

Instream Bacteria Influences from Bird Habitation of Bridges



Prepared by:

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July 2013

Prepared for:

Texas State Soil and Water Conservation Board

Temple, Texas

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List of Abbreviations and Acronyms

BST	Bacteria Source Tracking
cfs	Cubic Feet per Second
DSLPL	Days Since Last Precipitation
°C	Degrees Centigrade
DO	Dissolved Oxygen
FIB	Fecal Indicator Bacteria
km	Kilometer
LRW	Lampasas River Watershed
M	Non-Parametric Sign Test Statistic
m	Meter
µS/cm	Micro-Siemens per Centimeter
mg/L	Milligrams/Liter
mi	Mile
ml	Milliliter
MPN	Most Probable Number
S	Non-Parametric Signed-Rank Test Statistic
SC	Specific Conductivity
T	Water Temperature
t	Parametric t-Test Statistic
TCEQ	Texas Commission on Environmental Quality
TIAER	Texas Institute for Applied Environmental Research
TMDL	Total Maximum Daily Load
TSSWCB	Texas State Soil and Water Conservation Board
USEPA	U.S. Environmental Protection Agency
USGS	United States Geological Survey
W	Shapiro-Wilkes Test of Normality Statistic
WPP	Watershed Protection Plan

Executive Summary

Bridge crossings often afford a place of ready convenience and safe access for stream water quality sample collection. The representativeness of ambient water samples collected from bridge crossings has recently been challenged in public meetings and other forums. Critics contend that birds and bats roosting and nesting on bridge structures bias bacteria samples to more elevated concentrations than may exist in samples collected from river reaches not spanned by bridges. Water quality specialists recognize the potential legitimacy of the concern of bias from sample location but must weigh that concern against other factors that include personnel safety, cost, and ease of access. To minimize possible biases, the general practice by state agencies is to sample from the upstream side of the bridge whenever safety issues do not necessitate sampling from the downstream side.

This study was commissioned to determine what, if any, influence bridge-dwelling bird colonies have on instream bacteria concentrations collected in proximity to bridges. To this end, three bridges were selected in the Lampasas River watershed in central Texas for sampling of instream *Escherichia coli* (*E. coli*), an indicator bacterium known to exist in the intestinal tracts and feces of warm-blooded animals. Two bridges were inhabited by migratory cliff swallows and one was devoid of birds. During April – June of 2012 and January – June 2013, over 1,000 bacteria samples were collected from locations upstream, at the upstream bridgeface, and downstream of each bridge to determine whether significant increases in *E. coli* occurred in a downstream direction when birds were present and whether sampling a few meters upstream of a bridge was sufficient to avoid the influence of bridge bird colonies.

Results confirm that under dry-weather conditions, bird colonies can have a significant impact on bacteria concentrations in the vicinity of the bridges they inhabit. Not only were *E. coli* increases significant at bridgeface and downstream locations throughout the cliff swallow nesting season, they were also of high magnitude during the latter weeks of the cliff swallow nesting cycle. When bird activity peaked with fledgling emergence in *mid-* to late-May, bacteria geometric mean concentrations at bridgeface and downstream locations jumped from background levels < 50 MPN/100 ml to >190 MPN/100 ml, well above the state geometric mean criterion of 126 MPN/100 ml for primary contact recreation use.

CHAPTER 1

Introduction and Methods

Background

Surface water quality is monitored across the country by environmental agencies for fecal indicator bacteria (FIB), such as fecal coliform and *Escherichia coli* (*E. coli*). Although not necessarily pathogenic to humans, these bacterial forms indicate the possible presence of fecal pollution which is implicated in human health problems associated with water contact. Water bodies found to exceed established criteria for FIB are placed on the U.S. Environmental Protection Agency (EPA) Clean Water Act Section 303(d) list for bacteria impairment. Once a stream segment is listed, total maximum daily load (TMDL) programs and watershed protections plans (WPP) are commonly established to encourage practices that reduce instream bacteria loads. Since 1995, over 10,000 water bodies have been placed on the 303(d) list for FIB according to the U.S. Environmental Protection Agency (USEPA, 2013).

