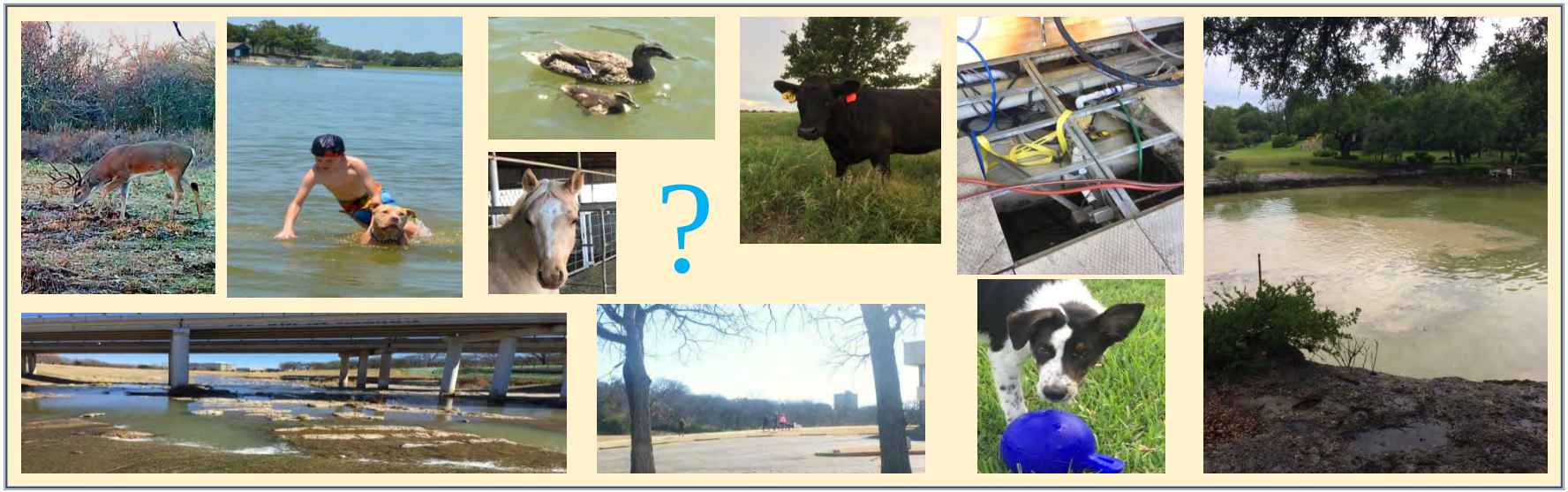
Bacterial Source Tracking Study

BST Assessment of Fecal Pollution Sources Impacting the Trinity River Floodway

trwd Tarrant Regional Water District

Objective

Ten sets of water samples were collected from each of 8 sites over an 10 week period in an attempt to identify sources contributing to bacterial loading in the Trinity River Floodway.



Method

2 Methods for E. coli Source Identification

The University of Texas Health Science Center at Houston (UTHealth) School of Public Health in El Paso

PCR Sample Analysis (Library Independent)

- Used during first 2 weeks of study

DNA RiboPrint (Library Dependent)

- Used for last 8 weeks of study

Supplemental Support Data Collected

Baylor University Chemical Tracers

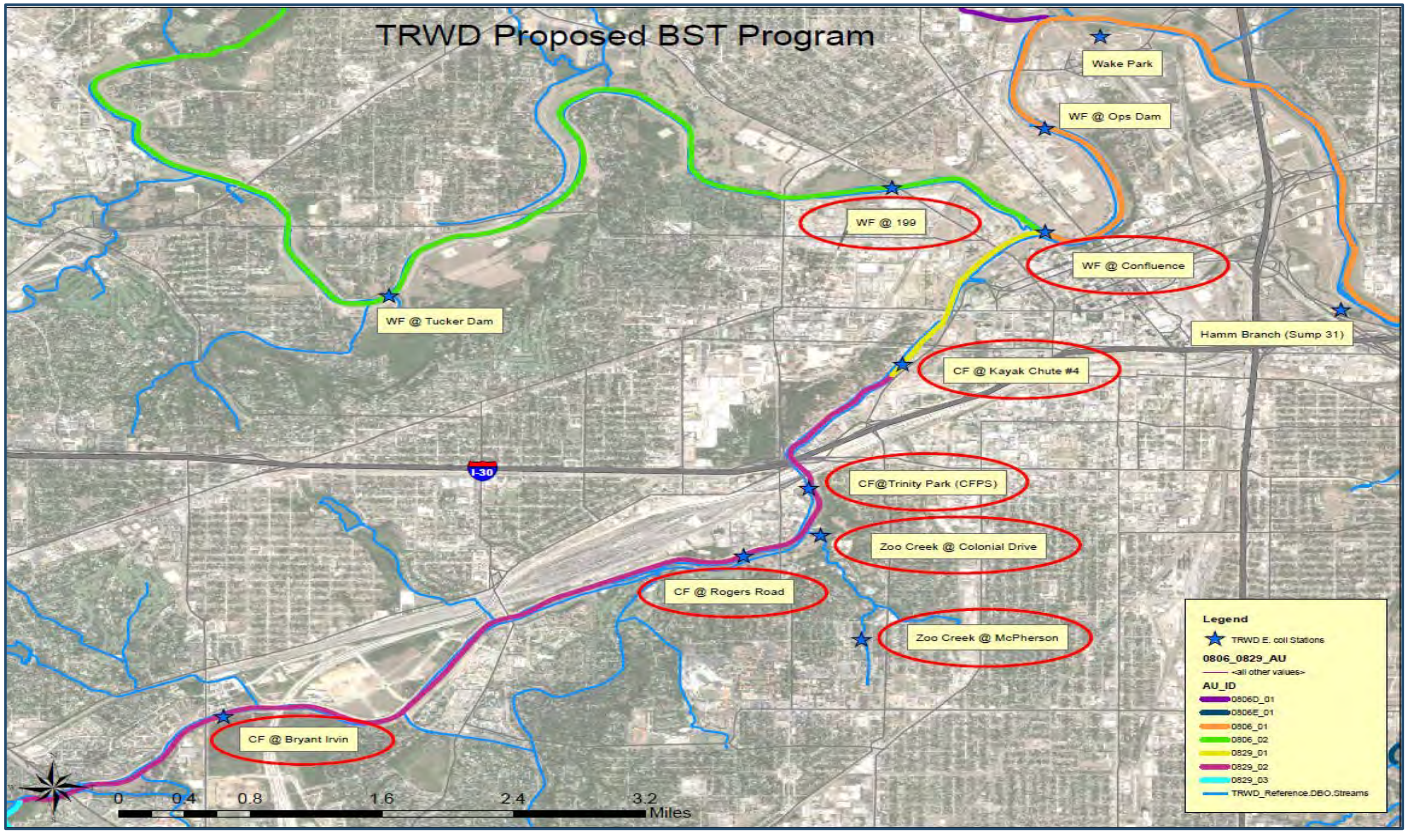
- Acetaminophen
- Caffeine
- Diltiazem
- Ketamine
- Sulfamethoxazole
- Benzoylecogine
- Carbamazepine
- Diphenhydramine
- Sucralose
- Trimethoprim

Pace Analytical Water Quality

- Temperature
- pH
- Dissolved Oxygen
- Specific Conductance
- Turbidity
- TDS
- NH3
- NOx
- TKN
- TP
- OPO4
- Chloride
- E. coli (Idexx)

Sampling Sites

- West Fork @ 199 (Henderson) (WF1)
- West Fork @ Confluence (WFC)
- Clear Fork @ Kayak Creek Chute #4 (CFK)
- Clear Fork @ Trinity Park (CFT)
- Zoo Creek @ Colonial (ZCC)
- Zoo Creek @ McPherson (ZCM)
- Clear Fork @ Rogers Rd (CFR)
- Clear Fork @ Bryant Irving (CFB)



TRWD BST of qPCR Data, Weeks 1-2

The first two sets of water samples were analyzed for the qPCR markers:

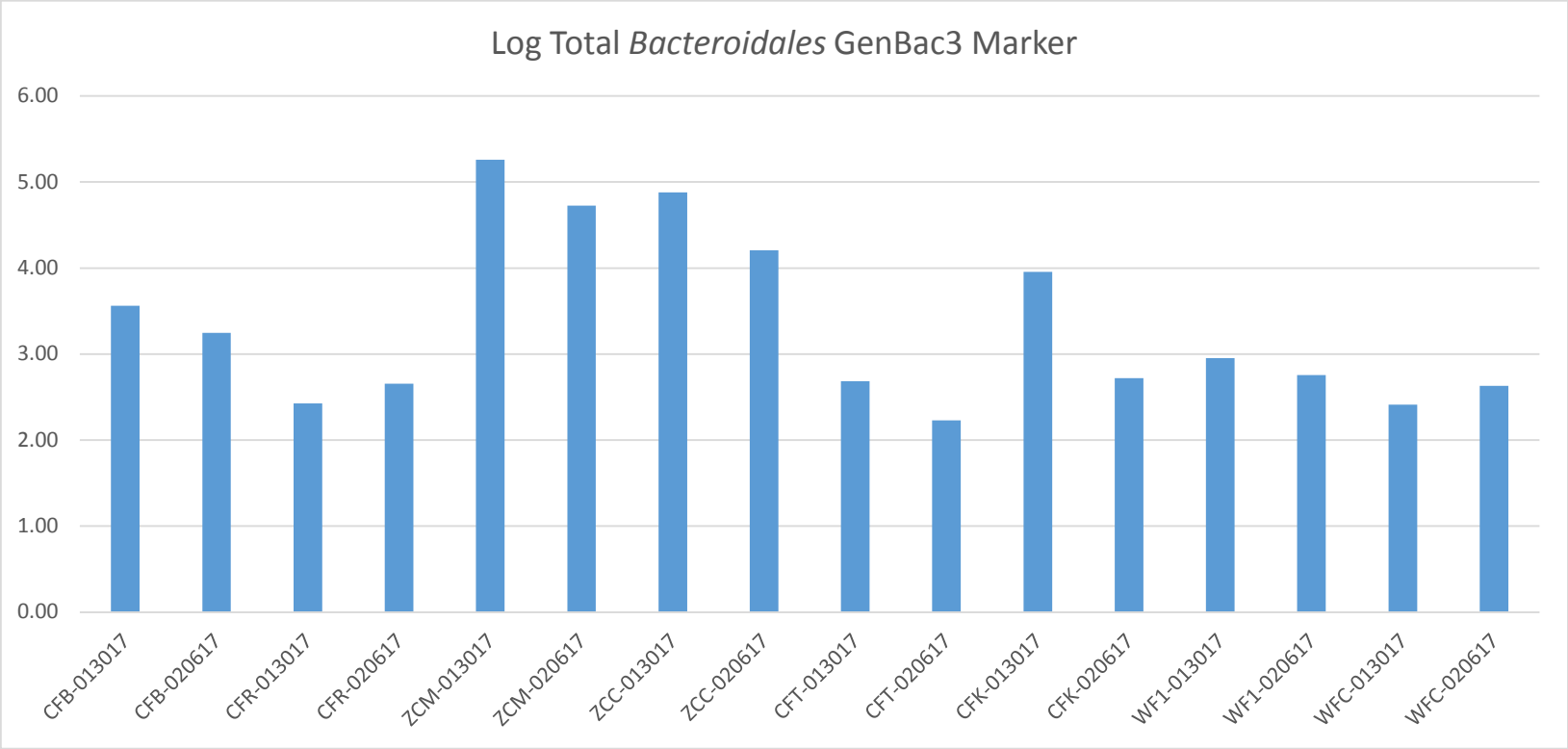
(1) ***Bacteroidales*** GenBac3 (general/non-specific)

a) **HF183/BacR287** (human)

