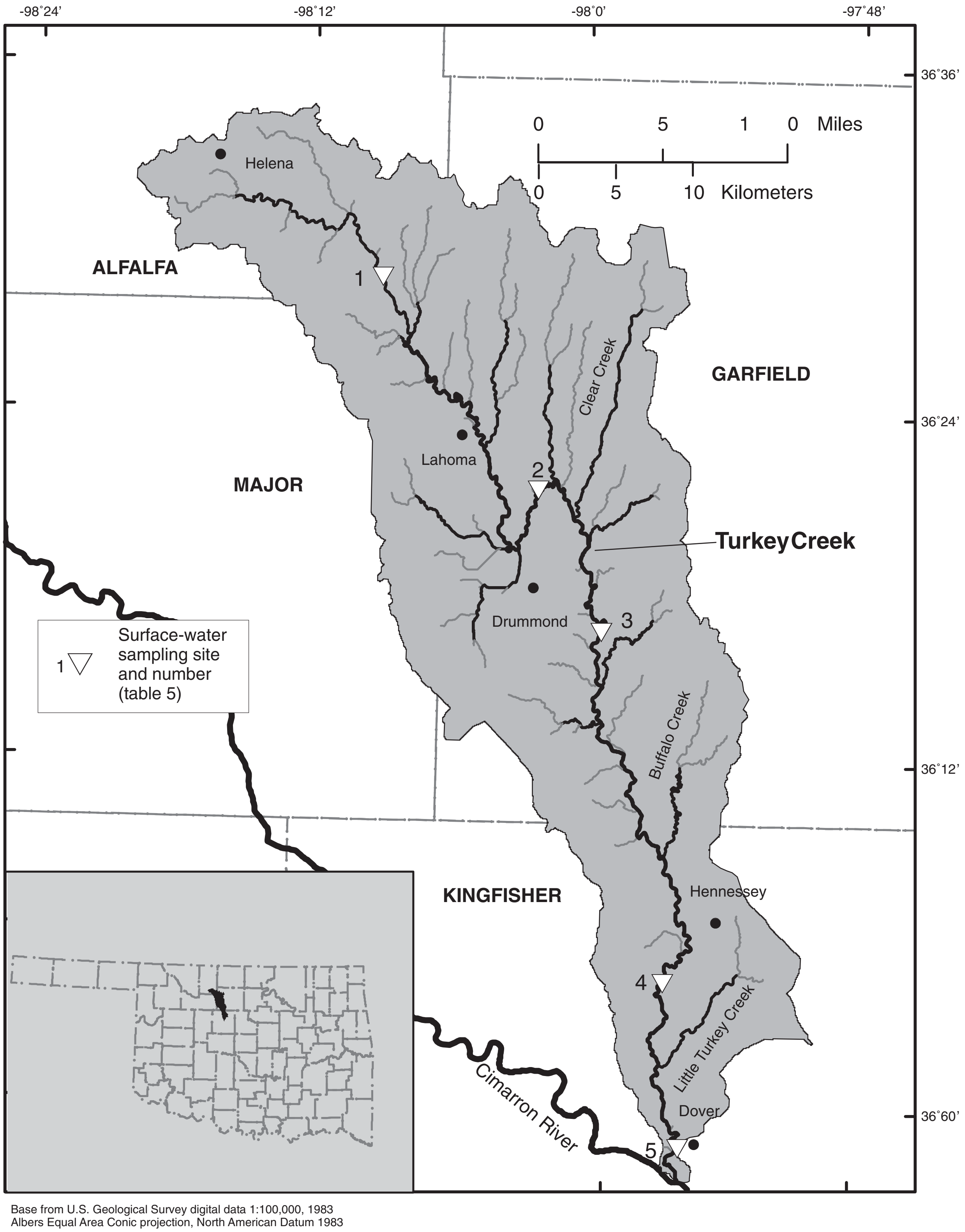


Figure 1. Surface-water quality sampling sites on Turkey Creek in the Turkey Creek watershed, northwest Oklahoma.

There is a need to better understand the sources of nutrients and bacteria as related to land use activities in the Turkey Creek watershed. As a result, the U.S. Geological Survey in cooperation with the Oklahoma Department of Environmental Quality and the U.S. Environmental Protection Agency investigated the distribution of surface-water quality and possible sources of nutrients and *Escherichia coli* (*E. coli*) bacteria to surface water in the Turkey Creek watershed. The objectives of the study were to investigate 1) the seasonal variations of the nutrients-nitrogen and phosphorus-in surface water, 2) the contributions of fertilizers to nitrogen in surface water, and 3) the possible sources of bacteria in surface water to match isolates to source groups using a microbial source tracking system called the DuPont Qualicon RiboPrinter® (Dupont Qualicon, 2003).

Purpose and Scope

This report describes the surface-water-quality data collected in the Turkey Creek watershed from June 2002 to June 2003, and the results of stable nitrogen isotopes to determine possible sources of nutrients and a bacterial source tracking method used to identify possible sources of *E. coli* bacteria to surface water. More than 40 surface-water samples were collected eight times over a 12-month period. The water properties—specific conductance, pH, water temperature, and dissolved oxygen—and stream discharge were measured at five sites on the main stem of Turkey Creek (fig. 1). Additionally, nitrite as nitrogen, nitrite plus nitrate as nitrogen, ammonia as nitrogen, ammonia plus organic nitrogen as nitrogen, total phosphorus, and orthophosphate as phosphorus were analyzed. The stable nitrogen isotopes ^{15}N and ^{14}N in nitrate nitrogen also were analyzed to determine the seasonal contributions of fertilizer to nitrogen in surface water. At four of the five sites, surface-water also was collected for the isolation of *E. coli* bacteria during three of the eight sampling episodes. A bacterial source tracking method was then used to identify possible sources by comparing the relatedness of DNA fragments from ribosomal genes, referred to as ribopatterns, to *E. coli* bacteria cultured from cattle, deer, horses, and effluent from septic tanks and city wastewater lagoons in the watershed.

Acknowledgments

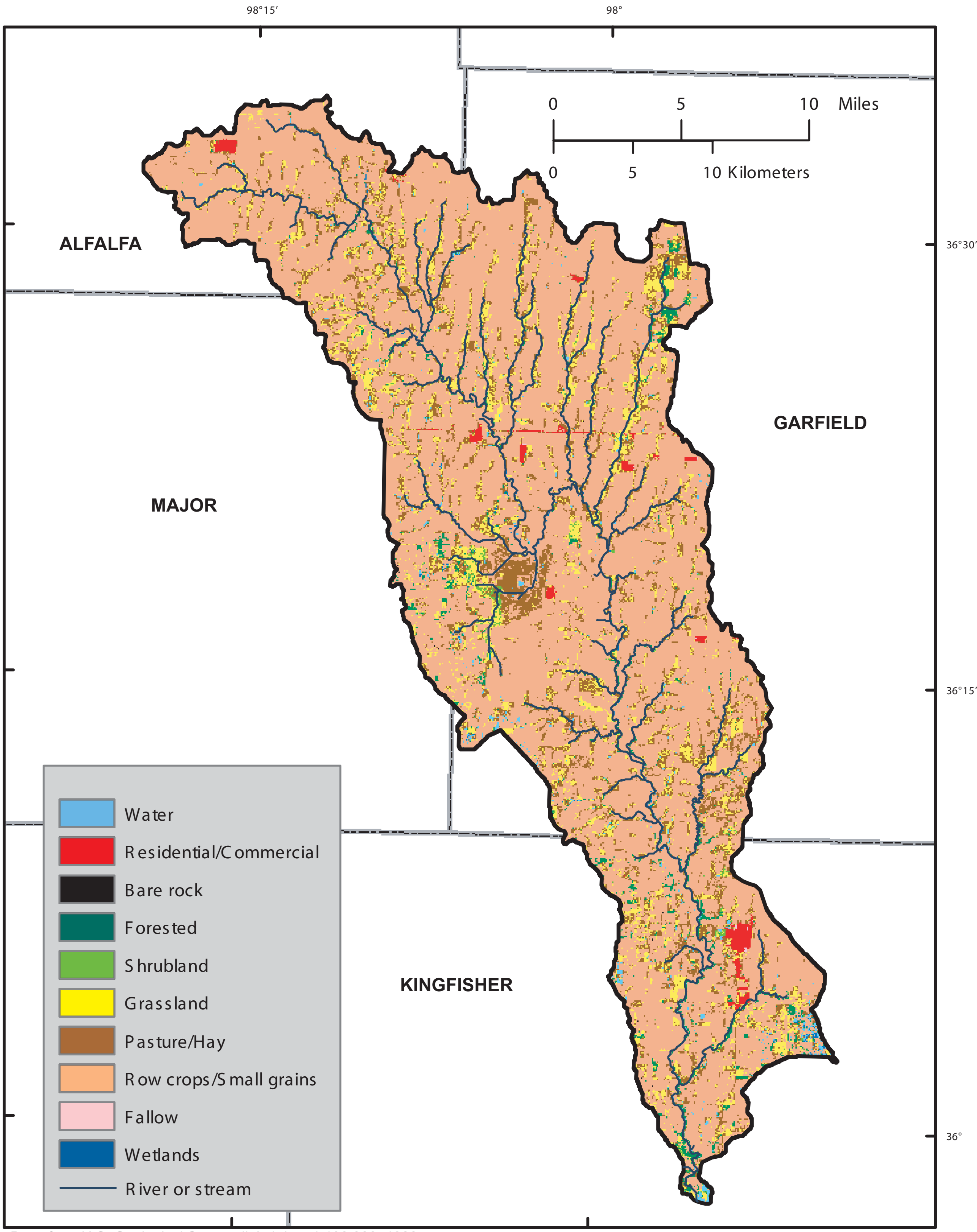
The author thanks Mike Houts, Oklahoma Department of Environmental Quality; Steve Hutchins and Yolanda Olivas, U.S. Environmental Protection Agency in Ada, Oklahoma; Doug Vandagriff, City of Hennessey; Scott Nolting, City of Lahoma; Mike Bowers, City of Helena; Ronald Welsh and Laura Dye, OSU Animal Disease Diagnostic Laboratory, Stillwater, Oklahoma; Eddie Pryor, and the many residents and livestock owners in the Turkey Creek watershed who graciously assisted with this project, and Lee Ann Alf, U.S. Geological Survey.