In Texas, numerous watersheds have been placed in the TMDL program by the Texas Commission on Environmental Quality (TCEQ) as a result of inclusion on the 303(d) list pertaining to elevated *E. coli*. The TCEQ encourages and provides guidance for collecting stream assessment data several meters above bridge crossings because road crossings are conveniently located and landowner permission is generally not required for access to the streambank within the bridge right-of-way (TCEQ, 2012). However, this protocol opens bacteria samples to potential bias due to the presence of birds and bats nesting and defecating into the stream at the bridge transect. State departments of transportation have studied bridge runoff impacts on downstream water quality for decades (e.g. Gupta *et al.*, 1981), and Barrett *et al.* (1995 & 1998) recorded large pulses of FIB in bridge runoff during rainfall events. It is also well documented that avian fauna are major sources of FIB in freshwater (Standridge *et al.*, 1979; Palmer, 1983; Kirschner *et al.*, 2004; Meays *et al.*, 2006a) and estuarine systems (Hussong *et al.*, 1979), sometimes overtaking non-avian vertebrate wildlife as the principal source of *E. coli* (Meays *et al.*, 2006a). Palmer (1983) attributed 17 – 35% of fecal coliform loadings in an Ontario stream location to feral rock pigeons roosting on a bridge. O’Keefe *et al.* (2005) also noted that power-washing a bridge caused dramatic spikes in fecal coliform in the adjacent stream. These studies suggest that the fecal residue from migratory bird populations that nest under bridges could impact surrounding streams as a constituent of rainfall runoff even after the birds have departed. Until recently, little research has been published quantifying the impacts of avian fecal deposits on FIB samples collected in close proximity to bridges.

In 2011, Sejkora *et al.* published a study on the influence of migratory cliff swallows (*Petrochelidon pyrrhonota*) on *E. coli* samples taken near bridges in Austin, Texas. They found significantly higher readings in downstream *E. coli* samples compared to upstream samples during dry-weather (i.e., ambient) sampling when birds were nesting. Those significantly higher readings, which hovered around the single sample criterion of 396 CFU/100 ml, persisted for 1.25 km downstream. Peak *E. coli* measurements coincided with the late-nesting and fledging period when direct deposition of fecal matter to the stream was at its highest level due to the greater amount of time spent at nests by the parent birds and the addition of fledglings at each nest. During foraging phases, when birds spent most of their active time away from the nest, the differences between upstream and downstream samples were not significant.

There was, however, a minor but significant difference between upstream and downstream samples when swallows were completely absent, possibly indicating a residual presence of swallow feces. Differences between upstream and downstream samples were also insignificant for wet weather samples when the bacteria signal was overwhelmed by the pulse of bacteria that typically enters streams via overland runoff. Sejkora *et al.* (2011) also performed one diurnal survey during the nesting period and found that time of day was not significantly correlated with *E. coli* values, a conclusion at odds with other recent studies of diurnal variability in lotic freshwater systems that attributed strong daytime decay rates primarily to sunlight radiation (Traister and Anisfeld, 2006; Desai and Rifai, 2013)

Sejkora *et al.* (2011) claimed to be the first to examine FIB contributions to streams from bridge-dwelling cliff swallows and the literature search described herein confirms their claim. In contrast to Sejkora *et al.* (2011), the present study focused exclusively on the impacts of swallow bridge communities on instream *E. coli* under dry-weather conditions. Sampling resources were allocated to tackle the central research question: Do nesting birds at bridges increase *E. coli* sample concentrations taken at road crossings? The results potentially have far-reaching implications for state and national stream assessment protocols since sampling water quality at road crossings is commonplace. Based on the results of Sejkora *et al.* (2011) and earlier studies described above, we hypothesized that under ambient conditions at bridges occupied by cliff swallows, *E. coli* values would increase significantly between upstream, bridgeface, and downstream samples. Furthermore, we hypothesized that higher densities of active nests would be associated with greater differences between upstream and downstream samples since Kirschner *et al.* (2004) recorded significant correlations between waterfowl fecal pellet counts and FIB in the water column.

