
MEMORANDUM

TO: ANDREW SANSOM, PH.D
FROM: NICK DORNAK
SUBJECT: 2017 CYPRESS CREEK BACTERIAL SOURCE TRACKING – CUMULATIVE REPORT
DATE: DECEMBER 11, 2017
CC: EMILY WARREN, MEREDITH MILLER, MICHAEL JONES, CLAUDIA CAMPOS, TOM HEGEMIER, THOMAS HARDY, PH.D

Dr. Sansom,

In an effort to support Cypress Creek watershed protection planning and better characterize instream *E. coli* bacteria sources near downtown Wimberley, TX, the Meadows Center for Water and the Environment (Meadows Center) has entered into a partnership with the Texas A&M AgriLife Research – Soil & Aquatic Microbiology Laboratory (SAML) to conduct a short-term Bacterial Source Tracking (BST) study of Cypress Creek.

*BST is a valuable tool that can identify, and also rule-out, significant sources of E. coli pollution in a watershed. Using DNA fingerprints and bacterial markers, fecal pollution sources are identified by comparing E. coli DNA to those in a statewide library of known sources.*¹

The purpose of this memorandum is to provide a summary of the Cypress Creek BST study and to present laboratory results from three sampling events conducted by Meadows Center staff on the following dates:

- Round 1 – Wet Weather Conditions – August 7, 2017
- Round 2 – Dry Conditions – September 12, 2017
- Round 3 – Dry Conditions – October 16, 2017

Summary of Cypress Creek BST Study

Goal: Collection and effective interpretation of high quality *E. coli* bacteria source data toward the purpose of implementing more strategic management practices for addressing nonpoint source pollution in the Cypress Creek watershed.

SAML will perform *E. coli* bacterial enumeration and BST analysis of raw water samples collected once per month from Cypress Creek by Meadows Center staff from August 2017 through October 2017. Meadows Center staff will collect standard field data during each sampling event.² Findings will be reported to stakeholders on a monthly basis as data becomes available and will be summarized in a final report.

Beginning in August 2017, duplicate samples will be collected on a monthly basis from two locations on Cypress Creek (**Figure 1**). Site #1 is located on Cypress Creek approximately 0.33 mile upstream of the RR12 bridge located in downtown Wimberley. Site #1 was selected to provide data on upstream *E. coli* sources with minimal potential impact from septic systems or other *E. coli* sources in the downtown Wimberley area. Site #2 is located on Cypress Creek approximately 0.10 mile downstream of the RR12

¹ From Texas Water Resources Institute website, <http://texasbst.tamu.edu/>

² Meadows Center staff utilized for this study have been trained and certified in standard industry water quality sampling methods and collect data under multiple state and federal quality assurance plans. A copy of the approved Quality Assurance Project Plan for this study is on file at both the Meadows Center and SAML.

bridge and was selected to provide data on Cypress Creek *E. coli* sources including the downtown Wimberley area.

