Sediment *E. coli*, Phase 2: Colony Enumeration and Sediment Habitat Preferences

Figure 1: Downstream view of sample location.

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Introduction

Over the past two decades, fecal bacteria, or *E. coli,* has become a concern for many stakeholders in the Trinity River basin, and throughout the state of Texas in general. According to the 2020 *Texas Integrated Report of Surface Water Quality for the Clean Water Act Sections 305(b) and 303(d)*, there are currently more than 300 waterbody segments in Texas listed as impaired for recreational use due to elevated levels of fecal bacteria. These impairments can sometimes be attributed to point sources of bacteria such as malfunctioning human sewage infrastructure, but commonly bacteria impairments arise from nonpoint source pollution that is delivered to the stream as runoff during storm events. However, streams may retain elevated levels of bacteria well after disturbances from storm events have taken place. Current scientific literature indicates that shallow bed sediments can be a significant reservoir of bacteria when resuspended by a disturbance event such as incoming stormwater runoff, floodwater erosion, or other in-stream physical agitation (wildlife, livestock, or human activity). However, the majority of these studies focus on coastal tidal zones, or along beaches of reservoirs or lakes. These are areas where flow velocity slows, where sediment conditions are more likely to be accretive, and where unconsolidated bed sediments are common. A literature review suggests that very little is known about this phenomenon in inland, eroding systems where particle sizes, sediment consolidation conditions, and fluvial geomorphology may differ from those conditions evaluated in preceding studies.

Furthermore, there is indirect acknowledgement that sediments can affect water quality; the Texas Commission on Environmental Quality's (TCEQ) *Surface Water Quality Monitoring Procedures, Volume 1: Physical and Chemical Monitoring Methods* (RG-415, Revised August 2012) provides guidance that seeks to minimize sediment disturbance when taking water quality samples. However, studies to evaluate sediment influence on water quality are limited.

A first phase of this study was conducted from 2018 to 2021 to determine the extent to which bacteria in sediment may affect water column concentrations. The report for Phase 1 of this study can be found on the Trinity River Authority Basin Planning Reports webpage. At the conclusion of the first phase, it was determined that another phase should be undertaken that would attempt to identify which specific sediment types may have the greatest effect on bacteria in water column concentrations.

Background

See the Sediment *E. coli* Phase 1 report for information on standards and assessment protocols for bacteria, as well as, the habitat preferences of bacteria. During Phase 1, it was noted that the variability in the sediments at each of the seven sites may have lead to difficulty interpreting the data. Sediment particle sites ranged from predominantly hard packed clay at some sites to predominantly gravel at other sites. The seven sites were located on five streams in two adjacent watersheds. Therefore, the climate and weather patterns were similar. Sites in each watershed represented both rural and suburban development. However, it was theorized that land uses in the immediate vicinity of each site may have contributed to the difficulty in drawing any overall conclusions from the data.

Study Area

In order to further address the main question of how much sediment *E. coli* concentrations affect water column concentrations, Phase 2 was developed to sample sites with various sediment particle sizes with as little spatial difference as possible between sites.

To that end, three locations that were in close proximity to each other in one stream were selected. These locations are in an area of Fish Creek that centers on station 15294 - Fish Creek South Branch at Great Southwest Parkway/Lakeridge Parkway in Grand Prairie (see Figure 2). This site was selected for several reasons. 1) It is near the TRA offices and allows for the frequency of sampling necessary to complete the project before the end the FY2020-2021 contract cycle. Due to weather related delays, the frequency was set to biweekly for six months. 2) TRA field staff are very familiar with this portion of the stream. 3) The site is wadeable and prior scouting determined that the three sediment size categories needed for this project were located near the bridge. 4) The flow at the site is flashy and returns to low/normal flows quickly after a rain event which helped ensure that the project was completed before the end of contract cycle. The location within the stream was considered representative of a small flowing system in a suburban area with bacterial loadings primarily from wildlife and pets and containing discrete areas of predominately fine sediments, sand, and gravel.