Cliff swallows create some of the most dense avian bridge colonies in the United States, reaching 3,500 individuals in a single colony (Brown and Brown, 1995). Originally isolated to the western United States, the steady encroachment of cliff swallows eastward for the last 100 – 150 years has been attributed to the development of the U.S. highway system, which the birds have coopted for nesting habitat (Gorenzel and Salmon, 1982; Brown and Brown, 1995). The birds typically arrive in Texas from the southwest in early March and migrate to north-central Texas by April 1 (eBird, 2013; Figure 1-1). Average reported counts are defined by eBird as “the average number of birds seen on checklists with a positive observation for the species within a specified date range and region”. The values can be interpreted as the flock size commonly reported at sites where the species is observed.

Once settled at a nesting site, the swallows are highly active for several weeks of feeding and building or repairing their nests. During this time, cliff swallows are known to spend up to 9.5 hours a day foraging compared to 11.5 hours in the nest, plus 3 hours of nest building (Withers, 1977). During incubation, the foraging hours drop 30% as several more hours a day are spent in the nest. Although total time away from the nest decreases during incubation, feeding sorties are more frequent and of shorter distance. Since defecation commonly occurs during take-off from the nest (Brown and Brown, 1995), increased sorties presumably translate to increases in fecal loading within a few meters below the nest. During nestling (mid-May – early June), activity at the nest peaks as parents forage not only for themselves, but also for the fledglings, and perch at the nest rim to feed their young. As young birds spend more time roosting on the edge of their nests, both parents and fledglings contribute fecal material to the land or water beneath the nest through direct deposition and removal of fledgling fecal sacs from the nest by adults (Gorenzel and Salmon, 1982).

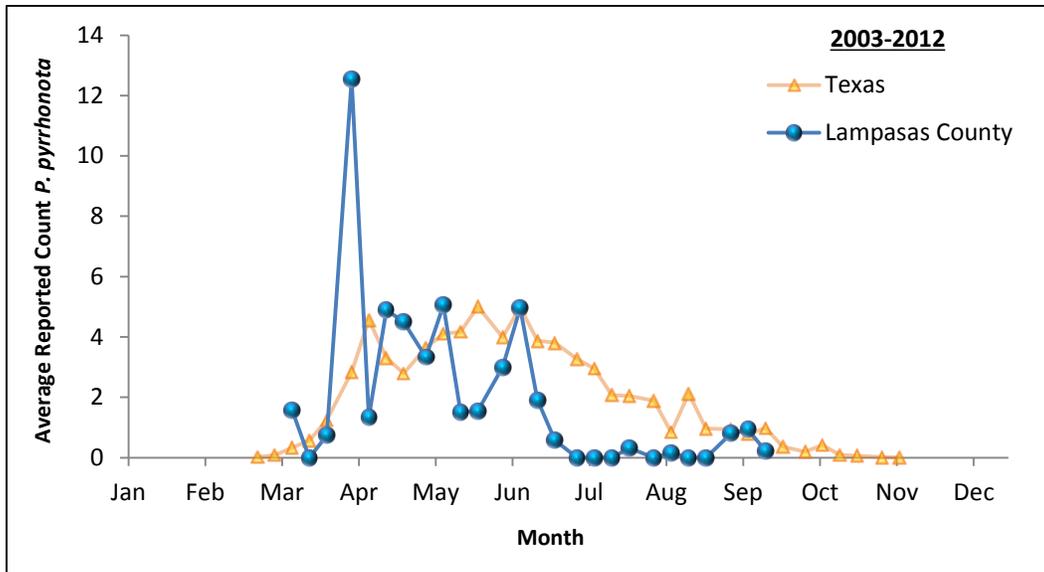


Figure 1-1. Average reported counts of *P. pyrrhonota* by month for the State of Texas (orange) and Lampasas County (blue). Data from www.eBird.com, accessed 19 March 2013

The purpose of this study was to quantify the impacts of cliff swallow colonies on instream *E. coli* at bridges on the Lampasas River and to supply data to address concerns regarding bacteria sample collection in close proximity to bridges colonized by birds. A full literature review is provided in Appendix A to aid contextualization of this report among published studies.