Description of Study Area

Turkey Creek drains about 376 square miles of rural Oklahoma, starting just north of Helena in Alfalfa County and flowing south to the confluence with the Cimarron River (fig. 1). The area is situated in the Central Redbed Plains, which is characterized by gently, rolling hills and broad, flat plains (Curtis and Ham, 1979). Land use in the watershed is primarily agricultural; land use/cover information for 1992 shows that about 76 percent of land in the watershed was used for row crops and small grains and about 11 percent was used for pasture and hay (fig. 2 and table 1) (U.S. Environmental Protection Agency, 2000). Wheat and rye are the predominant crops grown in the area with minor amounts of alfalfa (Keith Bovers, Oklahoma State University, Agricultural Extension Director, Kingfisher County, oral commun., 2003). The average annual precipitation for a 30-year period (1961-90) for the north-central part of the State ranges from 28 to 32 inches (Johnson and Duchon, 1994). The total precipitation measured at Lahoma in Garfield County was 32.4 inches from June 2002 to June 2003 (Oklahoma Climatological Survey, 2004).

Nutrients in the Watershed

Important sources of nutrients in the Turkey Creek watershed are commercial fertilizers, animals including humans, vegetation, soils, and precipitation. The most commonly used nitrogen fertilizers in the watershed are urea, anhydrous ammonia, and ammonium nitrate. Phosphorus also may be applied in combination with ammonium nitrate. Commercial fertilizers are applied to the ground when wheat is planted in September through October and then again in January through February to promote seed production (Keith Bovers, Oklahoma State University, Extension Director, Kingfisher County, oral commun., 2003). The amount of nitrogen applied in commercial fertilizers is estimated to average between 18.4 to 26.3 tons per square mile per year (tons/mi^2/yr) and phosphorus 1.4 to 3.1 tons/mi^2/yr in Alfalfa, Garfield, Kingfisher, and Major counties (Storm and others, 2000, table 11). Estimates were calculated using application rates on crop and pastureland and average fertilizer sales in the four counties from 1987 to 1996. Estimates of the total amount of nitrogen and phosphorus from animal wastes available to crop and pasture land for Alfalfa, Garfield, Kingfisher, and Major counties averaged from 0.2 to 0.5 ton/mi^2/yr of nitrogen and 0.1 to 0.3 ton/mi^2/yr of phosphorus (Storm and others, 2000, table 14). Nitrogen also occurs naturally in soils from the decomposition of organic material and from precipitation, which deposits about 1.3 to 1.7 tons/mi^2/yr in the watershed (Mueller and Helsel, 1996). Phosphorus also is found in phosphorus-containing rocks, fecal waste from ducks and cattle, fallout from the atmosphere, and tree leaves (U.S. Environmental Protection Agency, 1986). Estimates of nutrient input from human waste in the watershed were unavailable.



Base from U.S. Geological Survey digital data 1:100,000, 1983
Albers Equal Area Conic projection, North American Datum, 1983

Figure 2. Land use in the Turkey Creek watershed, 1992 (U.S. Environmental Protection Agency, 2000).

Table 1. Land use in the Turkey Creek watershed, 1992, (U.S. Environmental Protection Agency, 2000)

Land use	Percent of land use in watershed
Water	0.68
Residential/commercial	0.78
Bare rock	0.01
Forested	1.78
Shrubland	0.60
Grassland	9.35
Pasture/hay	10.95
Row crops/small grains	75.63
Urban grasses	0.02
Wetlands	0.19

Bacteria in the Watershed

Possible contributors of bacteria to the watershed are humans from septic tank effluent, cattle, and effluent from wastewater lagoons. Additionally, horses, dogs, cats, deer, waterfowl, and other small mammals are possible sources of bacteria and nutrients to the watershed. Waterfowl may be a significant source during the fall migration season. About 15,200 people and 9,300 cattle were estimated to reside in the Turkey Creek watershed. These estimates were calculated from the 2001 census county estimates (U.S. Census Bureau, 2003), the livestock county estimates for January 1, 2002, (U.S. Department of Agriculture-National Agricultural Statistics Service, 2003), and the percentage of county area in the watershed. The population estimate includes a correctional center in Helena, which houses about 840 inmates (Oklahoma Department of Corrections, 2003). Cattle are present throughout the watershed and have access to Turkey Creek and tributaries in many areas. There is only one permitted confined animal facility in the basin, a dairy with about 400 dairy cows located one mile east of Lahoma.

The towns of Dover, Drummond, Helena, Hennessey, and Lahoma provide sewage utilities within city limits for residents and businesses. These towns use primary sewage treatment; sewage is pumped into one or more lagoons where solids settle out and the organic material decomposes. Drummond, Hennessey, and Lahoma are permitted by the State to discharge treated effluent into Turkey Creek, when necessary; whereas, Dover and Helena are permitted for land application of the sewage solids on alluvial deposits along Turkey Creek (Patrick Rosch, Oklahoma Department of Environmental Quality, oral commun., 1999). Households and businesses outside of city limits use private septic tank systems for wastewater disposal.

Methods

Surface-water data collection included measurement of water properties and stream discharge, and water-quality sampling and analysis for nutrients. Nutrient sources were investigated with stable isotope techniques. Bacterial source tracking was used to identify possible sources of bacteria to surface water.

Surface-Water-Data Collection

Five surface-water-quality collection sites (sites 1-5) (fig. 1) were chosen on the main stem of Turkey Creek and surface-water samples were collected eight times between June 2002 and June 2003. Water properties including specific conductance, pH, water temperature and dissolved oxygen, and stream discharge were measured on site according to procedures described by Wilde and Radtke (1998). Surface-water-quality samples were analyzed for nutrient compounds in filtered and unfiltered samples and the stable nitrogen isotopes ^{15}N and ^{14}N in nitrate nitrogen. Nitrite as nitrogen, nitrite plus nitrate as nitrogen, ammonia as nitrogen, and orthophosphate as phosphorus, were analyzed in filtered samples. Ammonia plus organic nitrogen and total phosphorus were analyzed in unfiltered samples. All samples were collected during low-flow conditions except during high-flow conditions in August 2002 when runoff occurred during heavy rain. Sampling techniques are described in Webb, Radtke, and Iwatsubo (2003). Table 2 shows the constituents analyzed in this study, the analyzing laboratory, laboratory method, minimum reporting level, and method references. Water-quality samples were collected and processed as described in Wilde and others (1999, 2003).

Quality assurance practices were implemented to maintain consistency with sampling protocols throughout the study and to assure clean practices. Ten percent of all water samples were quality-control samples consisting of duplicate samples. One replicate sample was collected sequentially for nitrogen isotopes and analyzed by the USGS National Water Quality Laboratory in Lakewood, Colorado.

Nutrient Sources

Stable nitrogen isotopes were used to determine possible seasonal contributions of fertilizer to nitrogen in surface water. Nitrogen has two stable isotopes, ^{15}N and the more abundant ^{14}N. The $\delta^{15}N$ in nitrate in surface-water samples was compared to $\delta^{15}N$ in nitrate in common sources of nitrogen (Clark and Fritz, 1997). The units used to report nitrogen isotopes ratios in a sample to those of a standard material are expressed as delta (δ) values in parts per thousand, denoted as permil (‰). The δ values for nitrogen stable-isotopic ratios discussed in this report, referred to as $\delta\ ^{15}N$, are referenced to standard atmospheric nitrogen, referenced to National Bureau of Standards, NBS-14 nitrogen gas (Fritz and Fontes, 1980, p. 16). Stable-isotopic ratios are computed as follows (Kendall and Caldwell, 1998):

$$\delta\ ^{15}N = \left[\frac{\left(\frac{^{15}N}{^{14}N} \right)_{Sample}}{\left(\frac{^{15}N}{^{14}N} \right)_{Standard}} - 1 \right] \mathrm{X} \quad 1{,}000$$

where $\frac{^{15}N}{^{14}N}$ is the ratio of the heavier, less abundant isotope to the lighter isotope.