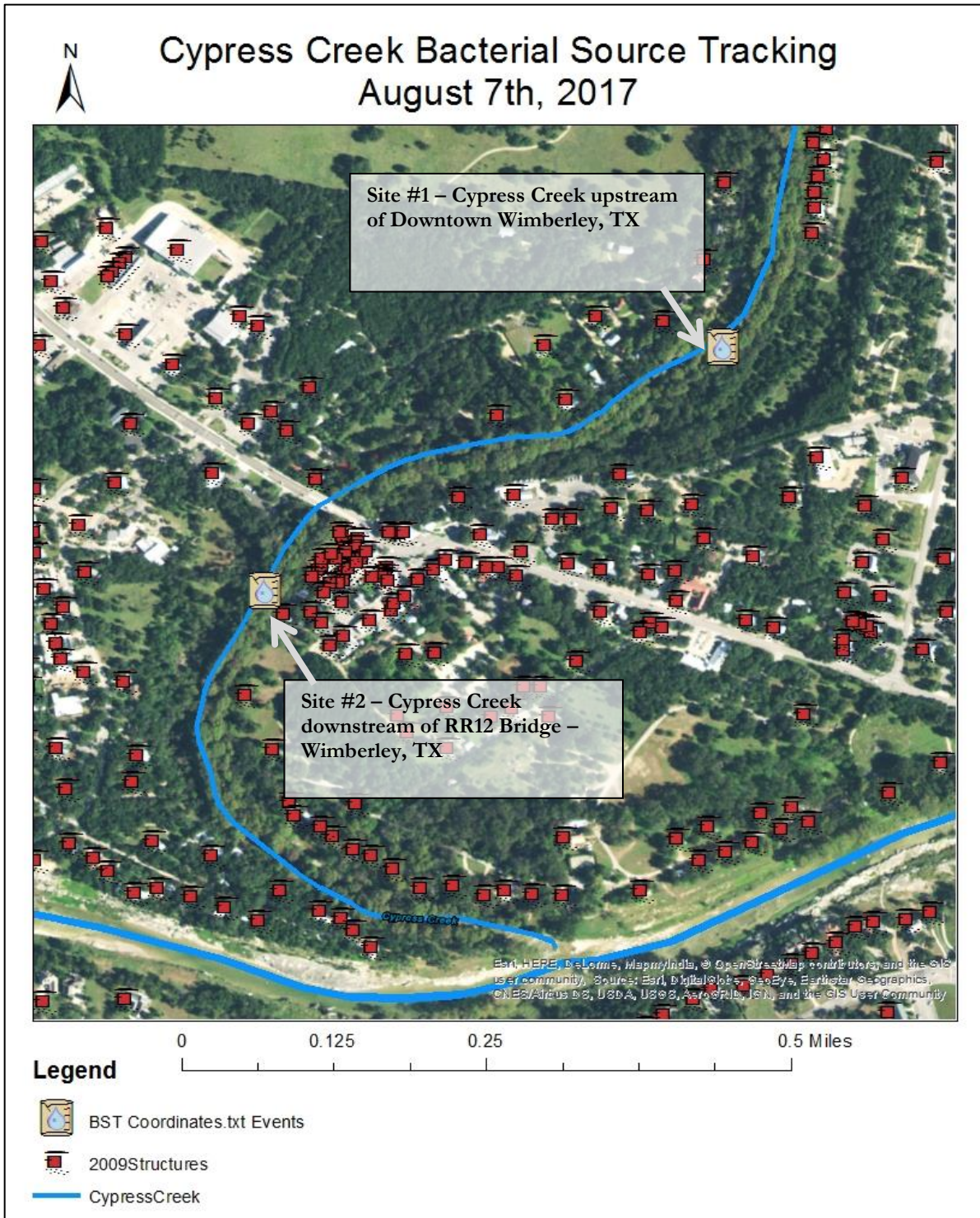


Figure 1. 2017 Cypress Creek Bacterial Source Tracking, Study area map

To provide a more representative sample set of routinely occurring *E. coli* sources in Cypress Creek, the project sampling plan specifies that one sampling event be conducted during “wet weather” conditions with an additional two sampling events to be conducted during “dry weather” conditions. BST analysis will be performed by SAML as follows:

- 3 events (2 dry, 1 wet - if possible)
- 6 isolates for each event from 2 sample sites (3 isolates per raw sample)
- 3 events x 6 isolates = 18 total isolates

In the Cypress Creek BST study, *E. coli* bacterial isolates from fecal pollution will be “fingerprinted” and their DNA compared to those in a statewide library of known sources.³ The bacterial isolates will be selected randomly and identified using both a 3-way categorization and a 7-way categorization protocol. The calculated “rate of correct classification” for SAML is 100% for the 3-way split and 91% for the 7-way split. More information on the Texas Statewide BST Library and BST classification can be obtained from the attached BST laboratory reports provided by SAML on September 12, 2017 and on December 1, 2017.

Bacterial Source Tracking Results for 8/7/17, 9/12/17 and 10/16/17 Sampling Events

For a more thorough analysis of the BST results, please see the attached BST laboratory reports.

** The ID of the closest library match for each isolate should be used for informational purposes only and not be interpreted as species-level source classification of the isolates since our current methods are not capable of doing this (e.g., they cannot distinguish between isolates from different species of non-avian wildlife, such as deer and feral hogs).*

Table 1. Classification of *E. coli* isolates from Site #1 (Upstream)

Sample Date	Weather Conditions	EPA Method 1603 Result (<i>E. coli</i> CFU/100 mL)	Isolate	3-way id	7-way id	Closest Match*
8/7/17	Wet	6,400	Cypress Creek-582572-8/7-A	Livestock and Domesticated Animals	Cattle	Cattle
			Cypress Creek-582572-8/7-C	Wildlife	Wildlife, Non-Avian	Mouse
			Cypress Creek-582572-8/7-D	Livestock and Domesticated Animals	Cattle	Cattle
			Cypress Creek-582572-8/7-E	Wildlife	Wildlife, Non-Avian	Feral Hog
9/12/17	Dry	36	Cypress Creek-9/12-582589-B	Wildlife	Wildlife, Non-Avian	Coyote
			Cypress Creek-9/12-582589-D	Wildlife	Wildlife, Non-Avian	Opossum
			Cypress Creek-9/12-582589-E	Wildlife	Wildlife, Avian	Vulture
10/16/17	Dry	60	Cypress Creek-10/16-582597-A	Wildlife	Wildlife, Non-Avian	Skunk
			Cypress Creek-10/16-582597-B	Livestock and Domesticated Animals	Livestock, Avian	Chicken
			Cypress Creek-10/16-582597-C	Livestock and Domesticated Animals	Livestock, Avian	Chicken

³ Bacterial isolates are defined as a pure strain of bacteria that has been separated from a mixed bacterial culture.