Site Descriptions

Three bridges were selected for this study based on the density of swallow nests, limited canopy cover (to avoid the influence of tree-roosting birds), accessibility, landowner cooperation, and hydrological reliability, among other factors. All stations were located on the Lampasas River in central Texas. During the first year of sampling, two bridges were considered as treatment locations due to the presence of nesting cliff swallows (Stations 16404 and 21186), and one bridge with an absence of birds and nests was considered as a control (Station 20018; Figure 1-2). During the second year of sampling, the birded Station 16404 was used as a temporal control with monitoring prior to the arrival of cliff swallows.

Treatment Station 16404 was located on the Lampasas River at the Farm-To-Market Road 2313 crossing approximately 3.5 mi. upstream of the confluence with Sulphur Creek (Figure 1-2). The approximate stream width in the sampling reach ranged between 4 – 9 m during 2012 and only 3 – 6 m during 2013 when streamflow was much reduced. Sparsely vegetated cobble beaches 10 – 30 m wide extended the full length of the sampling reach on the north shore and the upstream south shore. A steep shrubby bank lined the downstream south shore. The stream was characterized by riffle-run sequences with cobble-gravel substrate, occasional snag material, and a depth range of about 0.3 – 0.8 m. The water surface was entirely exposed to sunlight except for bridge shade.

