How One Small Environmental Lab Developed a Microbial Source Tracking Program

H. Halderman, D. Hernandez

San Antonio River Authority, Department of Environmental Sciences

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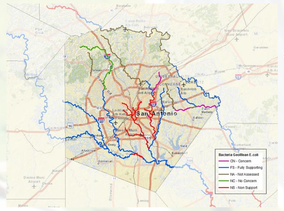

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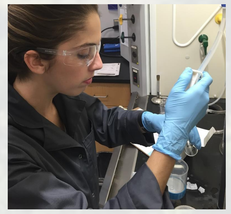

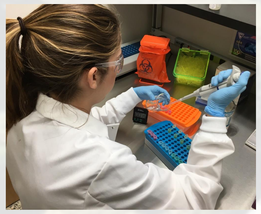

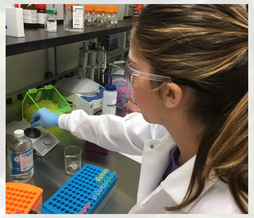

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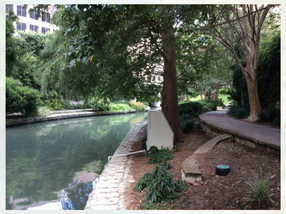

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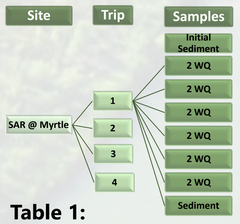

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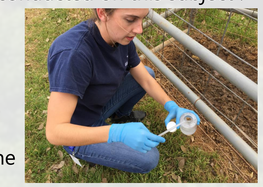

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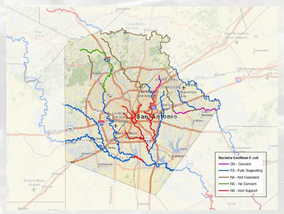

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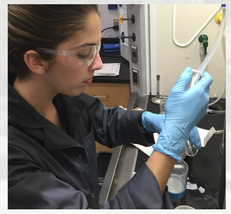

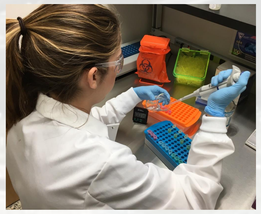

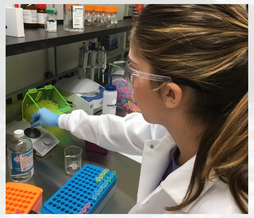

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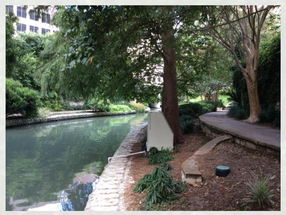

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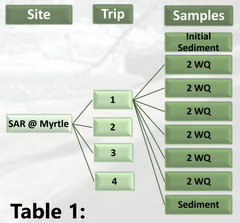

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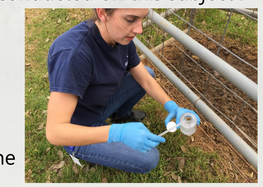

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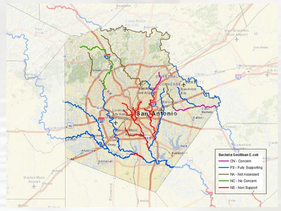

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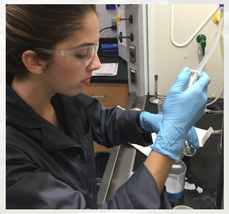

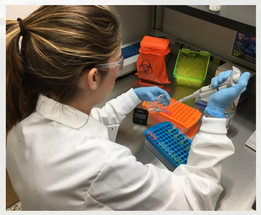

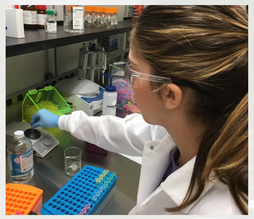

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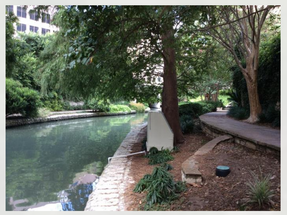