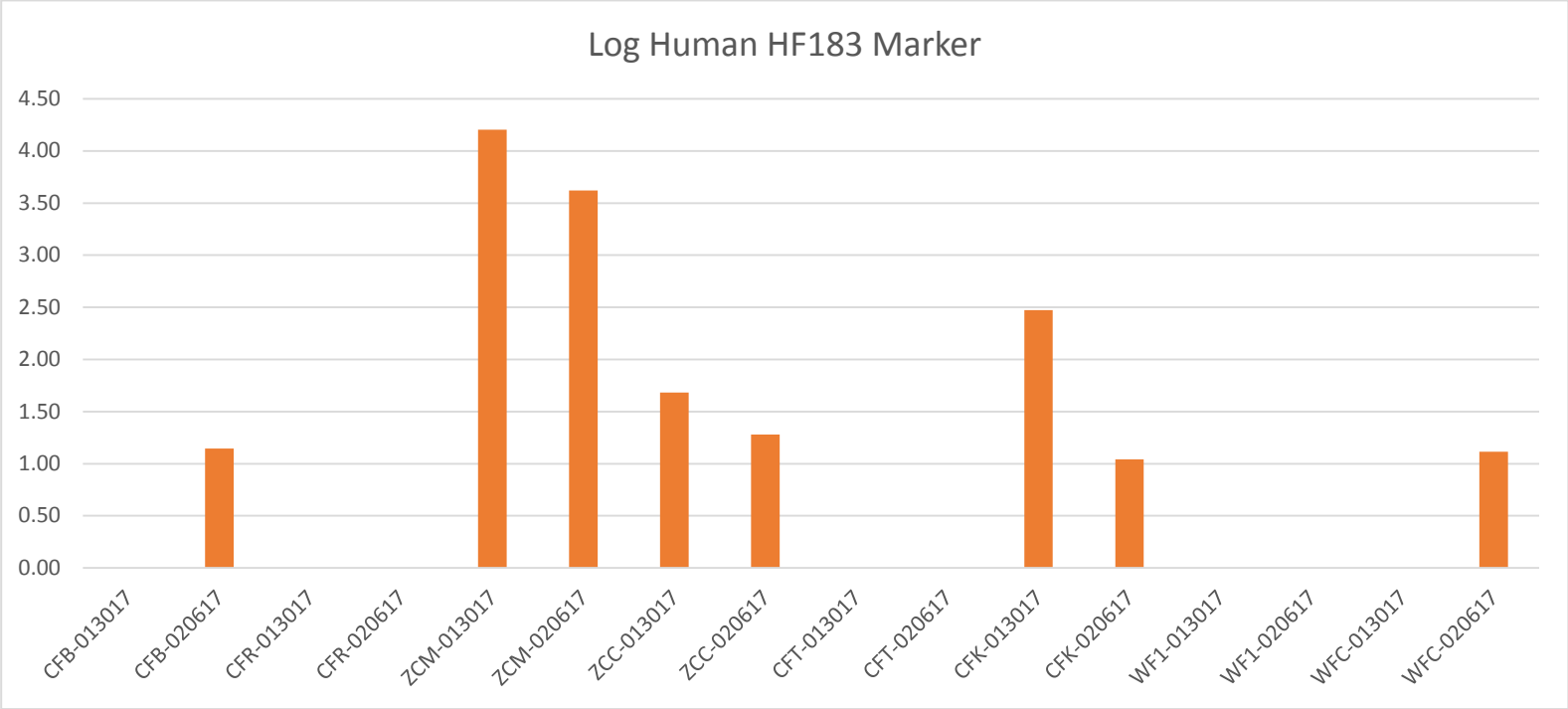
b) **BacCow** ruminant (and other animals)

c) ***Helicobacter*** **GFD** (bird)

qPCR Results Weeks 1&2

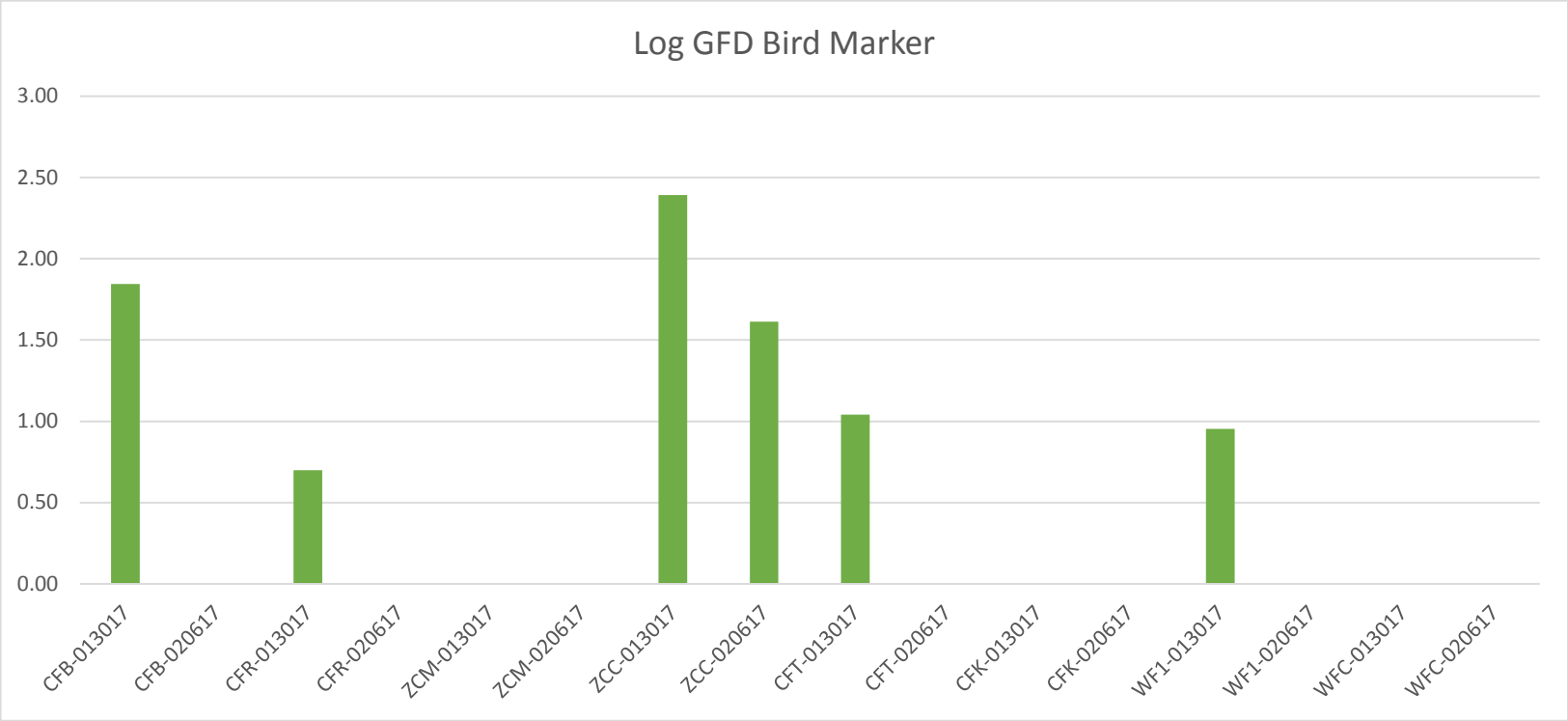


qPCR Results Weeks 1&2

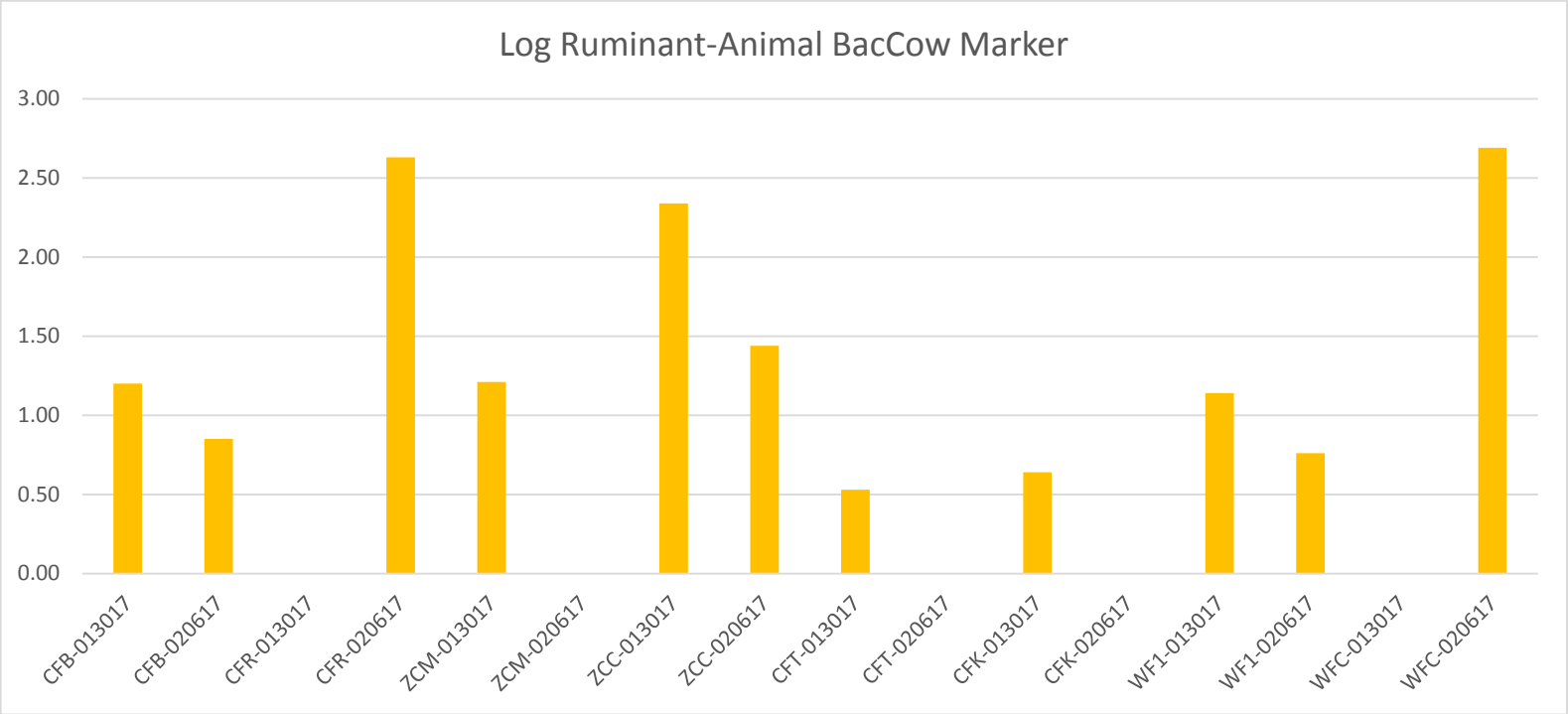


The Human HF183 Marker suggests that the site Zoo Creek at McPherson (ZCM) is *significantly impacted* by human fecal pollution; to a lesser extent, the sites Clear Fork at the Kayak Chute (CFK) and Zoo Creek at Colonial (ZCC) also appear to be impacted by human fecal pollution