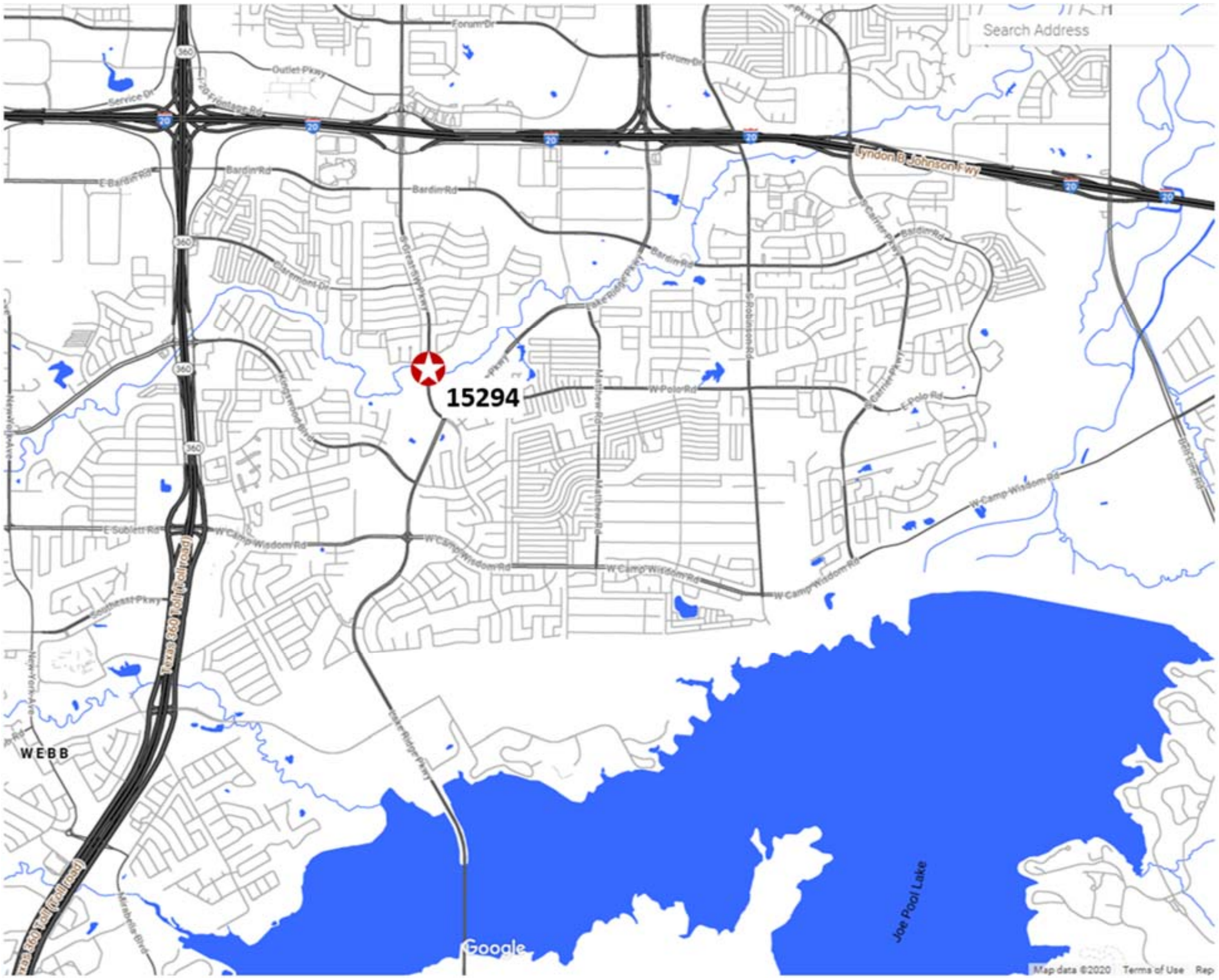

Figure 2: Location of sediment sampling site along Fish Creek

Methods

A project was begun in March 2018 to identify the extent to which bacteria in sediments may affect water column concentrations of *E. coli*. This project, Sediment *E. coli*: Phase 1, was initiated as a result of stakeholder interest during the development of the Village Creek-Lake Arlington Watershed Protection Plan. During Phase 1 of this project, samples were collected 12 times at seven locations within the Lake Arlington and Joe Pool Lake watersheds and included sediment and water column *E. coli* enumeration. Samples were collected across the width of the stream at each monitoring station and did not focus on any specific sediment size category within the stream.

Phase 2 of this project included 36 total sediment samples and 72 water column *E. coli* enumeration samples. However, sampling was conducted in one stream and focused on three sediment size categories at locations within this stream that were in relatively close proximity to each other. The goal of Phase 2 sampling was to determine if any sediment size category may be a preferred habitat for *E. coli* and to what extent bacteria in those sediments may be released into the water column during sediment disturbance events such as high flows and recreation.