Bacterial Source Tracking

Bacterial source tracking, also referred to as ribotyping, was performed to identify possible animal sources of *E. coli* bacteria to surface water. Bacterial source tracking relies on the assumption that genetically differentiable strains of *E. coli* are truly host specific. This assumption remains under investigation. Accordingly, the results should be characterized as experimental in nature. The ribotyping of ribosomal RNA genes from *E. coli* bacteria isolates was performed using the RiboPrinter® Microbial Characterization System at the Department of Biological Sciences, Purdue University Calumet in Hammond, Indiana, under the direction of Drs. Charles Tseng and Evert Ting. The method compared the relatedness of DNA fragments from ribosomal genes in *E. coli* isolates collected in surface-water samples to *E. coli* from five source groups: cattle, deer, horses, humans, and sewage. A total of 225 *E. coli* isolates were cultured from surface-water samples collected at sites 1, 2, 4, and 5 during August 2002 and February and June 2003 (table 3) and ribotyped for identification by comparison. At each site, water was collected in five, sterile 1-liter polypropylene bottles, filled a minimum of 3 minutes apart. Bottles were filled by facing the bottle into the current and dipping into the stream at 3 to 5 equally spaced locations in the stream cross section. The samples were placed on ice and delivered within 12 hours to the Oklahoma State University Animal Disease Diagnostic Laboratory, Stillwater, Oklahoma, for processing. The goal was to isolate five *E. coli* colonies from each bottle to acquire 25 colonies per site per sampling episode. There were difficulties in cultivating *E. coli* from the February samples because of the low numbers of *E. coli* present in the water. Bacteria were not enumerated in water samples.

A total of 195 *E. coli* isolates were cultured for ribotyping from fecal material collected from cattle, deer, and horses, and effluent from septic tanks and wastewater lagoons within the Turkey Creek watershed (fig. 3). Effluent from inside septic tanks was collected at a city ballpark, two businesses, and two churches. Overall, seven separate samples were collected; two septic tanks were sampled twice on different occasions. Three to 10 *E. coli* isolates were cultured from effluent from each septic tank; a total of 63 human *E. coli* isolates were ribotyped (table 4). Twenty-three *E. coli* isolates were cultured and ribotyped from untreated sewage collected from wastewater lagoons used by the towns of Helena, Lahoma, Drummond, and Hennessey. Sample locations were at the inflow where untreated sewage enters the lagoons. Zero to five *E. coli* isolates were cultured from each sample. *E. coli* also was cultured from untreated sewage accessed through a manhole near a middle school in the watershed. Bacteria from wastewater lagoons and septic tanks were grouped separately for the ribotyping process, because there is no assurance that bacteria cultured from sewage effluent is human. An effort was made to collect from public septic tanks receiving waste only from humans and servicing large numbers of people to ensure diversity of the bacterium.

E. coli bacteria were cultured from 69 cattle—12 dairy and 57 beef. One *E. coli* colony was isolated from each animal. The

Table 2. Constituents analyzed, analyzing laboratory, laboratory method, minimum reporting level, and method references used

[USGS, U.S. Geological Survey; ASF, automated-segmented flow; ID, identification number; m-FC, fecal coliform medium; TTC, triphenyltetrazolium chloride; ˚C, degrees Celsius; mg/L, milligrams per liter]

Constituents	Analyzing laboratory	Laboratory method	Minimum reporting level	Method references
(USGS schedule 1697) (1) Nitrite nitrogen, (filtered) (2) Nitrite + nitrate nitrogen, (filtered) (3) Ammonia nitrogen, (filtered) (4) Ammonia nitrogen + organic nitrogen, (unfiltered)	USGS National Water Quality Laboratory, Lakewood, Colorado	(1) Colorimetry, ASF, method ID: I-2540-90 (2) Colorimetry, ASF, cadmium reduction-diazotization, method ID: I-2545-90 (3) Colorimetry, ASF, Salicylate-hypochlorite; method ID: I-2522-90 (4) Colorimetry, ASF, microkjeldahl digestion; method ID: I-4515-91	(1) 0.008 mg/L (2) 0.060 mg/L (3) 0.04 mg/L (4) 0.10 mg/L	(1,2,3) Fishman (1993) (4) Patton and Truitt (2000)
(1) Total phosphorus, (unfiltered), (2) Orthophosphate, (filtered)	U.S. EPA National Risk Management Research Laboratory Robert S. Kerr Environmental Research Center Ada, Oklahoma	Lachat flow injection analysis method, 10-115-01-1-C (Total phosphorus), 410-115-01-1-A (Orthophosphate)	(1) 0.10 mg/L (2) 0.02 mg/L	(1) Lachat Instruments (2003a) (2) Lachat Instruments (2003b)
Ratio of the stable nitrogen isotopes in nitrate nitrogen ^{15}N and ^{14}N	Water Sciences Laboratory, University of Nebraska, Lincoln, Nebraska	Steam distillation		Bremner and Keeney (1965) Gormly and Spalding (1979) Kreitler (1975)
E. coliform bacteria culture and isolation	Oklahoma State University Animal Disease Diagnostic Laboratory, Stillwater, Oklahoma	Membrane filtration, m-FC media, 24 hours at 44.5˚C Swab inoculated Tergitol 7 agar with TTC incubated for 18 hours at 35 ˚C		Clesceri and others (1998) Murray and others (1999)
Bacterial source tracking	Purdue University Calumet, Hammond, Indiana	DuPont Qualicon, The RiboPrinter® Microbial Characterization System		Bruce and others (1997) DuPont Qualicon (2003)

Table 3. Date of collection and number of *Escherichia coli* isolates cultured from surface-water-quality samples collected at sites 1, 2, 4, and 5 on Turkey Creek for bacterial source tracking using the RiboPrinter® Microbial Characterization System (DuPont Qualicon, 2003) at the Department of Biological Sciences, Purdue University Calumet, Hammond, Indiana

Dates	Site 1	Site 2	Site 4	Site 5	Number of *Escherichia coli* isolates cultured
8-13-02	24	24	25	25	98
2-11-03	10	7	1	9	27
6-24-03	25	25	25	25	100
Total					225 ribotyped

beef cattle were scattered throughout the watershed and a maximum of five fecal samples from separate cattle were collected at each location. Collection of fecal material from beef cattle was split into summer and winter in case diet affected the bacteria strain. During the summer, diet consists primarily of green grass; whereas, during the winter, diet consists of dried grass and alfalfa pellets. Twenty-six *E. coli* isolates were cultured from horses and 14 from deer. A rectal swab was used for horses; whereas, fecal material was collected for deer. One *E. coli* colony was isolated from each horse and a maximum of five horses were swabbed at each location.

Water Quality

Stream discharge, measured water properties, and surface-water quality results are listed in table 5. Stream discharge on the main stem of Turkey Creek increased during low-flow conditions from an average of 5.4 cubic feet per second (ft^3/s) at site 1 to 39 ft^3/s at site 5, indicating that Turkey Creek gains water from ground-water discharge. A portion of the increase in stream discharge may be from discharges of treated effluent from city sewage lagoons. However, the volume and frequency of discharges are unknown. Surface-water-quality samples show that specific conductance ranged from 1,180 to 1,740 microsiemens per centimeter during low-flow conditions and in general, decreased downstream with site 1 or site 2 having the largest measurement and site 5 having the lowest. The pH values were slightly alkaline and ranged from 6.8 to 8.5 with a median of 8.2. Dissolved oxygen concentrations were larger during winter when surface-water temperatures were low. Dissolved oxygen ranged from 9.3 to 15.9 milligrams per liter (mg/L) in samples collected in the months of November, February, and March and ranged from 5.3 to 13.9 mg/L in samples collected in the months of June, July, and August.

Nitrogen

Analysis of surface-water-quality samples show that nitrite nitrogen comprises a small portion of the total nitrogen in all samples, with an average concentration of 0.028 mg/L. The median concentrations of nitrite plus nitrate as nitrogen and total nitrogen (sum of nitrite plus nitrate as nitrogen and ammonia plus organic nitrogen as nitrogen) are larger than the average median concentrations calculated from water-quality sites in Oklahoma and part of Arkansas (excluding sites in the Ozark Highland and the Ouachita Mountains ecoregions) having similar stream orders and stream slopes (table 6, figs. 4 and 5) (Haggard and others, 2003, table 7). The median nitrite plus nitrate as nitrogen concentration of 1.16 mg/L measured on Turkey Creek is about 3.3 times larger than the average median concentration of 0.35 mg/L for streams in Oklahoma described in table 6. Boxplots on figure 4 illustrate the distribution of nitrite plus nitrate as nitrogen and total nitrogen concentra-