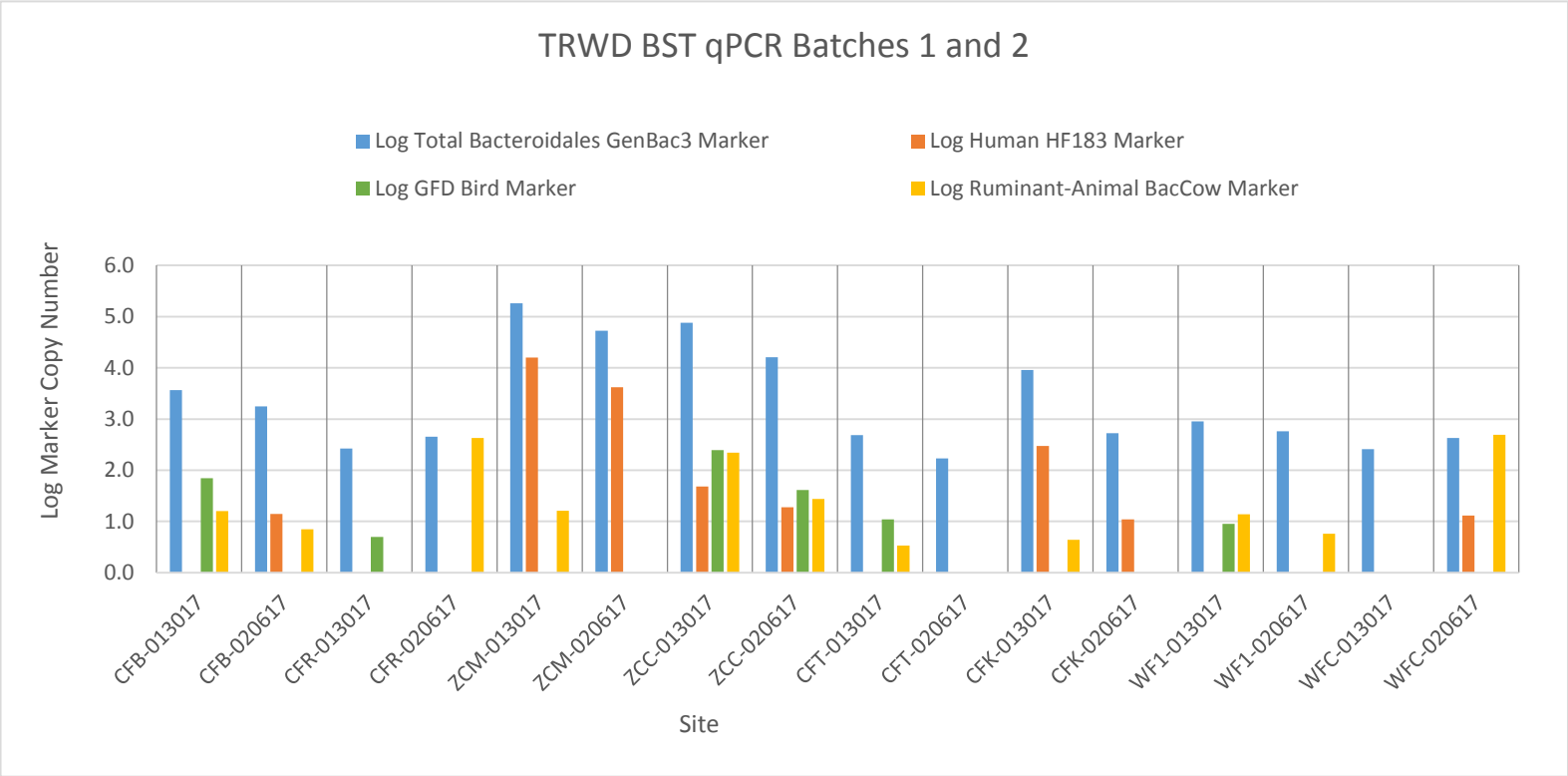
qPCR Results Weeks 1&2



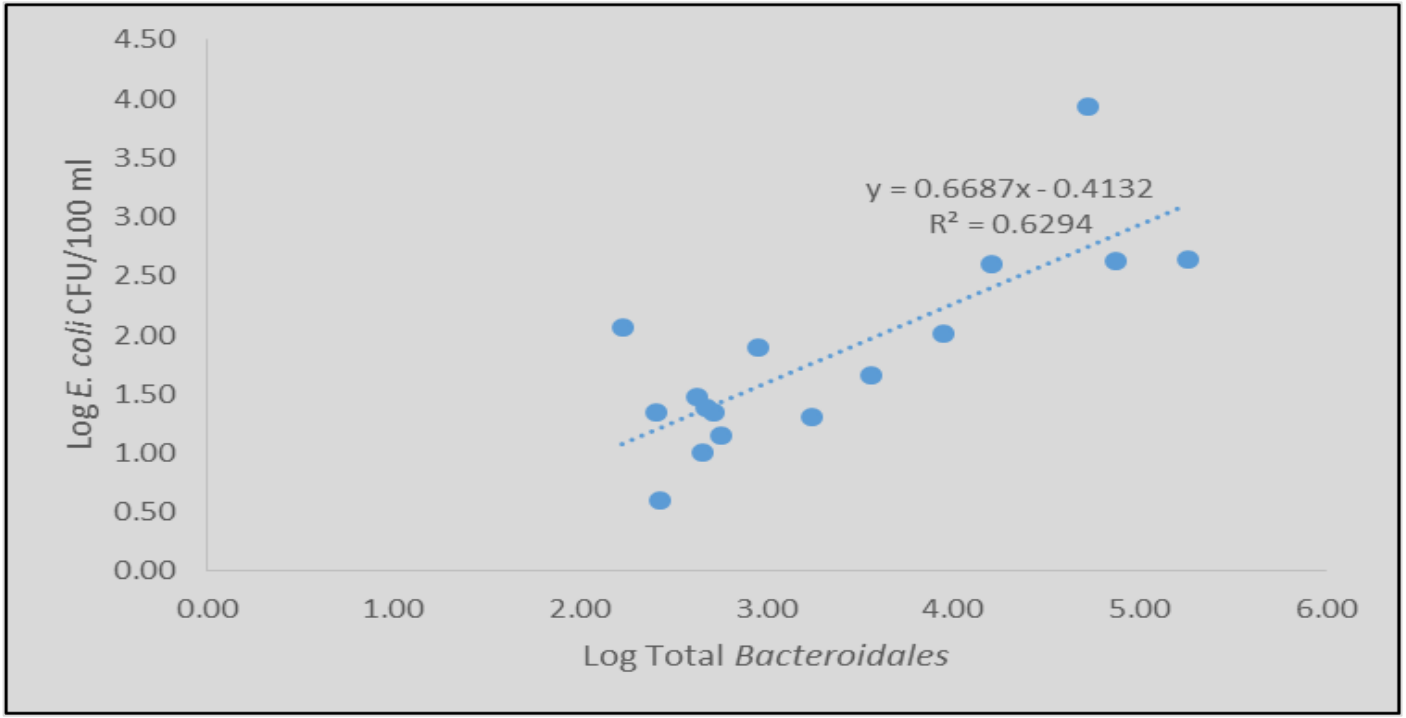
qPCR Results Weeks 1&2



qPCR Results Weeks 1&2



Log Total *Bacteroidales* vs Log *E. coli*



DNA Riboprinting/Library Dependent Analysis

- E. coli samples cultured on modified mTEC plates
- Up to 10 isolates collected from each cultured plate (some E. *coli* counts too low to collect 10 isolates)
- 615 isolates collected in total from 64 samples.
- ERIC-RP fingerprinting done and identified using the Texas E. coli BST Library (ver. 5-15)
- As with any BST method, percentages (or marker quantification) do **not** necessarily equate to percentage of fecal pollution loading--rather, results allow the identification and ranking of major fecal pollution sources.

4-Way Library Dependent Classifications

Human

- Sewage
- Septic

Domestic Animals

- Cat
- Dog
- Duck
- Turtle
- Cattle
- Chicken
- Duck
- Goose
- Guinea Fowl
- Turkey
- Donkey
- Goat
- Hog
- Horse
- Llama
- Pig
- Rabbit
- Sheep