Data Collection

Samples were collected approximately every other week - depending on weather and flow conditions - at three locations were within 20 meters of each other in Fish Creek in Grand Prairie. The three locations were, respectively, in an area that was predominately fine sediment, an area that was predominately sand, and an area that was predominately gravel. The exact location of these areas changed slightly over time due to redistribution of sediments after high flow pulses. However, based on previous work in this stream, it was determined that these three sediment size categories existed within less than a 1 kilometer reach that centers on station 15294 (Fish Creek South Branch at Great Southwest Parkway/Lakeridge Parkway in Grand Prairie) and had been recently been found directly under and adjacent to the bridge at this station (see Figure 3). Bacteria sampling activities consisted of the collection of sediment *E. coli* as well as water column *E. coli*. Water column *E. coli* samples were collected before and after artificial sediment disturbance.

Sediment samples were collected from a layer of sediment no deeper than 10 cm as significant contributions of *E. coli* to the water column from below this sediment depth are not expected under low to normal flow conditions. Research by Brinkmeyer et. al 2014 (DOI: 10.1016/j.scitotenv.2014.09.071) noted that *E. coli* in the sediments of two bayous in Houston, were consistently found to be highest in the top 1 cm and that there was "a significant order of magnitude or more reduction in [*E. coli*] and [enterococci] concentrations from the top 1 cm to the deeper 15, 30, and 60 cm horizons". Further, Pachepsky and Sheldon 2011 (http://dx.doi.org/10.1080/10643380903392718) noted that there was a more than twofold decrease in *E. coli* levels in 5 cm increments down to a depth of 15 cm in a rural Maryland creek.

Data generated during this project were not intended for submittal to SWQMIS for the following reasons. 1) Sampling was conducted on an approximately biweekly basis depending on weather and flow for a total of 12 samples. 2) Several individual samples were collected in close proximity to each other and not intended to be a single composite sample. 3) Water column samples were not collected in the centroid of flow but rather immediately adjacent to the area of the stream containing the desired sediment size. Additionally, the post disturbance *E. coli* samples were collected outside of normal CRP sampling protocols.

Figure 3: Sampling area.

A water column *E. coli* sample was collected prior to disturbing the sediments. Sediment *E. coli*, size, TOC, and percent solids data were then collected. An additional water column sample was collected after sediment has been disturbed and re-suspended. All data generated for this project was then used for data analysis to determine if any specific sediment size class affected the concentration of *E. coli* in the water column.

To ensure that samples were collected under flow severities of low to normal flow, staff regularly consulted weather data for the region to track recent rainfall, and scheduled sample collection accordingly. Samples were not collected during precipitation events that would generate runoff. It was expected that there would be limited sediment mobility under those flow conditions which should reduce background influences from sediments in the pre-disturbance samples. Therefore, high and flood flow conditions were not sampled. Additionally, the site was inspected prior to sampling to ensure that bed sediments were not being excessively mobilized at low and normal flows due to outside influences such as animal or human activity and channel disturbances such as construction and erosion. If it was apparent that bed sediments were being resuspended due to these influences, sampling was delayed until a time when the channel has stabilized and there was reduced sediment movement.

The following tasks were completed during each sample event:

1) Upon arrival at each site, an area of predominantly fine sediment, an area of predominantly sand, and an area of predominately gravel were identified. These locations were approached from downstream to avoid disturbing the sediment upstream of the sample areas.
2) A pre-disturbance water column *E. coli* sample was collected directly above each of the areas containing the desired sediment size.
3) Field staff collected a sediment sample for laboratory analysis of *E. coli* and sediment conventionals at each of the three locations. These samples were collected with a pre-cleaned metal scoop and placed into a pre-cleaned metal pan (see Figure 4). Cleaning prior to sampling consisted of spraying all equipment with denatured alcohol and then wiping with a paper towel until dry.
4) The sediments in each area were disturbed by kicking through the area containing the desired sediment size.
5) A post-disturbance water column *E. coli* sample was collected from the sediment plume that had been kicked up.
6) Because flow in the planned work area was generally low and the channel was wide, there was limited potential for cross-contamination from one sediment area to another. However, to further reduce the potential for cross-contamination and to the extent possible, the previous steps were conducted simultaneously by three individual field staff members to reduce the amount of time any water may have had to travel between the sampled areas. The exception to this being that the predominantly sandy area was frequently located immediately downstream of the predominantly gravel area. Therefore, the samples for the upstream gravel area were not collected until all collection activities for the downstream sandy area were completed during each event.
7) A sonde was placed upstream of the disturbed area for determination of field parameters.
8) Instantaneous flow was measured.
9) Metals pans and scoops were cleaned first by washing all sediments and residue from the equipment in the stream followed by triple rinsing. Pans and scoops were then sprayed with denatured alcohol and wiped dry with a paper towel.
10) Sediment *E. coli* and sediment conventionals samples were then shipped overnight to Ana-Lab for analysis.
11) Water column *E. coli* samples were delivered to TRA CRWS lab for analysis.