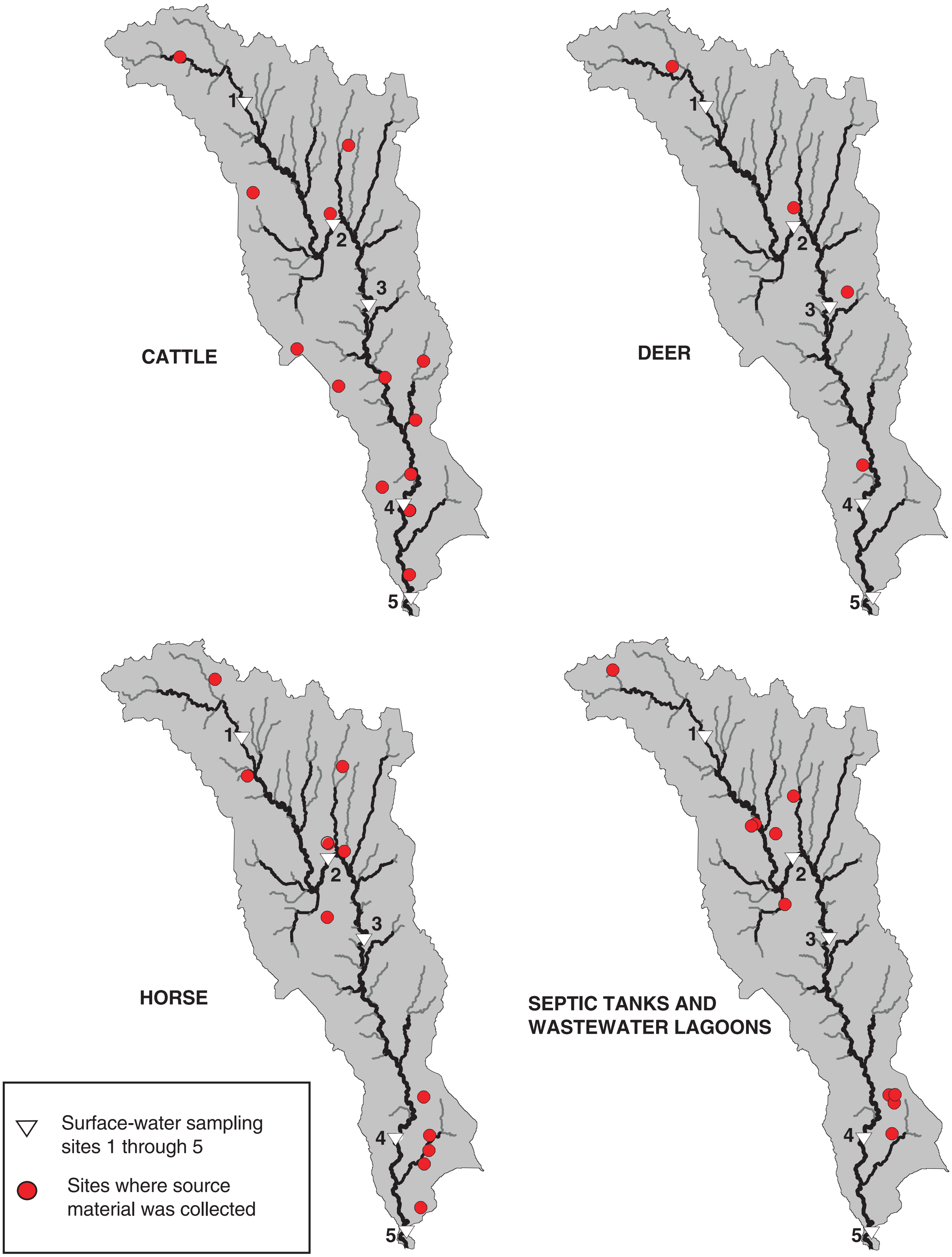


Figure 3. Sites where source material for bacterial source tracking was collected in the Turkey Creek watershed, northwest Oklahoma, June 2002 to June 2003.

Table 4. Number of *Escherichia coli* isolates cultured from source group material collected in the Turkey Creek watershed for bacterial source tracking using the RiboPrinter® Microbial Characterization System (DuPont Qualicon, 2003) at the Department of Biological Sciences, Purdue University Calumet, Hammond, Indiana

Source group material (number of samples)	Number of *Escherichia coli* isolates cultured
Cattle (69)	69
Deer (14)	14
Horse (26)	26
Human/septic tank (7)	63
Sewage (7)	23
Total	195 ribotyped

Table 5. Measured water properties and concentrations of nutrients and stable nitrogen isotopes in surface-water-quality samples from sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003

[Q, Discharge, instantaneous, cubic feet per second; SC, Specific conductance, microsiemens per centimeter at 25 degrees Celsius; pH, standard units; Temp, Temperature, water, degrees Celsius; DO, Dissolved oxygen, milligrams per liter; NO_2, Nitrite, filtered, milligrams per liter as nitrogen; NO_2+NO_3, Nitrite plus nitrate, filtered, milligrams per liter as nitrogen; NH_3, Ammonia, filtered, milligrams per liter as nitrogen; NH_3+OrgN, Ammonia plus organic nitrogen, unfiltered, milligrams per liter as nitrogen; Total P, Phosphorus, unfiltered, milligrams per liter; Ortho-PO_4, Orthophosphate, unfiltered, milligrams per liter as phosphorus; δ 15N, Nitrogen-15/Nitrogen-14 ratio, unfltered, per mil ; <, Less than; e, Estimated; --, No data; Analytes detected below minimum reporting levels were censored to reporting level; USGS, U.S. Geological Survey; ID, identifier]

Date	Q	SC	pH	Temp	DO	NO_2	NO_2+NO_3	NH_3	NH_3+OrgN	Total P	Ortho-PO_4	$\delta^{15}N$
Site 1, Turkey Creek near Goltry, USGS site ID 07158540												
Jun 3, 2002	0.81	1,510	8	23	7.6	0.012	0.06	0.05	1.3	0.28	0.28	--
Jul 2, 2002	2	1,580	8	22.8	11.9	<0.008	<0.05	<0.04	1.1	0.27	0.11	--
Aug 14, 2002	147	228	7.2	19.3	7.5	0.024	0.51	0.05	2.3	1.82	0.53	1.36
Nov 12, 2002	7.7	1,680	8.2	10.7	11.3	0.023	1.61	<0.04	0.4	0.17	0.13	7.74
Jan 14, 2003	6.3	1,570	7.2	3.5	--	0.02	1.29	0.13	0.64	<0.1	0.09	4.31
Feb 11, 2003	8	1,500	7.5	3.0	13.3	0.028	1.55	0.05	0.59	0.14	0.1	8.35
Mar 26, 2003	10	1,520	8.1	11.2	10.8	0.069	1.77	0.14	0.82	0.22	0.19	8.16
Jun 24, 2003	3	1,530	8.1	24.4	5.5	<0.008	<0.06	<0.04	1.6	0.42	0.26	--
Site 2, Turkey Creek near Lahoma, USGS site ID 07158900												
Jun 3, 2002	3.9	1,370	8.4	26.7	9.7	0.054	0.92	<0.04	1.1	0.39	0.44	7.08
Jul 2, 2002	2.5	1,740	8.5	24.4	12.8	0.008	0.07	<0.04	0.95	0.27	0.21	--
Aug 14, 2002	1,200	100	6.8	18.9	6.2	0.028	0.47	0.07	4.3	2.09	1.01	-0.47
Nov 12, 2002	23	1,730	8.3	11.2	10.3	0.053	1.69	0.16	0.7	0.23	0.26	9.65
Jan 14, 2003	16	1,730	8.2	3.9	--	0.017	1.16	<0.04	0.51	<0.1	0.1	6.00
Feb 11, 2003	16	1,630	8	5.3	15.4	0.02	1.16	<0.04	0.46	0.11	0.07	6.81
Mar 26, 2003	22	1,450	8.2	13.2	9.4	0.103	1.43	0.26	1.3	0.32	0.26	7.55
Jun 24, 2003	7.7	1,260	7.9	23.9	8.1	0.032	0.66	<0.04	1.3	0.58	0.44	3.63

Table 5. Measured water properties and concentrations of nutrients and stable nitrogen isotopes in surface-water-quality samples from sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003

[Q, Discharge, instantaneous, cubic feet per second; SC, Specific conductance, microsiemens per centimeter at 25 degrees Celsius; pH, standard units; Temp, Temperature, water, degrees Celsius; DO, Dissolved oxygen, milligrams per liter; NO_2, Nitrite, filtered, milligrams per liter as nitrogen; NO_2+NO_3, Nitrite plus nitrate, filtered, milligrams per liter as nitrogen; NH_3, Ammonia, filtered, milligrams per liter as nitrogen; NH_3+OrgN, Ammonia plus organic nitrogen, unfiltered, milligrams per liter as nitrogen; Total P, Phosphorus, unfiltered, milligrams per liter; Ortho-PO_4, Orthophosphate, unfiltered, milligrams per liter as phosphorus; δ 15N, Nitrogen-15/Nitrogen-14 ratio, unfltered, per mil ; <, Less than; e, Estimated; --, No data; Analytes detected below minimum reporting levels were censored to reporting level; USGS, U.S. Geological Survey; ID, identifier]