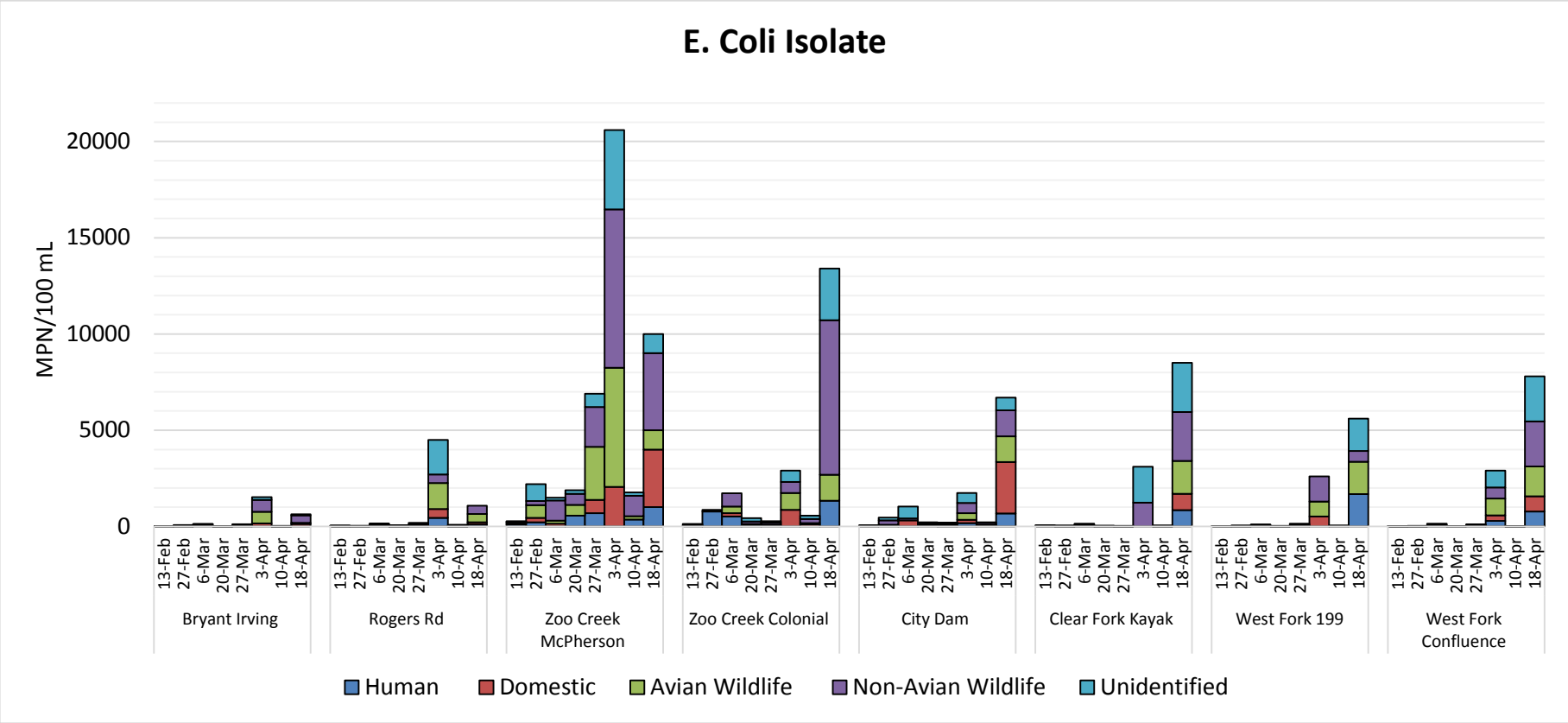
Avian Wildlife

- Blackbird
- Buzzard
- Blue jay
- Brown headed cowbird
- Crow
- Double crested cormorant
- Dove
- Duck
- Egret
- Goose
- Grackle
- Heron
- Killdeer
- Mockingbird
- Pelican
- Pigeon
- Roadrunner
- Gull
- Sparrow
- Starling
- Swallow
- Tern
- Turkey
- Vulture

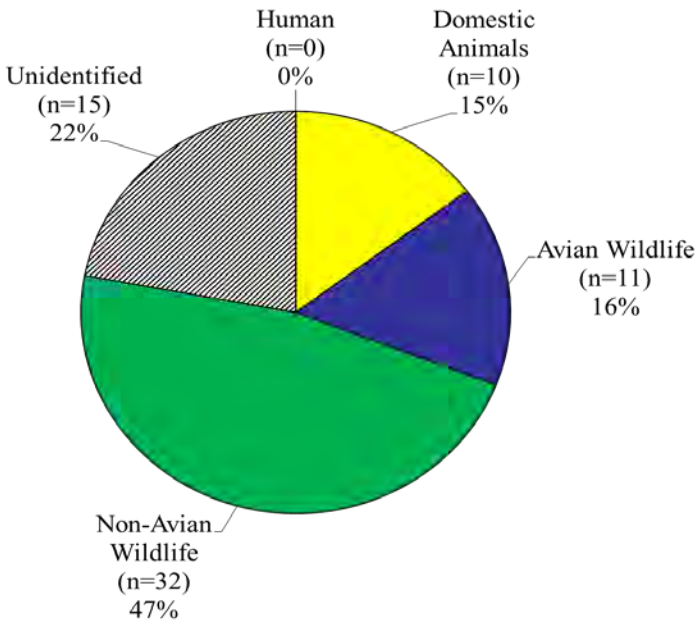
Non-Avian Wildlife

- Armadillo
- Beaver
- Bobcat
- Coyote
- Deer
- Feral hog
- Fox
- Javelina
- Mouse
- Opossum
- Porcupine
- Prairie dog
- Rabbit
- Raccoon
- Rat
- Skunk
- Squirrel
- Wolf

Isolate Data

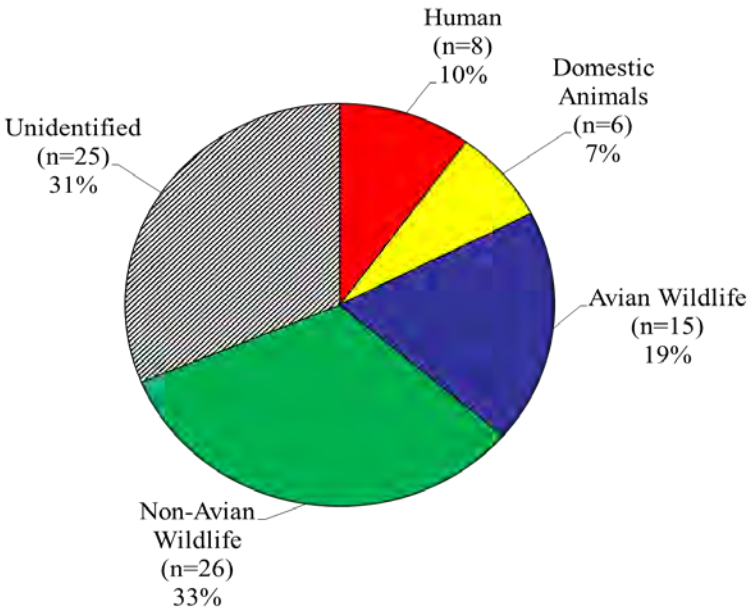


Clear Fork @ Bryant Irving



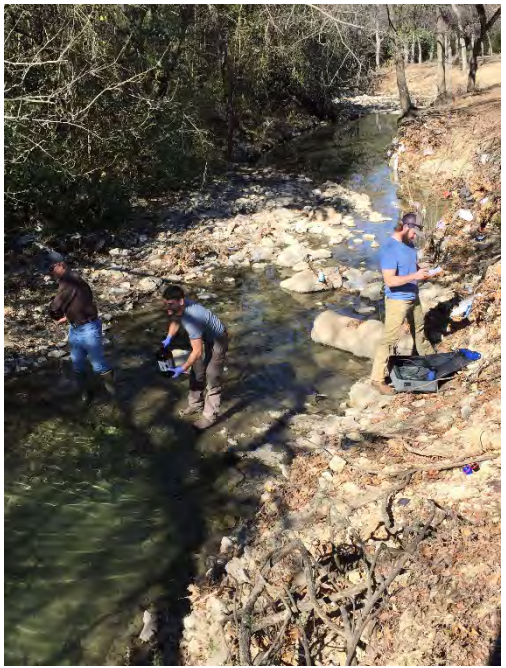
Geometric Mean = 65 CFU/100 ml (n=68 isolates; 8 samples)

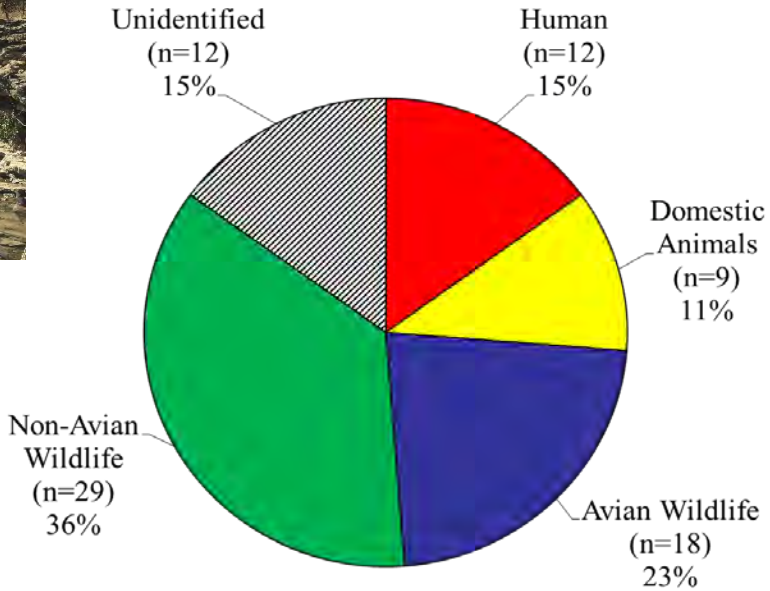
Clear Fork @ Rogers Rd.



Geometric Mean = 97 CFU/100 ml (n=80 isolates; 8 samples)

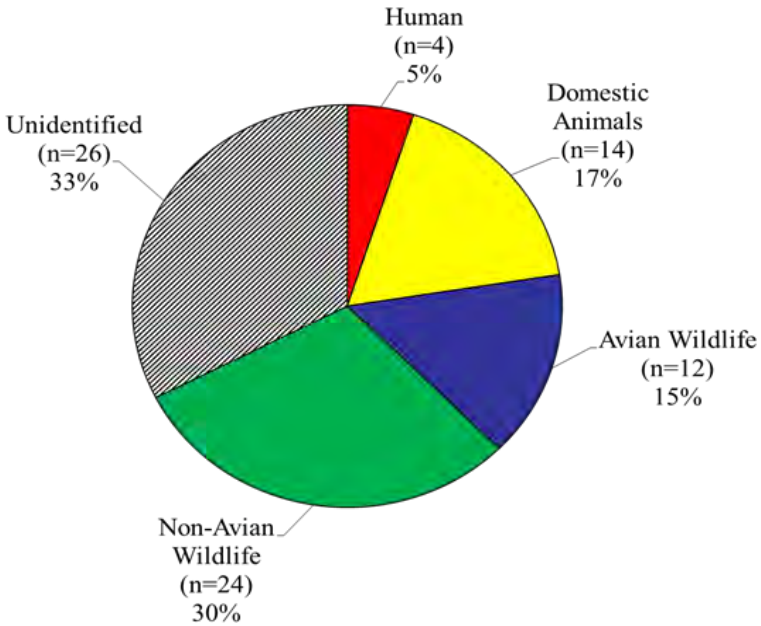
Zoo Creek @ McPherson





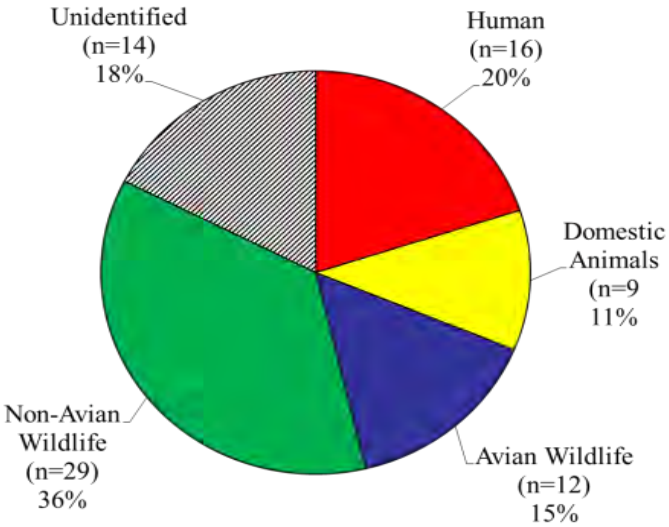
Geometric Mean = 2635 CFU/100 ml (n=80 isolates; 8 samples)