Table 2. Classification of *E. coli* isolates from Site #2 (Downstream)

Sample Date	Weather Conditions	EPA Method 1603 Result (<i>E. coli</i> CFU/100 mL)	Isolate	3-way id	7-way id	Closest Match*
8/7/17	Wet	18,000	Cypress Creek-582571-8/7-A	Human	Human	Raw Sewage
			Cypress Creek-582571-8/7-B	Unidentified	Unidentified	Cattle
			Cypress Creek-582571-8/7-C	Livestock and Domesticated Animals	Cattle	Cattle
			Cypress Creek-582571-8/7-E	Wildlife	Wildlife, Non-Avian	Feral Hog
9/12/17	Dry	276*	Cypress Creek-9/12-582587-C	Wildlife	Wildlife, Non-Avian	Deer
			Cypress Creek-9/12-582587-D	Wildlife	Wildlife, Non-Avian	Feral Hog
			Cypress Creek-9/12-582587-E	Wildlife	Wildlife, Non-Avian	Coyote
10/16/17	Dry	650	Cypress Creek-10/16-582596-A	Wildlife	Wildlife, Avian	Duck
			Cypress Creek-10/16-582596-B	Wildlife	Wildlife, Non-Avian	Feral Hog
			Cypress Creek-10/16-582596-C	Wildlife	Wildlife, Non-Avian	Raccoon

Field notes and observations

- The August 7 sampling event was conducted during a storm event under wet weather conditions. 24-hour precipitation for August 7 sampling⁴ = 2.5 in. Active surface runoff was documented during sampling.
- The August 7 sampling event was conducted during a storm event yielding much higher concentrations of *E. coli* bacteria than would be expected during baseflow conditions in Cypress Creek. Nonpoint source pollution carried by stormwater may also yield different *E. coli* bacterial sources carried over longer distances than would be expected under dry conditions.
- There was an odor of sulfur or sewage in the air near Site #2 on August 7.
- The September 12 and October 16 sampling events were conducted under dry conditions with water appearing clear. Similar stream width, depth and weather conditions were documented on for each sampling event during dry weather conditions.
- The odor of sulfur or sewage near Site #2 noted on August 7 was not present during the September 12 and October 16 sampling events.
- All three sampling events took place between 9AM and 10:30AM. Standards for sampling procedures are identified in the approved Cypress Creek BST QAPP.
- The smell of bat guano around the RR12 bridge was persistent during each sampling event.
- **Figure 2** demonstrates daily maximum discharge, daily minimum discharge and daily mean discharge from USGS Gauge 08170990 Jacobs Well Spring near Wimberley, TX from December 2016 through December 2017. Note that discharge data is not available for September 12.

⁴ NOAA Record of Climatological Observations, Station: WIMBERLEY 4.4 E, TX US1TXHYS003