Date	Q	SC	pH	Temp	DO	NO_2	NO_2+NO_3	NH_3	NH_3+OrgN	Total P	Ortho-PO_4	$\delta^{15}N$
Site 3, Turkey Creek near Waukomis, USGS site ID 07159010												
Jun 3, 2002	6.8	1,440	8.4	30.2	10.7	0.029	0.49	0.08	0.73	0.31	0.41	-0.06
Jul 2, 2002	8.6	1,590	8.4	26.4	13.9	e 0.004	0.05	<0.04	0.63	0.28	0.23	--
Aug 13, 2002	1,220	186	7.7	21.8	5.9	0.06	1.09	0.16	5.8	2.31	1.18	-0.51
Nov 12, 2002	33	1,610	8.4	13.1	10.9	0.024	1.85	e 0.04	0.45	0.19	0.16	6.31
Jan 14, 2003	27	1,610	8.3	5.1	--	0.012	1.42	<0.04	0.37	<0.1	0.08	5.45
Feb 11, 2003	32	1,570	8.3	9.5	14.8	0.017	1.27	<0.04	0.52	0.1	0.04	6.56
Mar 26 2003	36	1,380	8.3	17.4	9.8	0.072	1.65	0.14	1.1	0.27	0.23	6.64
Jun 24, 2003	12	1,370	8.4	31	10	0.01	0.27	<0.04	1	0.41	0.36	--
Site 4, Turkey Creek south of Hennessey, USGS site ID 07159060												
Jun 4, 2002	10	1,440	8.4	25.4	10	e 0.006	0.07	<0.04	0.77	0.37	0.48	--
Jul 3, 2002	14	1,460	8.3	24.5	10.9	<0.008	e 0.04	<0.04	0.54	0.42	0.4	--
Aug 13, 2002	1,420	467	7.6	23.3	5.3	0.087	1.35	0.14	5.2	2.11	0.87	1.91
Nov 13, 2002	45	1,570	8.4	9	11	0.013	1.82	<0.04	0.42	0.22	0.25	6.81
Jan 14, 2003	38	1,550	8.3	6.2	--	0.012	1.57	0.06	0.56	<0.1	0.08	6.46
Feb 11, 2003	45	1,660	8.4	6.1	15.9	0.015	1.38	<0.04	0.51	0.1	0.05	6.73
Mar 26, 2003	54	1,340	8.2	14	10.3	0.04	1.5	0.07	0.96	0.3	0.23	7.05
Jun 24, 2003	20	1,440	8.2	28.7	10	0.01	0.27	<0.04	1.1	0.58	0.42	--
Site 5, Turkey Creek lower near Dover, USGS site ID 20030211200												
Jun 4, 2002	16	1,270	8.5	25.7	10.9	0.015	0.54	<0.04	0.89	0.27	0.29	5.36
Jul 3, 2002	16	1,280	8.2	24.1	9.6	0.022	0.65	0.08	0.69	0.34	0.24	6.91

Table 5. Measured water properties and concentrations of nutrients and stable nitrogen isotopes in surface-water-quality samples from sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003

[Q, Discharge, instantaneous, cubic feet per second; SC, Specific conductance, microsiemens per centimeter at 25 degrees Celsius; pH, standard units; Temp, Temperature, water, degrees Celsius; DO, Dissolved oxygen, milligrams per liter; NO_2, Nitrite, filtered, milligrams per liter as nitrogen; NO_2+NO_3, Nitrite plus nitrate, filtered, milligrams per liter as nitrogen; NH_3, Ammonia, filtered, milligrams per liter as nitrogen; NH_3+OrgN, Ammonia plus organic nitrogen, unfiltered, milligrams per liter as nitrogen; Total P, Phosphorus, unfiltered, milligrams per liter; Ortho-PO_4, Orthophosphate, unfiltered, milligrams per liter as phosphorus; δ 15N, Nitrogen-15/Nitrogen-14 ratio, unfltered, per mil ; <, Less than; e, Estimated; --, No data; Analytes detected below minimum reporting levels were censored to reporting level; USGS, U.S. Geological Survey; ID, identifier]

Date	Q	SC	pH	Temp	DO	NO_2	NO_2+NO_3	NH_3	NH_3+OrgN	Total P	Ortho-PO_4	$\delta^{15}N$
Aug 13, 2002	612	280	7.7	22.4	5.7	0.065	1.16	0.17	4.6	1.75	0.72	1.04
Nov 13, 2002	51	1,460	8.5	10.1	11	0.011	2.12	<0.04	0.39	0.22	0.23	5.94
Jan 14, 2003	43	1,500	8.3	7	--	0.016	1.89	<0.04	0.51	<0.1	0.07	6.56
Feb 11, 2003	54	1,500	8.4	4	12.2	0.022	1.62	<0.04	0.42	<0.1	0.04	6.74
Mar 26, 2003	64	1,240	8.1	12.2	9.3	0.044	1.67	0.07	0.98	0.29	0.23	7.11
Jun 24, 2003	31	1,180	8	26.3	7.3	0.011	0.6	<0.04	1.5	0.56	0.35	5.08

Table 6. A comparison of median nutrient concentrations measured in surface-water-quality samples from sites 1-5 on Turkey Creek collected June 2002 to June 2003, to the average median concentrations calculated from water-quality sites in Oklahoma and part of Arkansas (excluding sites in the Ozark Highland and Ouachita Mountains ecoregions) having similar stream orders and stream slopes. Taken from Haggard and others (2003, table 7)

[mg/L, milligrams per liter; NA, not available; number in ( ) are number of water-quality sites used for calculation]

Nutrient	Turkey Creek median 40 analyses	Oklahoma (and part of Arkansas) average median [1]
Nitrite plus nitrate as nitrogen	1.16 mg/L	0.35 mg/L (127)
Total nitrogen[2] (calculated)	2.02 mg/L	1.06 mg/L (74)
Total phosphorus	0.275 mg/L	0.083 mg/L (133)
Orthophosphate as phosphorus	0.235 mg/L	NA

[1]The average median calculated from percentile distributions of median concentrations from water-quality sites having stream orders 1, 2, 3, and stream slope less than or equal to 17 feet per mile (SS2).

[2]Sum of nitrite plus nitrate as nitrogen and ammonia plus organic nitrogen as nitrogen concentrations.

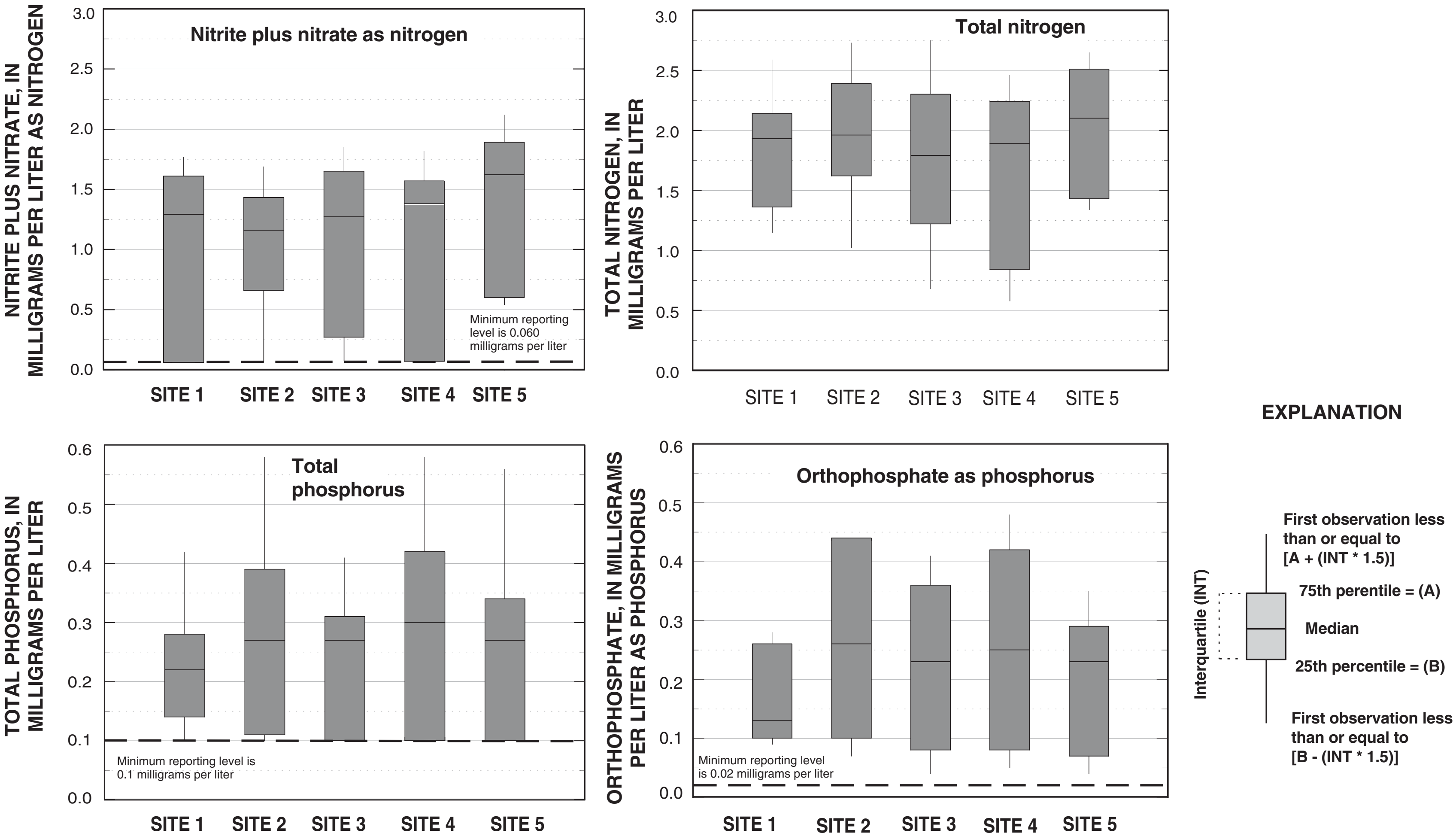


Figure 4. Distribution of nutrient concentrations measured in surface-water quality samples during low-flow conditions at sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003. Each box plot was constructed from seven analyses.