Zoo Creek @ Colonial



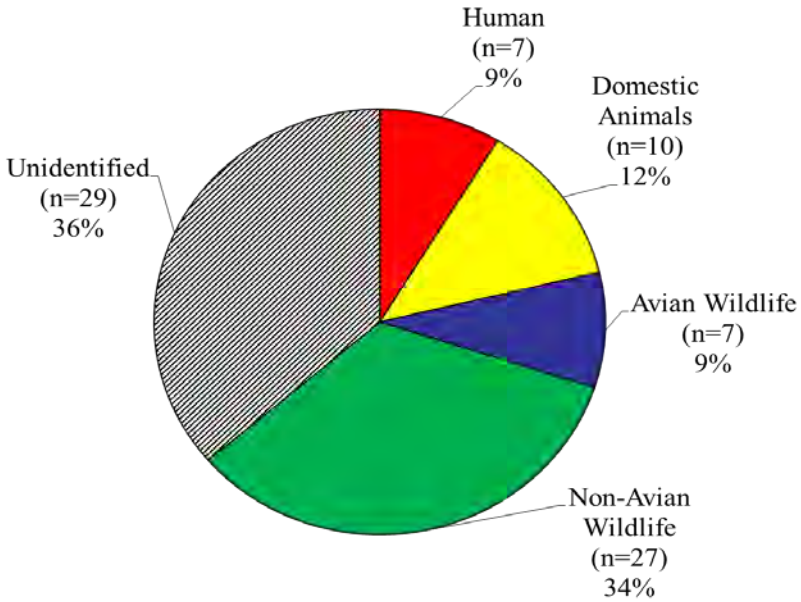
Geometric Mean = 786 CFU/100 ml (n=80 isolates; 8 samples)

Clear Fork @ Trinity Park



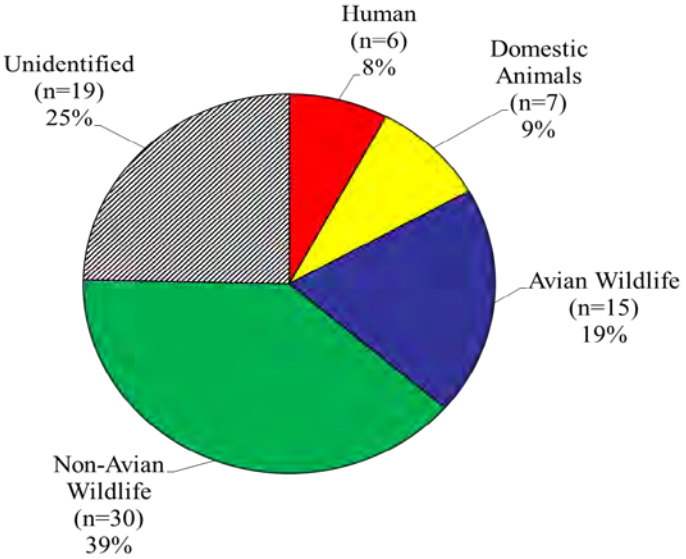
Geometric Mean = 314 CFU/100 ml (n=80 isolates; 8 samples)

Clear Fork @ Kayak Chute #4



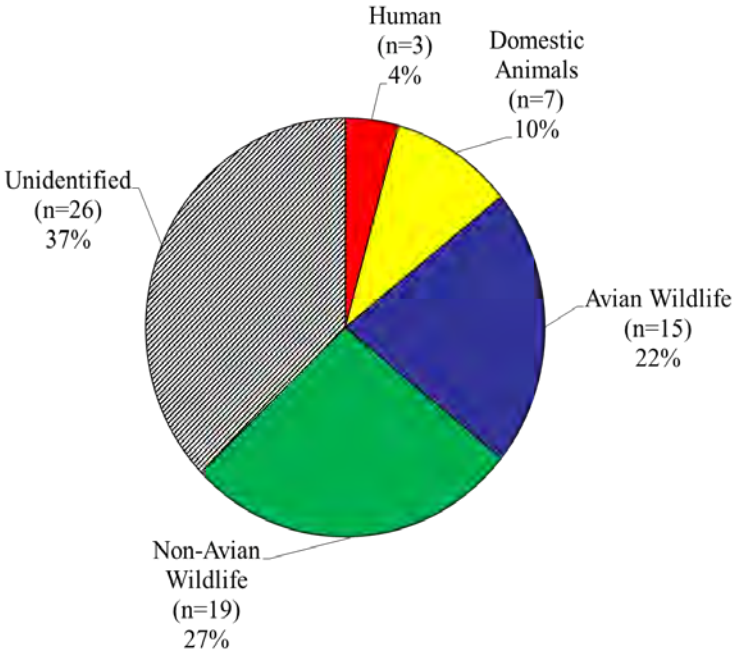
Geometric Mean = 122 CFU/100 ml (n=80 isolates; 8 samples)

West Fork @ 199 (Henderson)



Geometric Mean = 99 CFU/100 ml (n=77 isolates; 8 samples)

West Fork @ Confluence



Geometric Mean = 77 CFU/100 ml (n=70 isolates; 8 samples)

Four-way Split of Source Classes

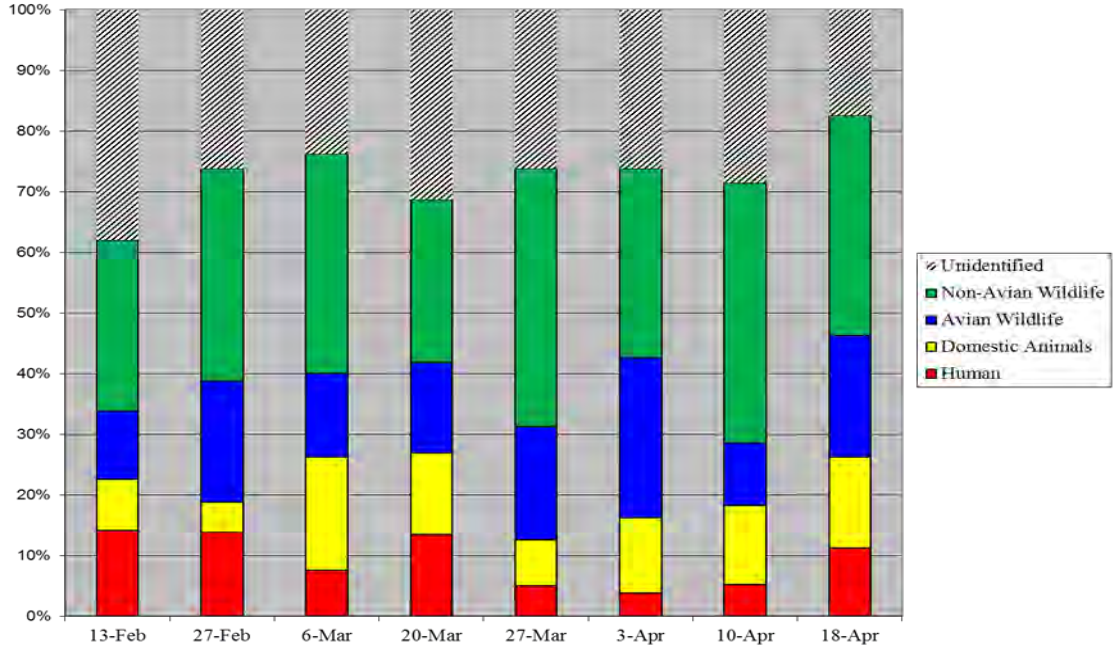
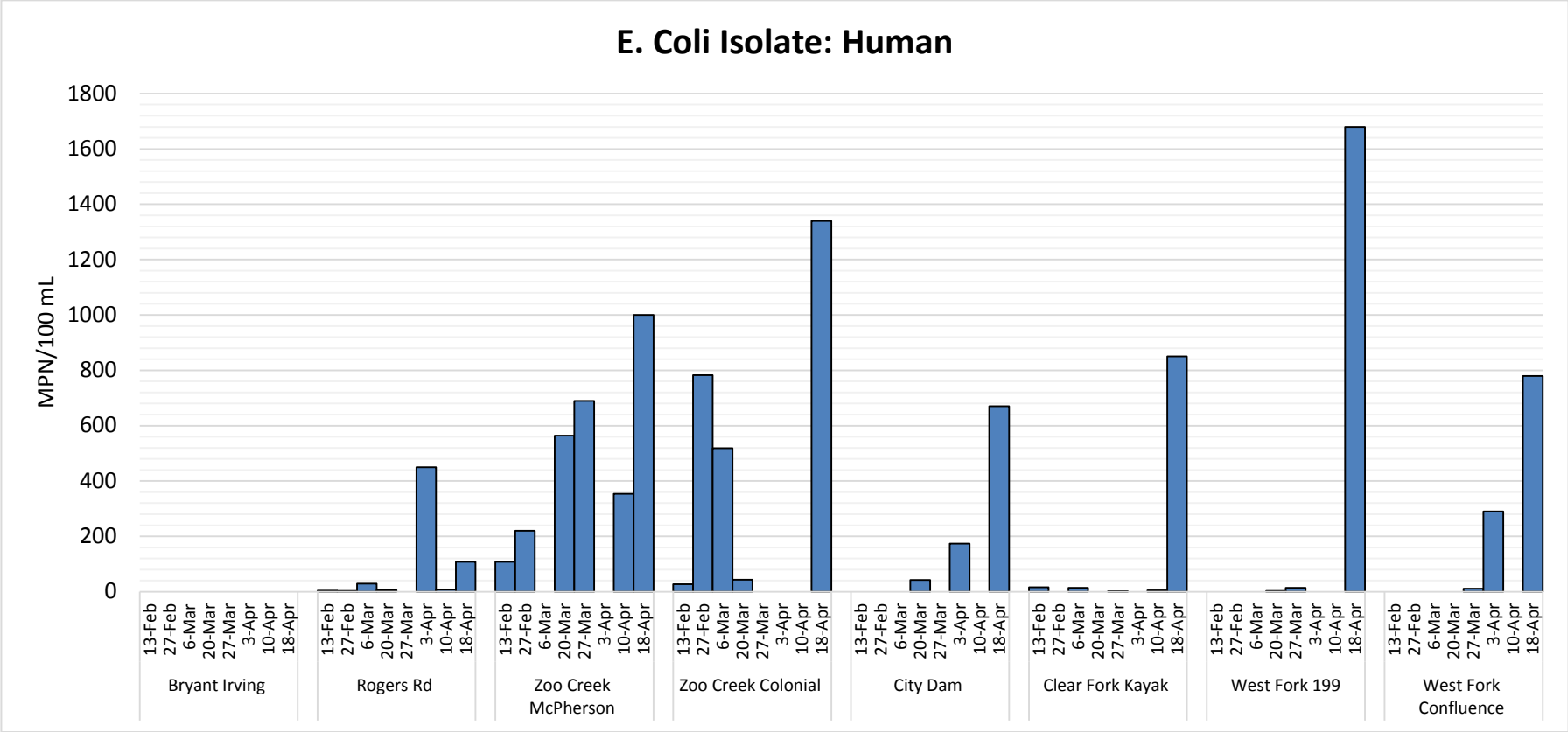
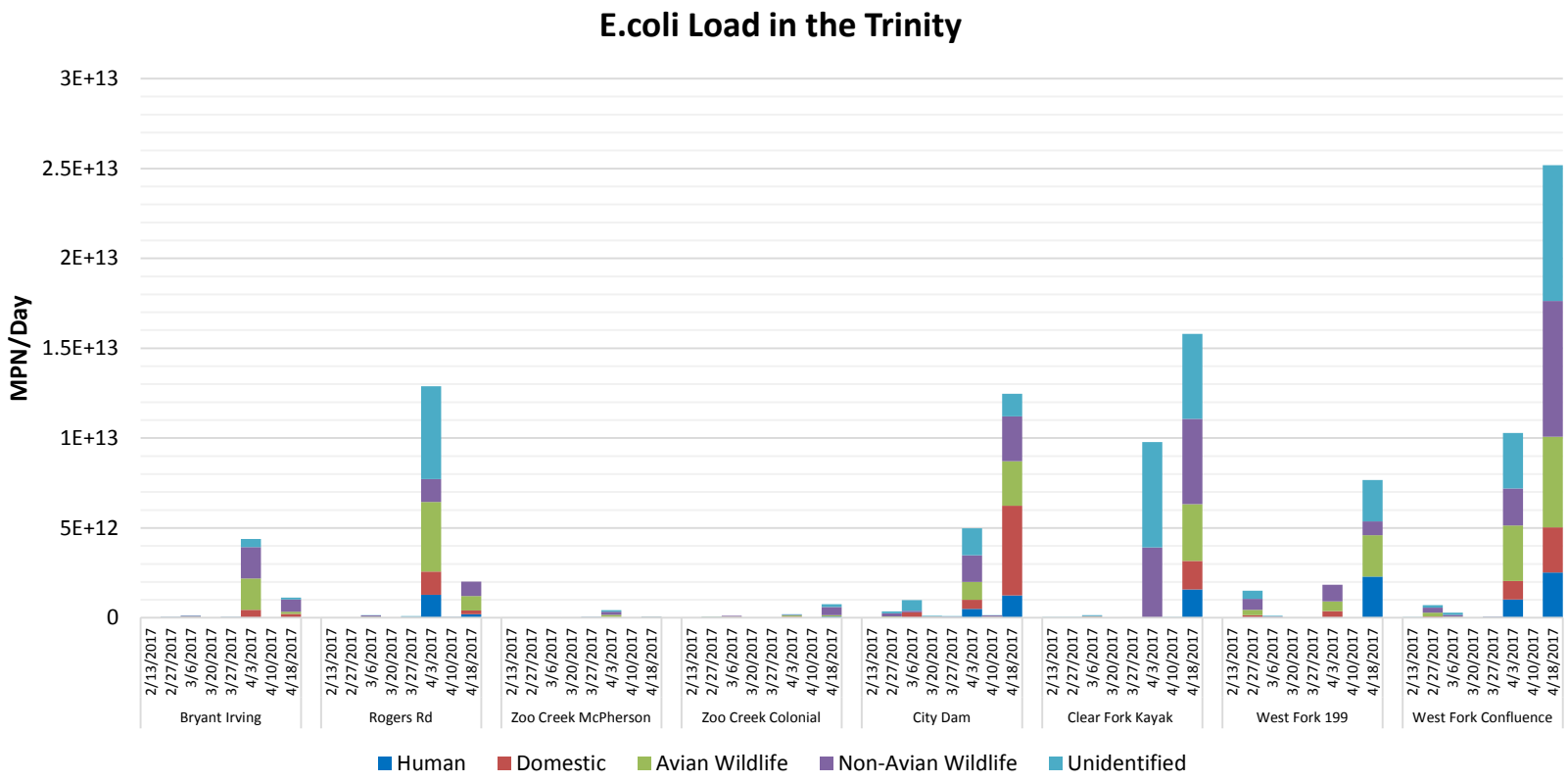