Figure 4: Sediment sample collection. From left to right - fine sediment, sandy sediment, and gravel.

Data Analysis

Comparing Pre- and Post-Disturbance *E. coli* Enumeration

As in Phase 1, pre-disturbance and post-disturbance *E. coli* enumerations were compared first in order to observe any changes. For each sediment type (fine, sandy, gravel), 12 pre-disturbance and 12 post-disturbance samples were taken for the comparison, for a total of 36 samples. 88.89% of the post disturbance samples across all three sediment types showed an increase in bacteria count. After calculating the pre- and post-disturbance *E. coli* geomeans for each sediment type and then using those values to find the relative percent increase of each, there was marked increase in each type. Sandy sediment had the lowest increase at 82%, Gravel sediment had a 101% increase, and the fine sediment had a relative percent increase of 1045%, the largest by a margin of over 900%. When comparing the post-disturbance sediment *E. coli* geomean for each sediment type, the fine sediment had a much higher geomean than the gravel or sandy sediment (see Table 1 and Figure 5).

Table 1: Sediment E. coli geomean and relative percent increase for sediment types

Sub-areas of Station 15294 - Fish Creek at Great Southwest Parkway in Grand Prairie	Pre-Disturbance *E. coli* Geomean (MPN/100 mL)	Post-Disturbance *E. coli* Geomean (MPN/100 mL)	Relative Percent Increase	Sediment *E. coli* Geomean (MPN/ 100 g)
Fine Sediment	133.73	1,531.58	1045%	53,432.69
Sandy Sediment	249.74	453.45	82%	8,424.89
Gravel	222.03	445.18	101%	4,461.44