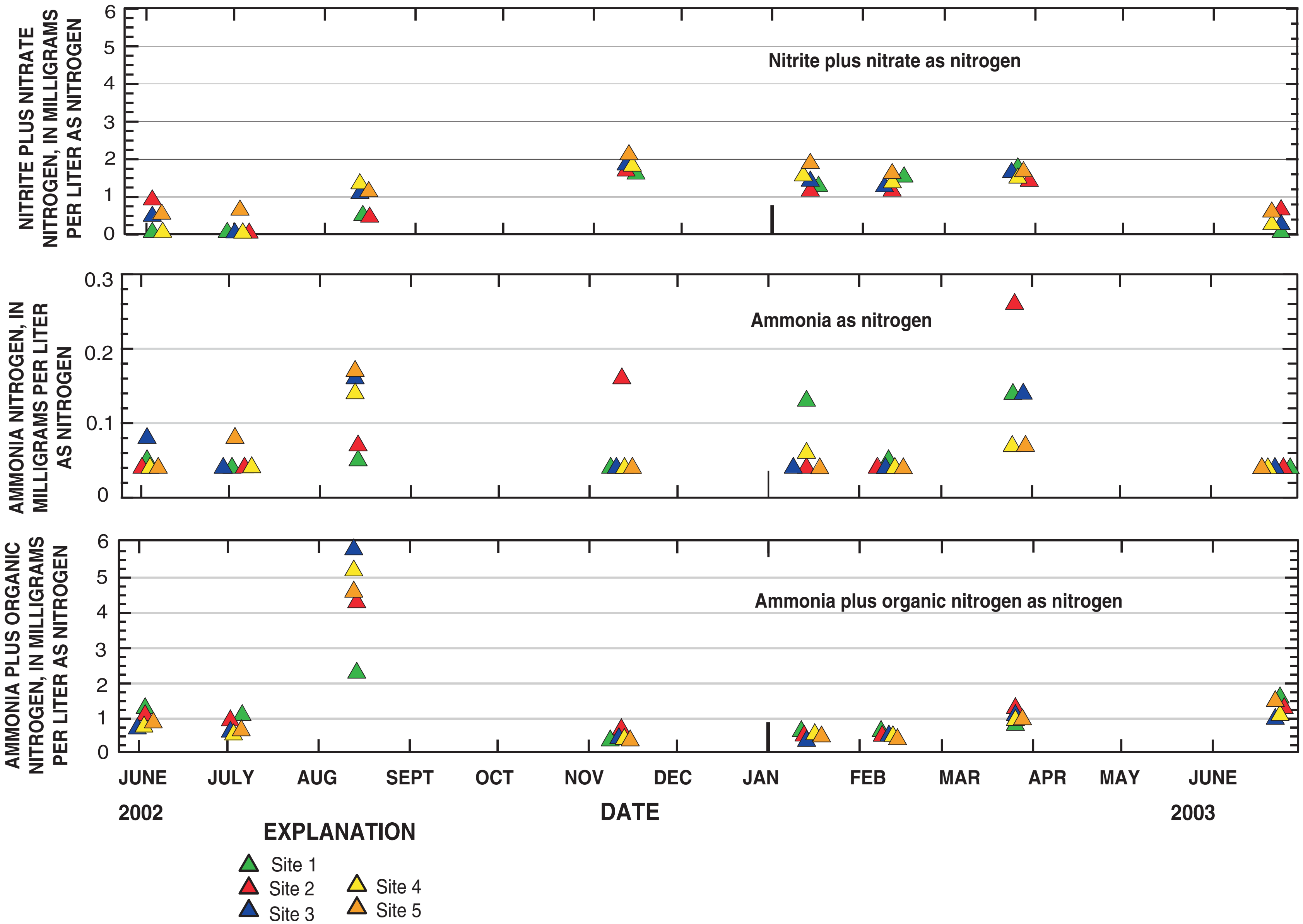


Figure 5. Distribution of nitrite plus nitrate as nitrogen, ammonia as nitrogen, ammonia plus organic nitrogen as nitrogen concentrations measured in surface-water quality samples from sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003.

tions measured during low-flow conditions at sites 1-5 from June 2002 to June 2003. Concentrations of nitrite plus nitrate as nitrogen were slightly larger in the winter samples; November and December 2002, January, February, and March 2003 (fig. 5). This may be from the lower uptake and use of nitrogen by vegetation during the winter months. Ammonia plus organic nitrogen as nitrogen concentrations were about 4 to 5 times larger during high-flow, possibly from organic material carried into the creek by runoff (fig. 5). During low-flow conditions, ammonia as nitrogen was generally less than 0.2 mg/L and ammonia plus organic nitrogen as nitrogen was less than 1.6 mg/L.

Phosphorus

Analysis of surface-water-quality samples show the median concentration of total phosphorus is larger than the average median concentration calculated from water-quality sites in Oklahoma and part of Arkansas (excluding sites in the Ozark Highland and the Ouachita Mountains ecoregions) having similar stream orders and stream slopes (table 6, figs. 4 and 5) (Haggard and others, 2003, table 7). Similar to nitrogen, the median total phosphorus concentration of 0.275 mg/L measured on Turkey Creek is about 3.3 times larger than the average median concentration of 0.083 mg/L for streams in Oklahoma described earlier. The distribution of total phosphorus concentrations on figure 6 shows that during August 2002, total phosphorus concentrations increased 7.7 times (average of sites 1-5, 2.016 mg/L) above the average concentration of 0.261 mg/L in samples collected during low-flow conditions. During the three winter months and March, (fig. 6) total phosphorus concentrations were lowest; compared to nitrite plus nitrate as nitrogen concentrations that were largest during this period. Figure 7 shows orthophosphate as phosphorus concentrations increased 3.5 to 4 times during the August 2002 sampling. Plots show similar trends in orthophosphate as phosphorus concentrations during the eight sampling episodes, both tended to increase during the summer months and decrease during winter months. A large fraction of all phosphorus measured in samples during low-flow conditions was present as orthophosphate as phosphorus. Samples for the analysis of orthophosphate as phosphorus were filtered with 0.45-micron filters and may have contained undissolved phosphorus sorbed onto colloidal material that passed through the filter.

Possible Sources of Nutrients and Bacteria

Nitrogen Isotopes

Values of $\delta^{15}N$ measured in surface-water samples and in common sources of nitrate are shown on figure 8. The minimum concentration of nitrate nitrogen as nitrogen required for the analysis of nitrogen isotopes was 0.3 mg/L. As a result, nitrogen isotopes were not measured in all samples from June and July 2002 and June 2003. Almost all samples collected during low-flow conditions show $\delta^{15}N$ values between 4 and 10 parts per thousand, above the range for atmospheric nitrogen and synthetic fertilizer and below the range for animal waste (Clark and Fritz, 1997). These samples may represent a mixture of nitrate from these two sources and other sources enriched with $\delta^{15}N$, such as soils and plants. The sample from site 3, June 2002, is depleted in $\delta^{15}N$ and is within the range encompassing synthetic fertilizer, plants, or atmospheric nitrogen (Clark and Fritz, 1997). The high-flow samples from August 2002, taken after a large precipitation event, are depleted in $\delta^{15}N$ and may include a large amount of nitrate derived from the atmosphere or synthetic fertilizer.

Bacterial Source Tracking

Results of the bacterial source tracking indicated that the two source groups having the greatest number of ribopattern matches with surface-water isolates were the cattle group, 53 isolates or 23.5 percent and the human group, 41 isolates or 18.2 percent. Fewer surface-water isolates matched the deer and horse groups, 8.0 percent and 3.5 percent, respectively. About 43 percent or 96 surface-water isolates were not matched to any source group (table 7 and fig. 9). The distribution of isolates matching source groups was relatively similar from site to site. An exception occurred at site 1, where a greater number of isolates matched the deer group than at the other three sites. Eight of the 14 deer fecal samples were collected upstream from site 1 and might have increased the probability of a match occurring.

Summary

The U.S. Geological Survey in cooperation with the Oklahoma Department of Environmental Quality and the U.S. Environmental Protection Agency investigated the distribution of surface-water quality and possible sources of nutrients and *E. coli* bacteria to surface water in the Turkey Creek watershed. Turkey Creek drains about 376 square miles of rural Oklahoma, starting just north of Helena in Alfalfa County and flowing south to the confluence with the Cimarron River. Land use in the Turkey Creek watershed is primarily agricultural; land use/cover information for 1992 shows that about 76 percent of land in the watershed was used for row crops and small grains and about 11 percent was used for pasture and hay. Important sources of nutrients in the Turkey Creek watershed are commercial fertilizers, animals including humans, vegetation, soils, and precipitation. Possible contributors of bacteria to the watershed are humans from septic tank effluent, cattle, and effluent from wastewater lagoons. Additionally, horses, dogs, cats, deer, waterfowl, and other small mammals are possible sources of bacteria and nutrients in the watershed.