Figure 8: Identification of *E. coli* water isolates using a four-way split of source classes. Results are normalized to percent of isolates per sampling date (67 to 80 isolates per date; all sites combined). Note: samples taken 3-Apr and 18-Apr were after rain events.

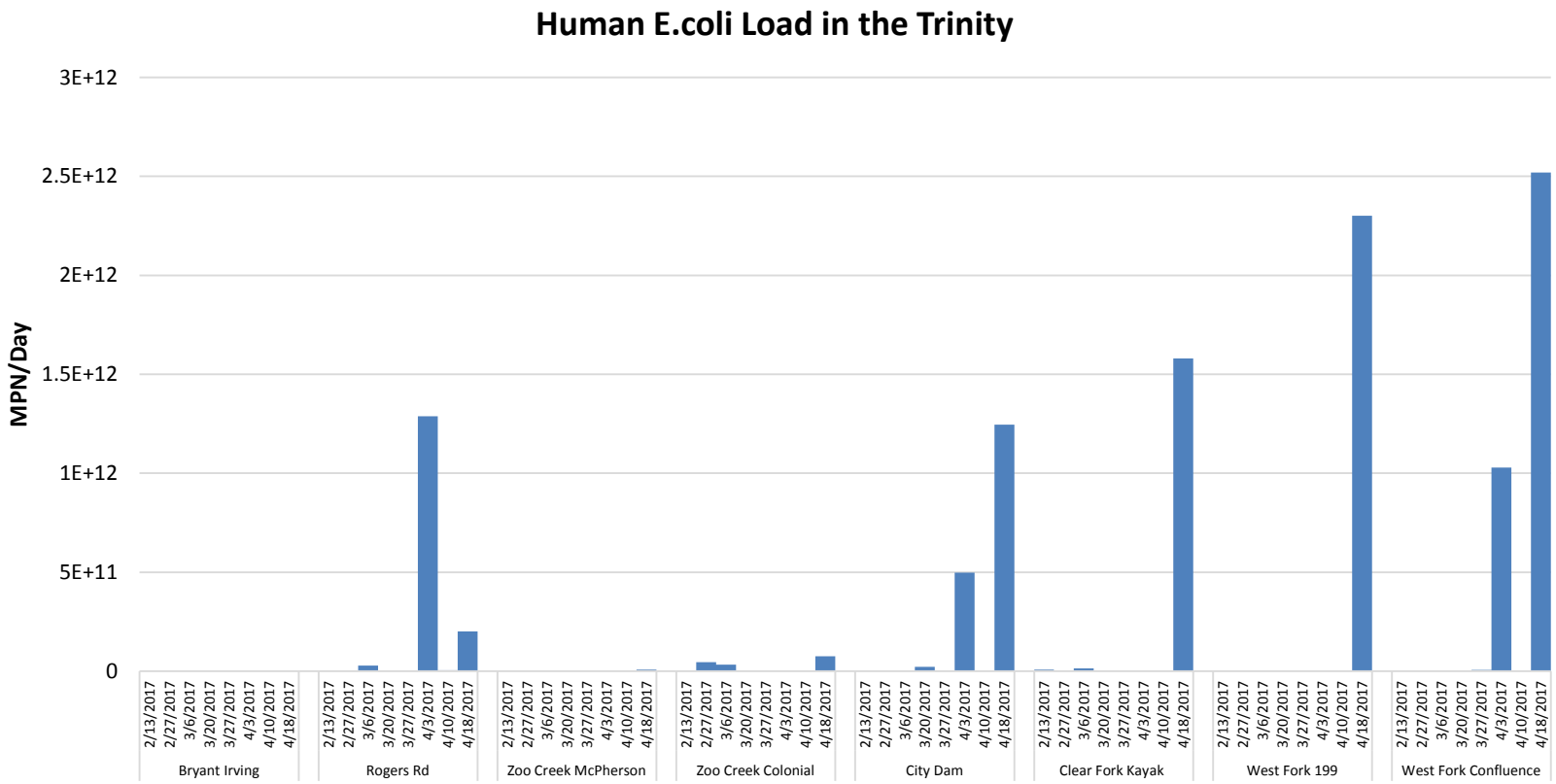
Isolate Data



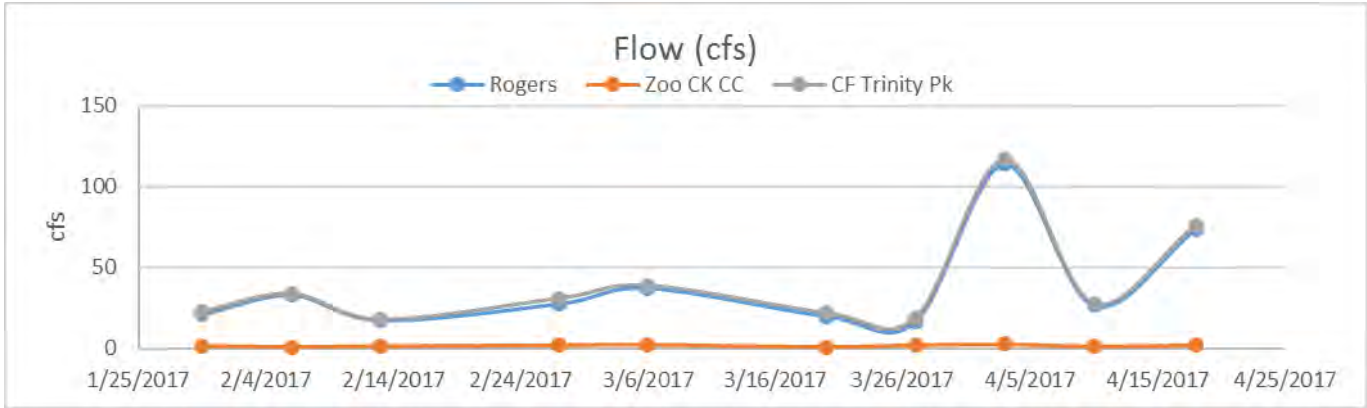
Load Data

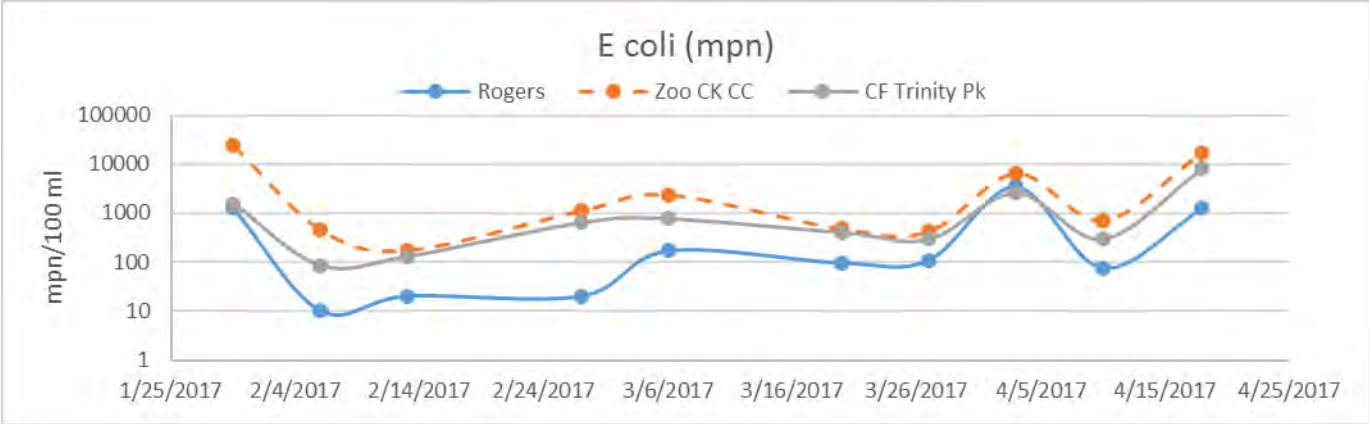

E.coli Load in the Trinity
MPN/Day
3E+13
2.5E+13
2E+13
1.5E+13
1E+13
5E+12
0
2/13/2017
2/27/2017
3/6/2017
3/20/2017
3/27/2017
4/3/2017
4/10/2017
4/18/2017
Bryant Irving
Rogers Rd
Zoo Creek McPherson
Zoo Creek Colonial
City Dam
Clear Fork Kayak
West Fork 199
West Fork Confluence
Human
Domestic
Avian Wildlife
Non-Avian Wildlife
Unidentified