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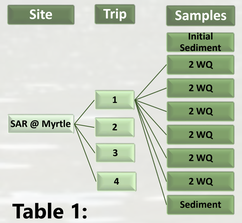

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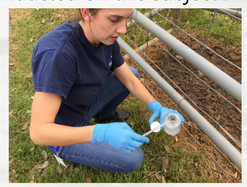

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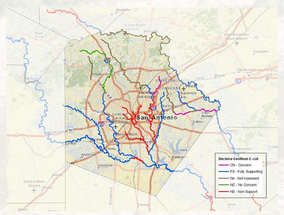

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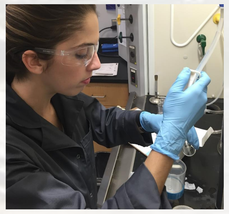

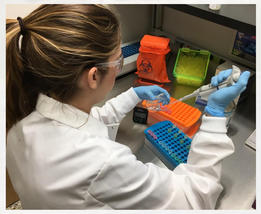

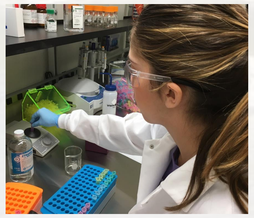

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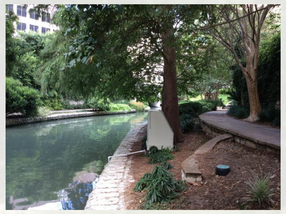

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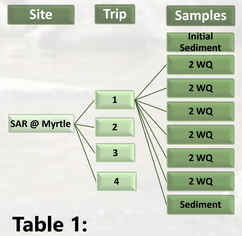

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How One Small Environmental Lab Developed a Microbial Source Tracking Program

H. Halderman, D. Hernandez

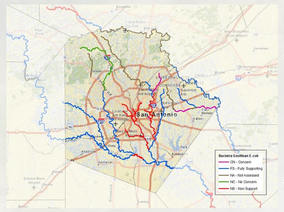
San Antonio River Authority, Department of Environmental Sciences

What is Microbial Source Tracking?

Microbial source tracking or MST identifies the source of fecal contamination in a waterway and can differentiate between species. This capability is an enormously helpful resource in mitigating bacterial loads in water bodies. However, laboratories with molecular biology capabilities and experience in the environmental monitoring realm are few and far between. Incorporating these methodologies into an environmental laboratory can be expensive, complex, and overall not feasible.

Objectives

- Improve the water quality of the San Antonio River Basin
- Identify the sources of bacteria found in our watershed to begin mitigating the bacterial loads to meet the TCEQ recreational waters criteria



- Reduce outsourcing of samples
- Become the first River Authority to provide molecular services such as MST
- Expand the River Authority's monitoring capabilities

Challenges

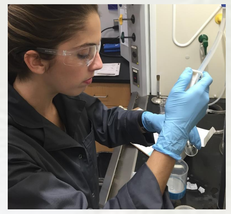
- Research and development of the analysis with few resources available to the team.
- Gathering support for the project
- Creation of a clean room & lab setup
- Staff training
- Method development

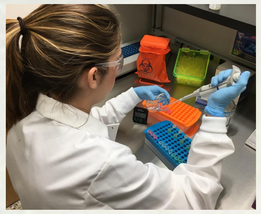
Method & Results

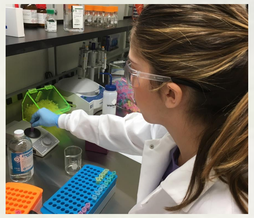
The first steps in creating a molecular biology section in an environmental lab was to start by researching what type of requirements would be needed to successfully analyze qPCR in house. Two analysts were sent to an "Intro to qPCR workshop" to start developing a strategy moving forward. Once the training was complete equipment was purchased and a lab was designated to run MST samples. The lab was found to require:

- A "clean room" dedicated to molecular biology techniques only.
- New equipment such as a -80°C freezer, a qPCR machine, stainless steel bench tops, and other smaller pieces of equipment.
- Brand new consumables that would be dedicated only to MST. This included pipettes, tubes, lab coats, a refrigerator, an ice maker, etc.