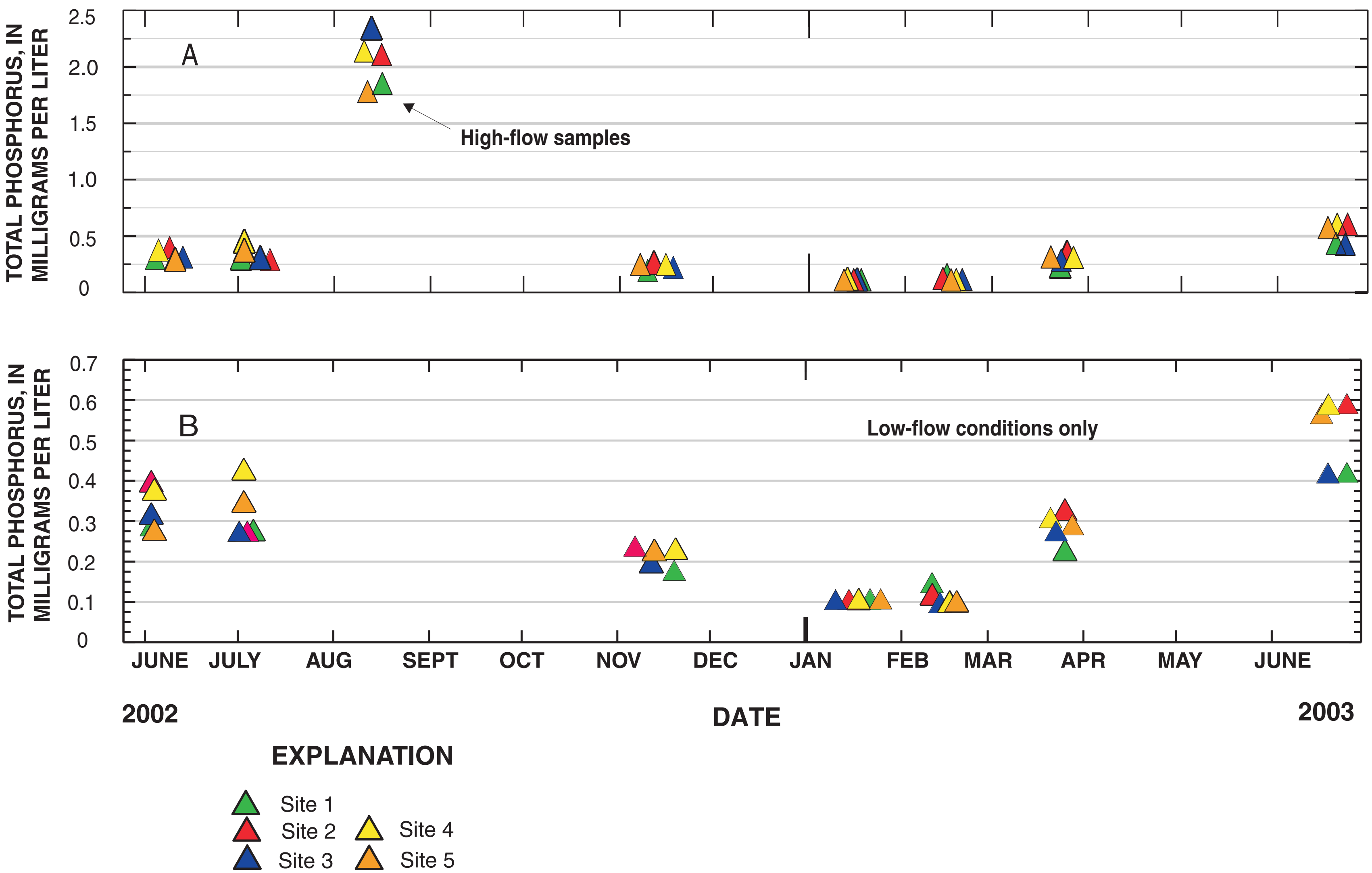


Figure 6. Distribution of concentrations of total phosphorus in surface-water quality samples from sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003. Plot A shows concentrations of all samples, plot B shows concentrations of samples collected during low-flow conditions only.

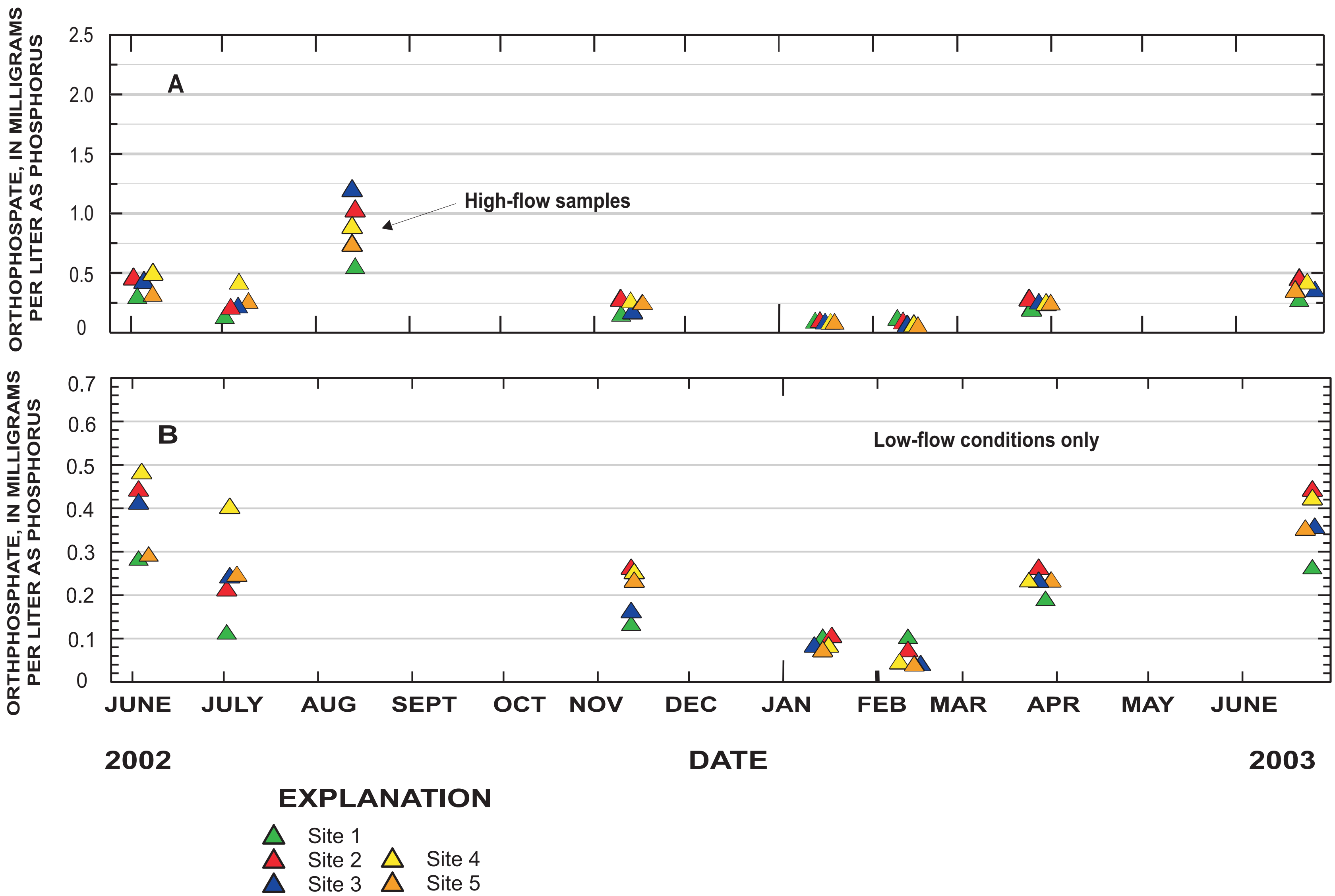


Figure 7. Distribution of concentrations of orthophosphate in surface-water quality samples from sites 1-5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003. Plot A shows concentrations of all samples, plot B shows concentrations of samples collected during low-flow conditions only.