Load Data

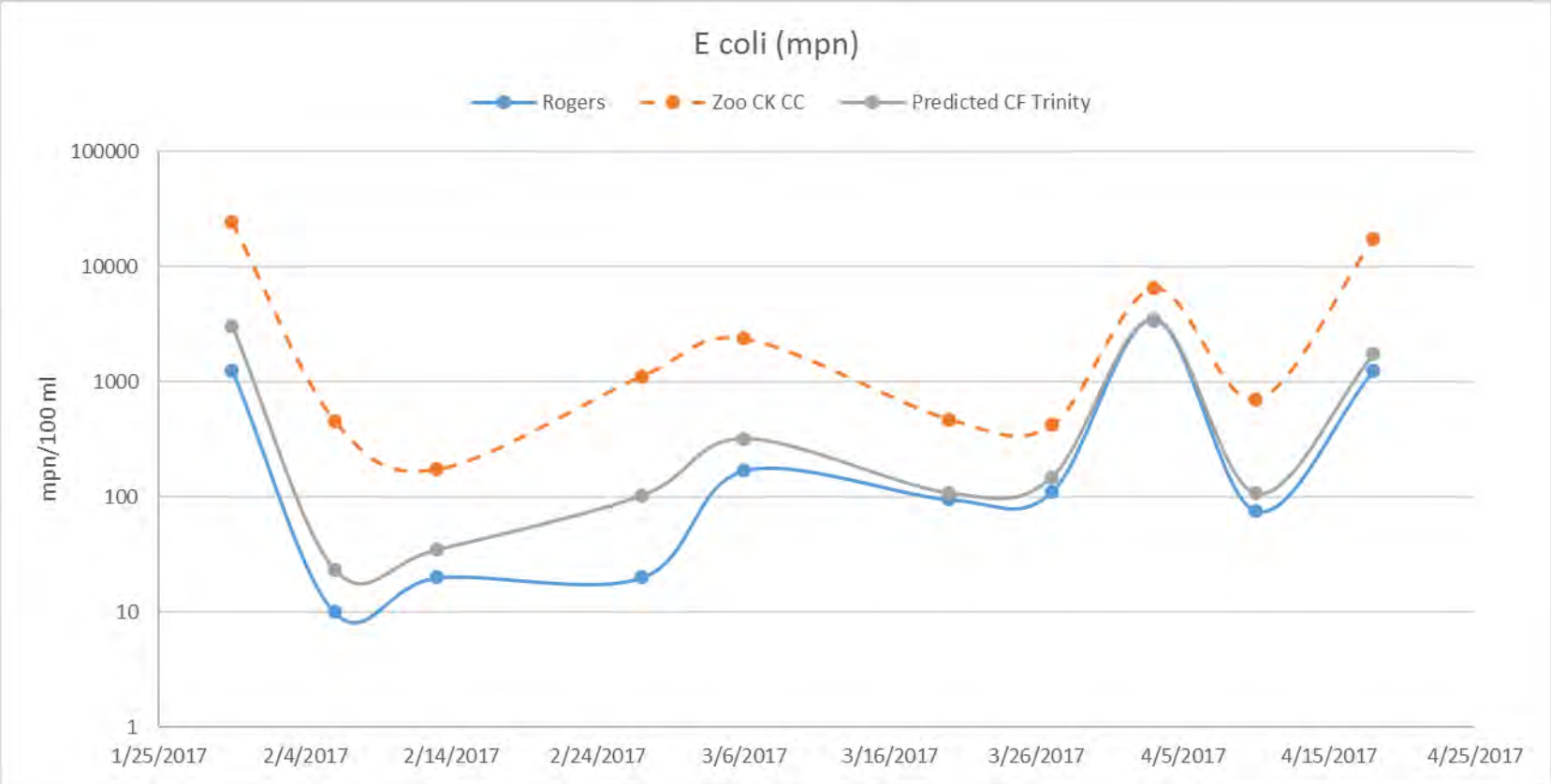

Human E.coli Load in the Trinity
MPN/Day
3E+12
2.5E+12
2E+12
1.5E+12
1E+12
5E+11
0
2/13/2017
2/27/2017
3/6/2017
3/20/2017
3/27/2017
4/3/2017
4/10/2017
4/18/2017
Bryant Irving
Rogers Rd
Zoo Creek McPherson
Zoo Creek Colonial
City Dam
Clear Fork Kayak
West Fork 199
West Fork Confluence