Once the laboratory was outfitted for microbial source tracking analysis work began on the method. The team sent analysts all over the nation from California to Virginia to learn from some of the best in the business how to preform MST analysis. With the help of many different labs and the review of numerous papers the team came together to create a standard operating procedure for MST.







The procedure developed includes four main steps:

- Filtration of the sample
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Comparative studies were performed using an outside lab to confirm the validity of the in house results. A DOC or demonstration of capability was also performed in house using a known human sewage sample and multiple replicates for additional confirmation.

References

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Current Studies

The most recent study conducted in the lab was the Urban Reach project. The project spanned one year and included riverbed scanning, quarterly monitoring of conventional water quality parameters and sediment, and microbial source tracking. The goal of this project was to monitor and characterize *E. coli* levels in water and sediment at two locations along the urban reach of the Upper San Antonio River, pictured below.

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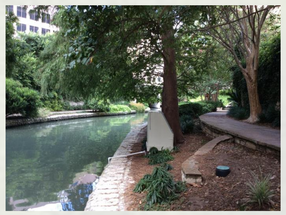

Samples were collected every 30 minutes over a 24 hour period at two sites along the San Antonio River. (Figure 1)

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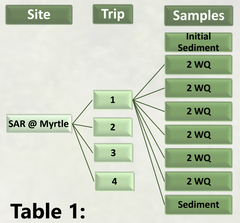

Due to the time frame of the project and the establishment of the MST parameter, MST samples were only collected in the last 2 quarters of the project. A corresponding *E. coli* MPN sample was analyzed with each MST sample. The MST samples were run in-house and outsourced to another lab for a comparison study (Table 1).

Table 1:

SAMPLE	SARA LAB		OUTSOURCE LAB			
	E. coli MPN/100 mL	Human BST	Human	Bird	Dog	Ruminant
SAR @ Myrtle 2/9/2016 14:39	46	Present	NA	NA	NA	NA
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Findings of the intensive monitoring events revealed bacteria levels can vary significantly when collected at the same monitoring site at different times of the day. Continued research is being conducted on this subject .

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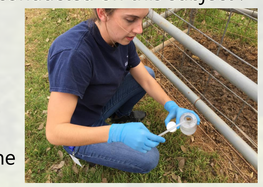

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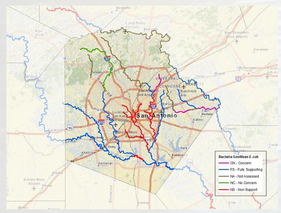

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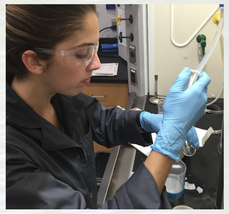

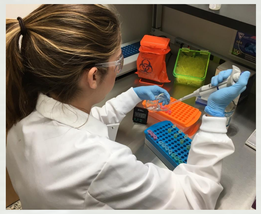

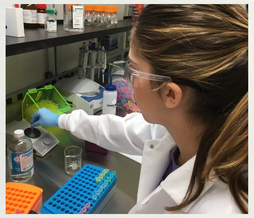

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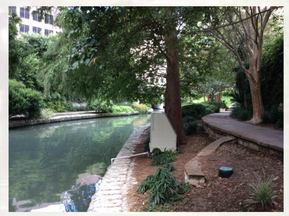

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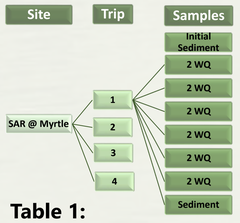

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