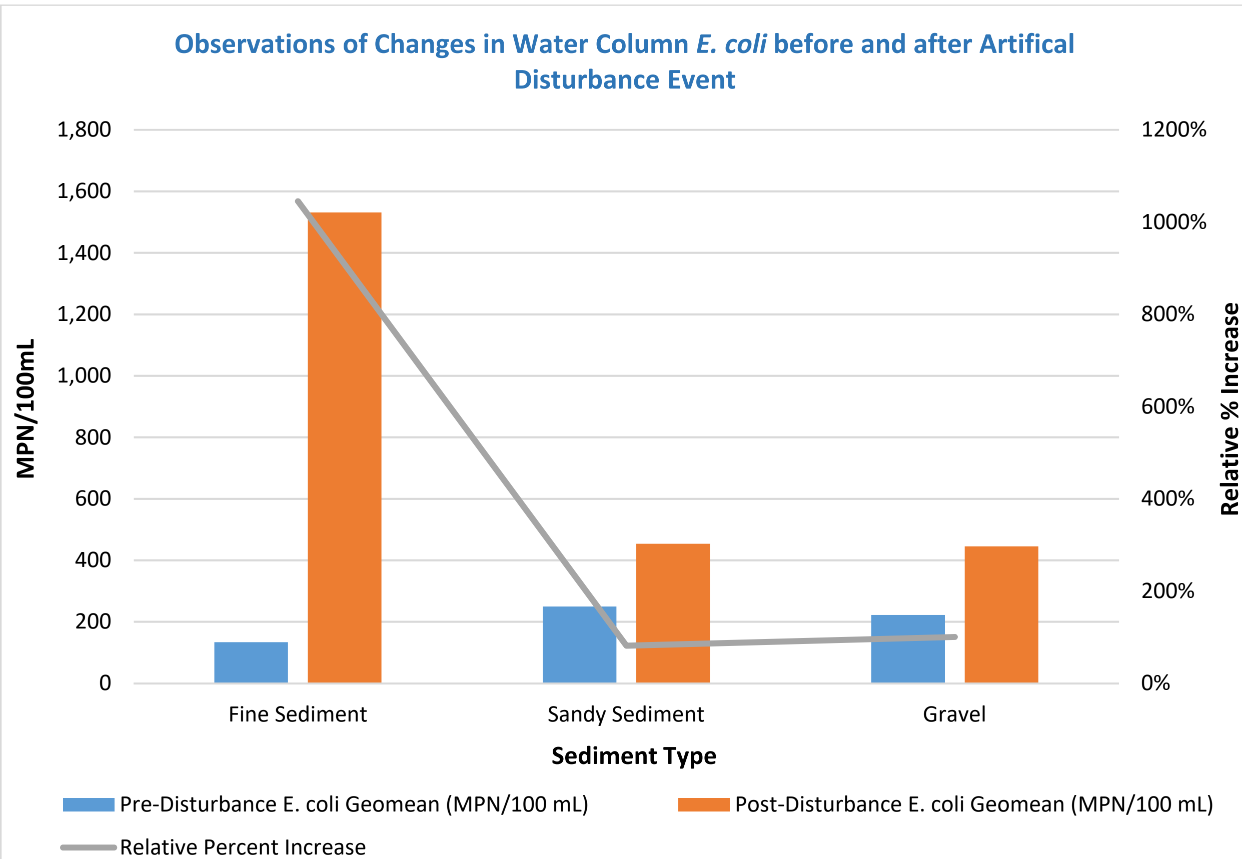

Figure 5: Observations in changes of water column E. Coli

Multiple – Variable Regression

Multiple regression analysis was used to determine if the specific sediment partitioning of each general sediment type (Fine, Sandy, Gravel) had an influence on the *E. coli* abundance in the sediments (see Table 2).

Table 2: Multiple regression analysis on sediment partitioning

Sediment Type	Fine	Sandy	Gravel
Multiple R	0.55191438	0.814591187	0.903426836
Multi R Square	0.304609482	0.663558802	0.816180047
Adjusted R Square	-0.529859139	0.259829365	0.595596104
Standard Error	266465.7404	13094.18673	14825.79639
Observations	12	12	12

The fine sediment had a multiple R-squared value of 0.30; much lower than the sandy or gravel sediments with multiple R-squared values of 0.66 and 0.82, respectively. This indicates that the array of sediment particle sizes observed in the fine sediment had the least amount of impact on the sediment *E. coli* concentrations as compared to the sandy and gravel sediments. This makes sense considering that the composition of fine sediment samples were on average 46.6% clay/silt, 48.4% fine sand, 3.3% medium/coarse sand, and 1.8% gravel – a more homogenous mixture of fine grained sediments than the sand or gravel samples. By comparison, the sandy sediments were 23.8% silt/clay, 43.8% fine sand, 23.9% medium/coarse sand, and 8.6% gravel while the gravel sediments were 4.4% silt/clay, 5.5% fine sand, 34.9% medium/coarse sand, and 55.3% gravel.

Conclusions

Core Concepts

The primary objective of this study was to determine if a particular sediment size category was a preferred habitat for *E. coli*, and how that sediment size might impact the extent to which bacteria was released into the water column during sediment disturbance events such as high flows, animal activity, and anthropogenic recreation. It was found that in the chosen sampling site, the fine sediment had both the highest post-disturbance concentration of *E. coli* in the water column and the highest sediment *E. coli* concentration by a large margin. This would suggest that the fine sediment is acting as a prime vector for *E. coli* in three capacities. 1) The increased cumulative surface area of the fines provides more area for *E. coli* to sorb onto particles. 2) That increased amount of *E. coli* bacteria is then more easily released into the water column as the fine sediment requires less disturbance to release a greater amount of sediment and thereby a greater amount of *E. coli*. 3) The density of fine sediment particles likely provides protection from predation by other bacteria and protozoa, as well as, exposure to sunlight thereby reducing ultraviolet die-off.

Additional Findings

While the fine sediments showed the greatest increase of post-disturbance *E. coli* found in the water column, both the sandy and gravel sediments also experienced increases in *E. coli* bacteria post disturbance. This should be something to consider when applying standards and assessing waterbodies for *E. coli*. While initial water samples might show low bacteria counts, if there is a high risk of disturbance events such as high flows, anthropogenic recreation, or animal activities, then the sediment types of the assessed areas could be taken in to account to explain a possible sudden and significant increase in the *E. coli* bacteria found in the water column as opposed to assuming that increases are solely due to run-off from the surrounding or upstream watershed. Additionally, sediment particle size composition may need to be taken into consideration when sampling for *E. coli* prior to an organized contact recreation event. A possible solution for addressing these issues could be to implement a modified sampling schedule during known high activity times or increased precipitation for areas of concern to get a better estimate of *E. coli* levels that could be present during these times.

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