Mass Balance Study – Clear Fork at Trinity Park

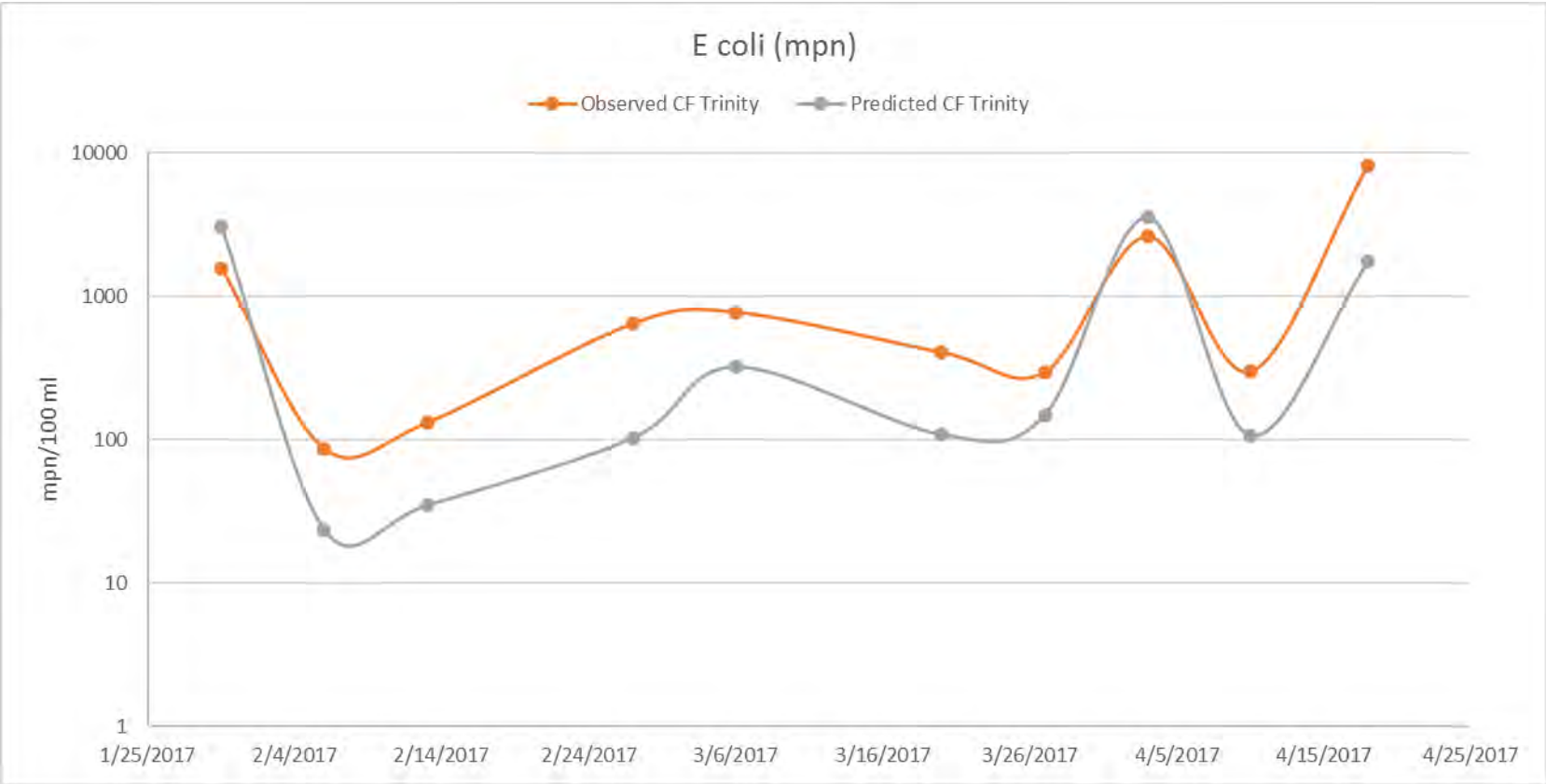




Mass Balance Study – Clear Fork at Trinity Park



Mass Balance Study – Clear Fork at Trinity Park

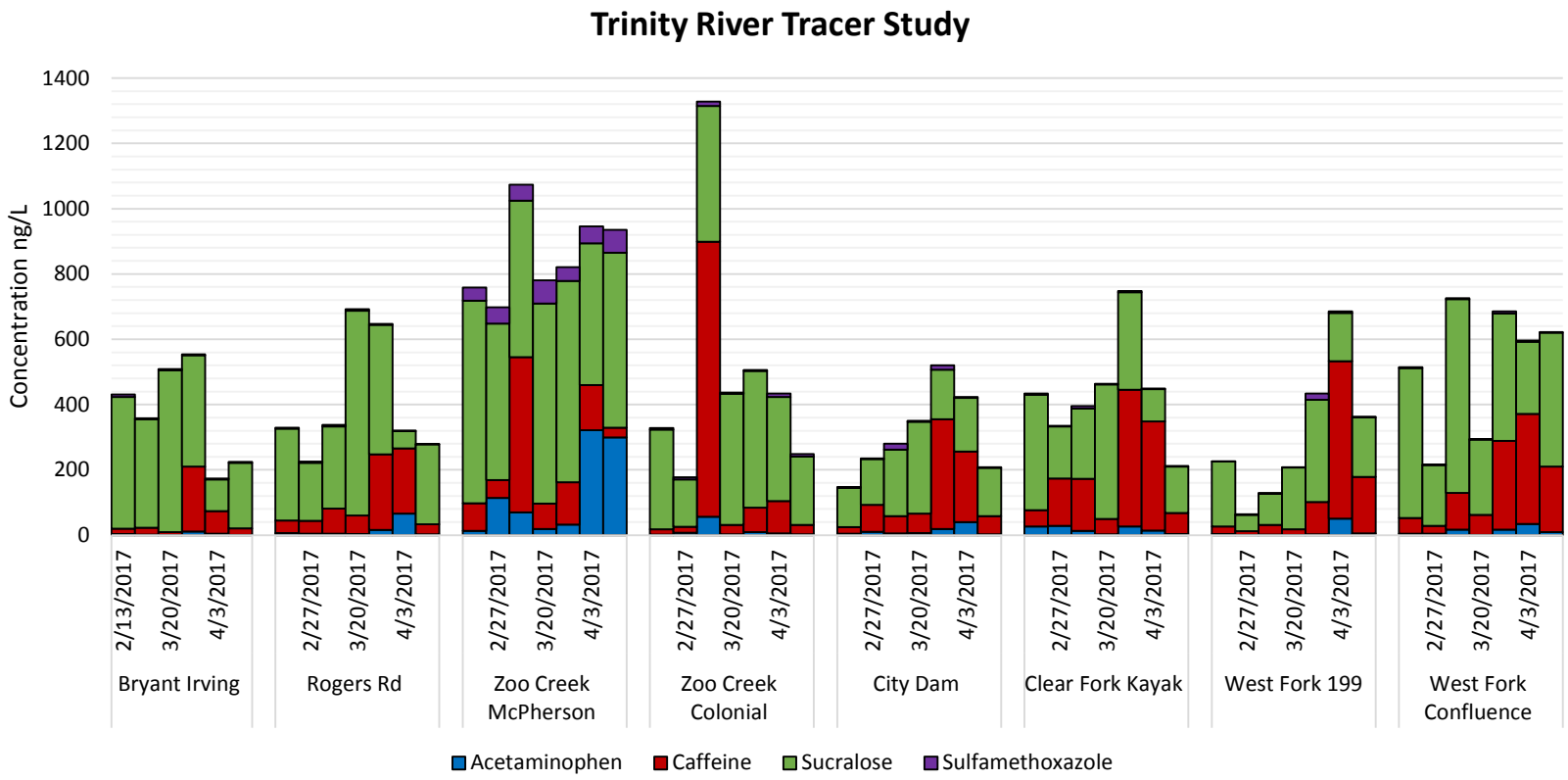


Trinity River Tracer Study

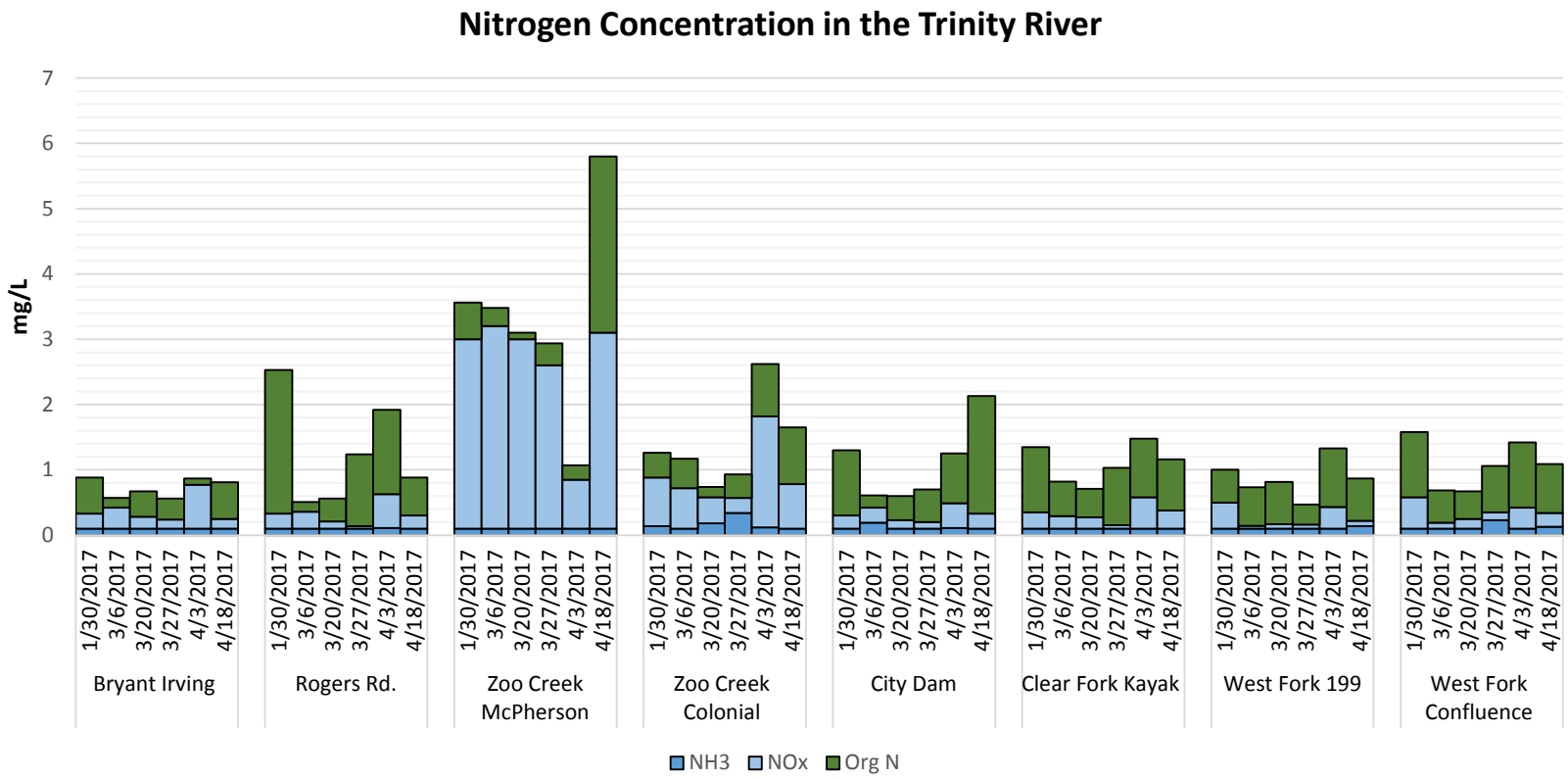
4 Tracer Compounds Used for Analysis:

- Acetaminophen (Pain Reliever)
- Caffeine
- Sucralose
- Sulfamethoxazole (Antibiotic)

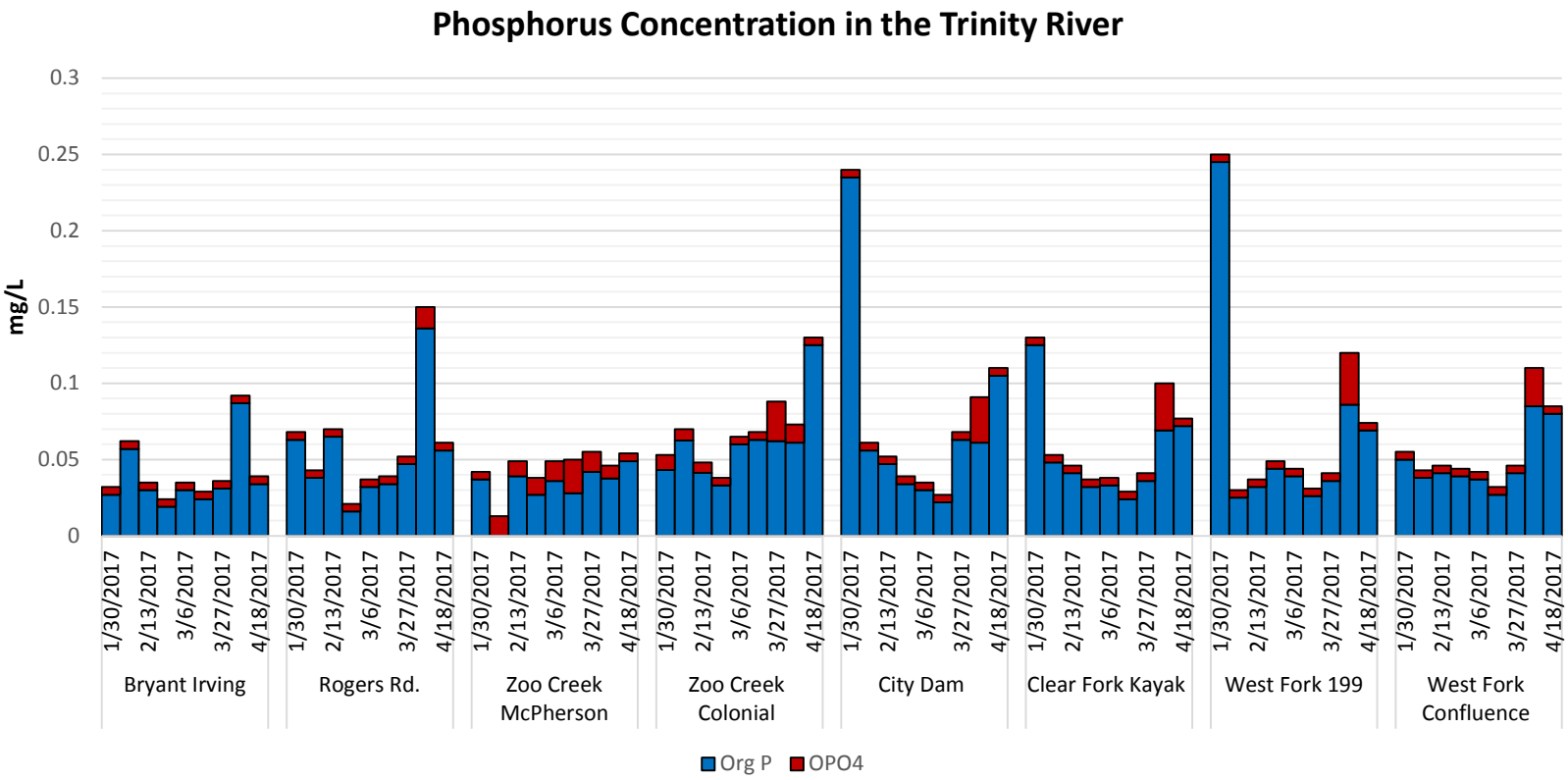
Tracer Study



Total Nitrogen



Total Phosphorus



Continuing Work….

Future Plans:

- Conduct Similar Study in Summer Months
- Document Avian Seasonality in Upper Clear Fork
- Add Nutria DNA to Texas Library
- Field Investigate Outfalls above Clear Fork @ Trinity Park Site