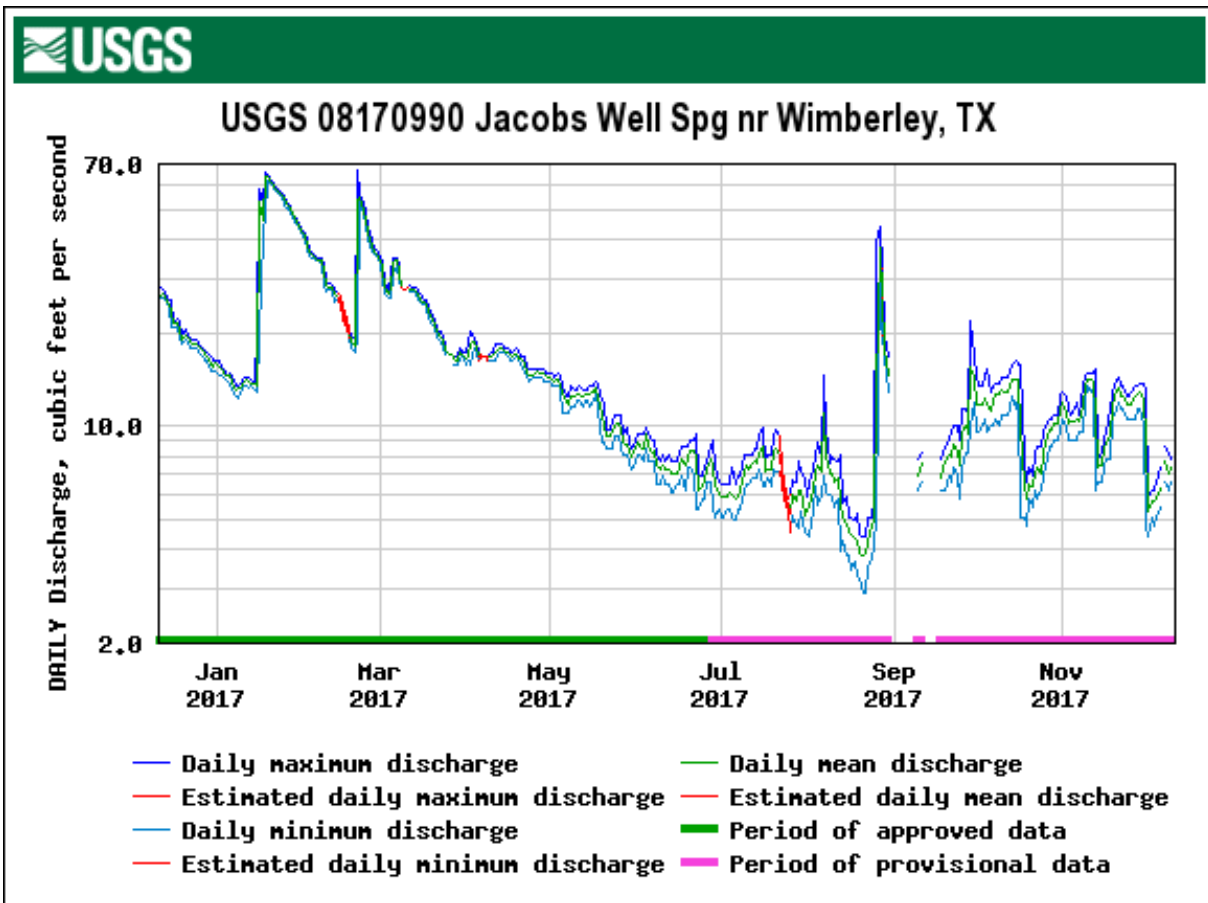


Figure 2. Discharge from USGS Gauge 08170990 Jacobs Well Spring near Wimberley, TX from December 2016 through December 2017

Analysis and recommendations

- The BST results in this report should be interpreted cautiously since they represent only 3 to 4 *E. coli* isolates from each sample. Analysis of *E. coli* isolates from additional sampling events may strengthen and further validate these initial results.
- Livestock or wildlife were identified as the source for 90% (18 of 20) of the isolates analyzed by SAML using a 7-way ID. While this does not eliminate other sources, it does indicate that livestock and wildlife are a substantial source of *E. coli* bacteria present in this reach of Cypress Creek during both dry and wet weather conditions.
- This study indicates substantial bacteria loading in Cypress Creek at one or more locations over the approximately 1/2 mile reach of Cypress Creek studied. There were considerable increases in recorded *E. coli* concentrations noted during each sampling event (both wet and dry weather conditions) moving from upstream Site #1 to downstream Site #2. The data indicate Site #1 is achieving water quality protective of safe contact recreation during baseflow conditions while *E. coli* concentrations at Site #2 exceeded contact recreation standards during each sampling event.
- As would be expected, the highest concentration of *E. coli* bacteria and most diverse identification of sources was recorded during wet weather conditions with active runoff. The only 7-way ID for

human was recorded during the wet weather event at Site #2. Note also smell of sulfur or sewage at Site #2 on August 7.

- Based on observable land use for this reach of Cypress Creek, direct deposition of fecal matter into the stream by livestock and/or wildlife as well as the possibility of leaking septic systems would be the most likely sources of *E. coli* bacteria found in the study area *during dry weather conditions*.
- During dry weather conditions, non-avian wildlife were identified as the source for 5 of 6 isolates at Site #2. Further investigation of potential *E. coli* contributions in Cypress Creek from the colony of bats living under the RR12 bridge in the study area should be considered.
- With clear and dramatic increases in *E. coli* concentrations observed in Cypress Creek between Site #1 and Site #2, this study demonstrates an ideal location for the implementation of best management practices for reducing direct, in-stream deposition of bacteria. Based on EPA Method 1603 test results, BST findings and observable land use, both the bat colony under the RR12 bridge and the possibility of leaking septic systems in the downtown Wimberley area should be further investigated.

Thank you for the opportunity to provide results and analyses for this first ever Cypress Creek BST study. I look forward to continuing this effort with the Meadows Center and SAML to enhance the characterization of *E. coli* bacteria sources in the Cypress Creek watershed.

Sincerely,



Nick Dornak

Attachments:

1. Cypress Creek BST Reports from SAML for August 7, September 12 and October 16 Sampling Events
2. Cypress Creek SAML Test Results for August 7, September 12 and October 16 Sampling Events, EPA Method 1603