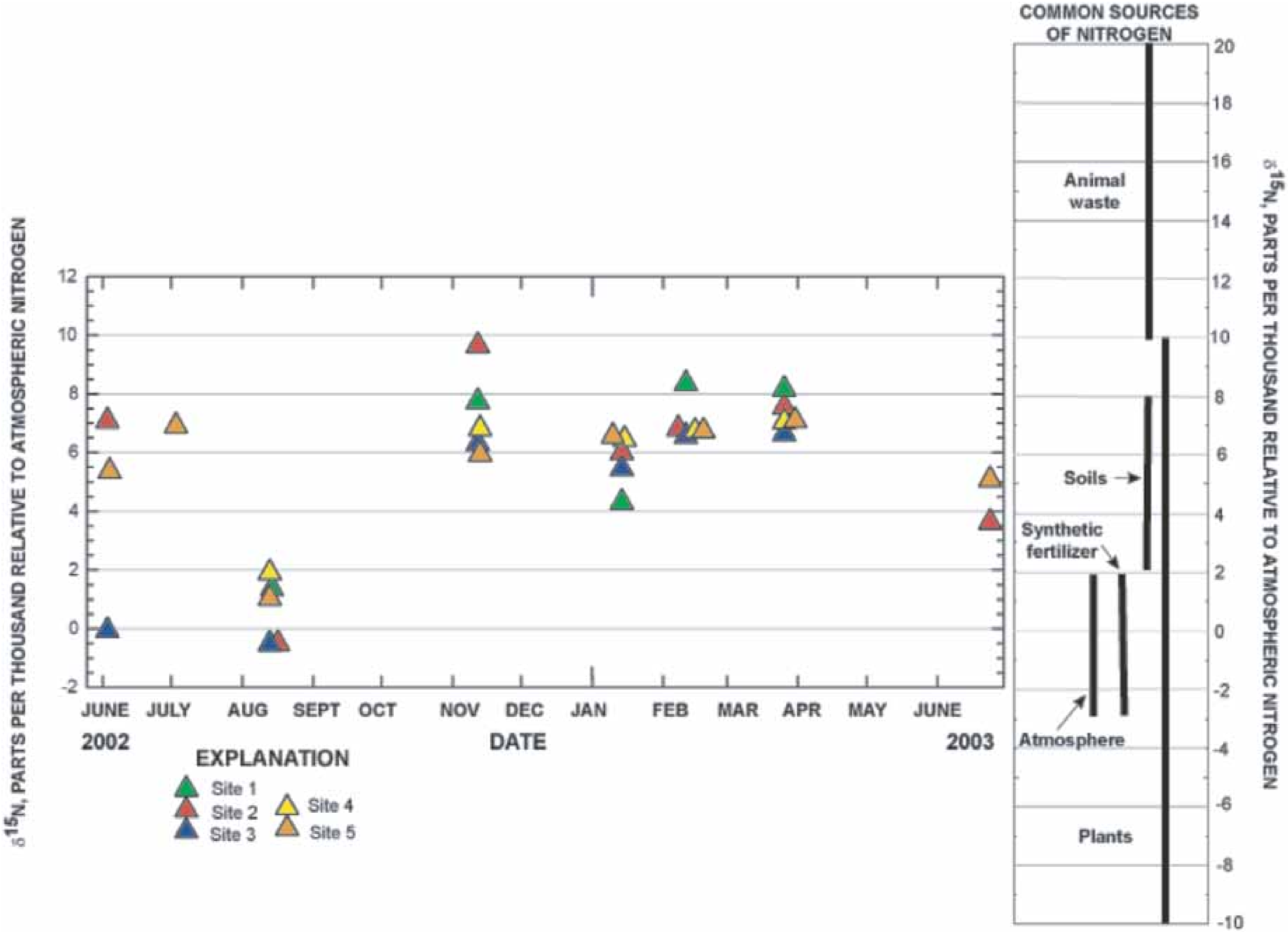


Figure 8. Distribution of $\delta^{15}N$ in nitrate found in common sources of nitrogen and in surface-water-quality samples from sites 1–5 on Turkey Creek, northwest Oklahoma, June 2002 to June 2003. Values of $\delta^{15}N$ in nitrate found in common sources of nitrogen taken from Clark and Fritz (1997).

Table 7. Number of *Escherichia coli* isolates cultured from surface-water-quality samples matching source groups collected in the Turkey Creek watershed, northwest Oklahoma. Matching results are based on an identification pattern comparison of greater than or equal to 95 percent

Source group	Site 1	Site 2	Site 4	Site 5	Total percentages
Cattle	8	15	15	15	23.5
Deer	11	5	0	2	8.0
Horse	3	1	1	3	3.5
Human/septic tank	11	7	10	13	18.2
Wastewater lagoons	2	4	0	3	4.0
Unidentified	24	24	25	23	42.7

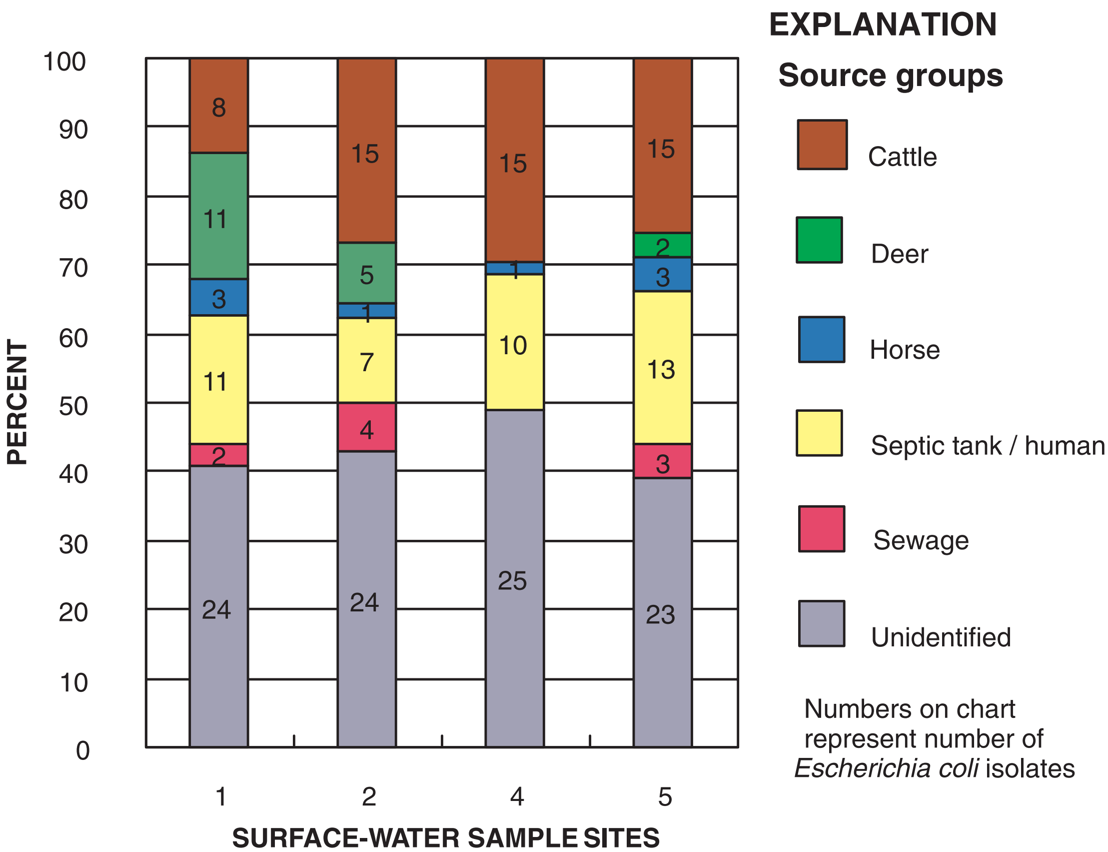


Figure 9. Percent of *Escherichia coli* isolates cultured from surface-water-quality samples from sites 1, 2, 4, and 5 matching ribopatterns of *Escherichia coli* from source groups in Turkey Creek watershed, northwest Oklahoma, June 2002 to June 2003. Bacterial source tracking was performed using the RiboPrinter® Microbial Characterization System (DuPont Qualicon, 2003) at the Department of Biological Sciences, Purdue University Calumet, Hammond, Indiana.

Five surface-water-quality collection sites (sites 1-5) were chosen on the main stem of Turkey Creek and surface-water samples were collected eight times between June 2002 and June 2003. Surface-water-quality samples were analyzed for nutrient compounds in filtered and unfiltered samples and the stable nitrogen isotopes ^{15}N and ^{14}N in nitrate nitrogen. Bacterial source tracking, also referred to as ribotyping, was performed to identify possible animal sources of *E. coli* bacteria to surface water. A total of 225 *E. coli* isolates were cultured from surface-water samples collected at sites 1, 2, 4, and 5 during August 2002 and February and June 2003 and ribotyped for identification by comparison. A total of 195 *E. coli* isolates were cultured for ribotyping from fecal material collected from cattle, deer, and horses, and effluent from septic tanks and wastewater lagoons within the Turkey Creek watershed.

Results show that stream discharge on the main stem of Turkey Creek increased during low-flow conditions from an average of 5.4 cubic feet per second at site 1 to 39 cubic feet per second at site 5 in the watershed, indicating that Turkey Creek gains water from ground-water discharge. A portion of the increase in stream discharge may be from discharges of treated effluent from city sewage lagoons. However, the volume and frequency of discharges are unknown. Surface-water-quality samples show that specific conductance ranged from 1,180 to 1,740 microsiemens per centimeter during low-flow conditions and in general, decreased downstream with site 1 or site 2 having the largest measurement and site 5 having the lowest. The pH values were slightly alkaline and ranged from 6.8 to 8.5 with a median of 8.2. Dissolved oxygen ranged from 9.3 to 15.9 mg/L in samples collected in the months of November, February, and March and ranged from 5.3 to 13.9 mg/L in samples collected in the months of June, July, and August.

Analysis of surface-water-quality samples show that the median concentrations of nitrite plus nitrate as nitrogen (1.16 mg/L) and total phosphorus (0.275 mg/L) are larger than the average median concentrations of 0.35 mg/L and 0.083 mg/L, respectively, calculated from water-quality sites in Oklahoma and part of Arkansas (excluding sites in the Ozark Highland and the Ouachita Mountains ecoregions) having similar stream orders and stream slopes. Concentrations of total phosphorus and orthophosphate as phosphorus tended to increase during the summer months and decrease in the winter months, whereas, concentrations of nitrite plus nitrate as nitrogen increased slightly during the winter months and decreased in the summer months. During high-flow conditions total phosphorus increased 7.7 times above the average concentration of 0.261 mg/L in samples collected during low-flow conditions. Orthophosphate concentrations increased 3.5 to 4 times during high-flow conditions.

Almost all samples collected during low-flow conditions showed $\delta^{15}N$ values between 4 and 10 parts per thousand, above the range for atmospheric nitrogen and synthetic fertilizer and below the range for animal waste. These samples may represent a mixture of nitrate from these two sources and other sources enriched with $\delta^{15}N$, such as soils and plants.

Results of the bacterial source tracking indicated that the two source groups having the greatest number of ribopattern matches with surface-water isolates were the cattle group, 53 isolates or 23.5 percent and the human group, 41 isolates or 18.2 percent. Fewer surface-water isolates matched the deer and horse groups, 8.0 percent and 3.5 percent, respectively. About 43 percent or 96 surface-water isolates were not matched to any source group.

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