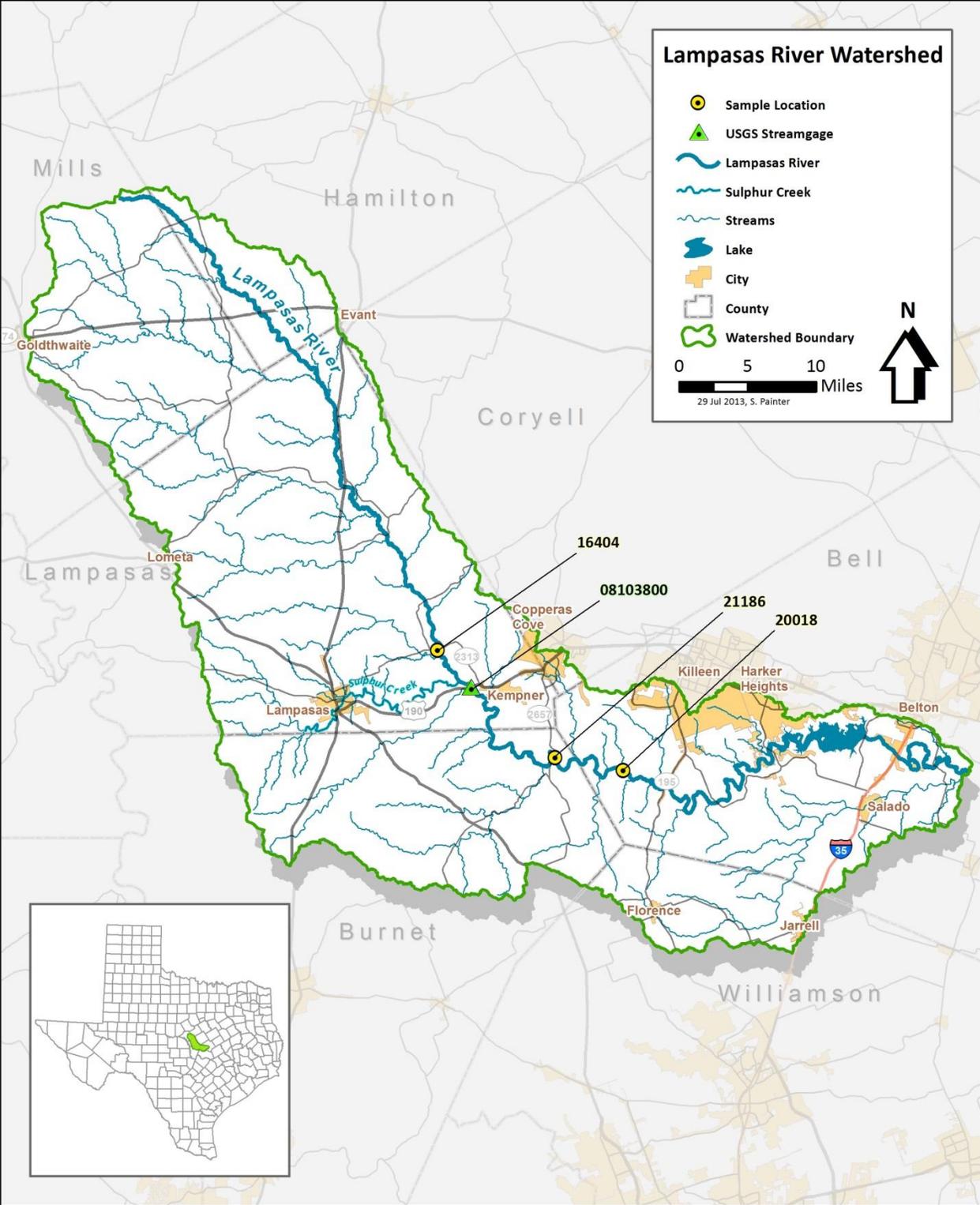


Figure 1-2. Map of the Lampasas River watershed and sampling bridge locations

Treatment Station 21186 was located on the Lampasas River at the crossing of Farm-To-Market Road 2657 about 6 mi. south of the City of Copperas Cove (Figure 1–2). The approximate stream width in the sampling reach ranged between 10 – 27 m during 2012 but ranged several meters narrower in 2013 due to low streamflow. A wide cobble beach (15 – 30 m), mostly unvegetated, extended the full length of the sampling reach on the northeast shore whereas the southwest shore was primarily a steep bank of trees and shrubs except for directly under the bridge where the bridge skirting came down nearly to the water’s edge. The upstream reach was characterized by laminar flow over bedrock, silt, and sand substrate with mostly uniform depth (~0.6 m). Under the bridge and for 30 m downstream the depth increased up to 2 m with occasional boulders near the southwest shore. Fishermen frequented the station but only once entered the water above a sampling location, generally preferring to sit on the bridge skirting on the southwest shore under the shade of the bridge. Except for the bridge shadow, the water surface was entirely exposed to sunlight.

The spatial control Station, 20018, was located at a retired bridge on old Maxdale Road in Maxdale, Texas (Figure 1-2). This truss bridge possessed no bird nests. It was, however, positioned about 300 m downstream from a newly constructed concrete bridge under which a small colony of cliff swallows was observed. The south shore was a flat cobble beach about 10 – 40 m wide behind which were steep, treed banks. The north shore was a very steep treed bank. Forty meters upstream of the bridge, at the sampling location, was a shallow cobble riffle that drained into a deep run or pool about 15 m wide and up to 2 m deep. This pool extended from 20 m upstream and downstream of the bridge before emptying into another shallow riffle. A small intermittent creek drained into the pool from the north side about 20 m upstream of the bridge.

Watershed Description

Small, mostly intermittent streams in Mills, Hamilton, and Lampasas counties feed the upper half of the Lampasas River. Sulphur Creek, a large, spring-fed stream that joins the Lampasas River about eight miles east of the City of Lampasas, contributed an average of 85 – 95% of the flow to the Lampasas River below the confluence on study dates between 2012 – 2013 based on the differences between instantaneous flow measurements at Station 16404 and USGS gage 08103800 at US 190 about 1 mile southeast of Station 16404. (USGS, 2013; Figure 1-2). The Lampasas River watershed (LRW) is dominated by scrub/grassland and forest with small percentages of land set aside for crops and hay production (Figure 1-3 and Table 1-1; USGS, 2011). The sub-watershed above Station 16404 is nearly devoid of developed land whereas Stations 21186 and 20018 receive flow emanating from the City of Lampasas (2012 est. pop. 6,854) and western portions of Copperas Cove (2012 est. pop. 33,374; U.S. Census Bureau, 2013). Since 2011, the LRW, along with most of Texas, has been in drought conditions ranging from severe to exceptional (U.S. Drought Monitor, 2013). Flow was always present during sampling periods, but was occasionally sluggish, particularly at Station 16404, which was situated above the sustaining discharge of Sulphur Creek.

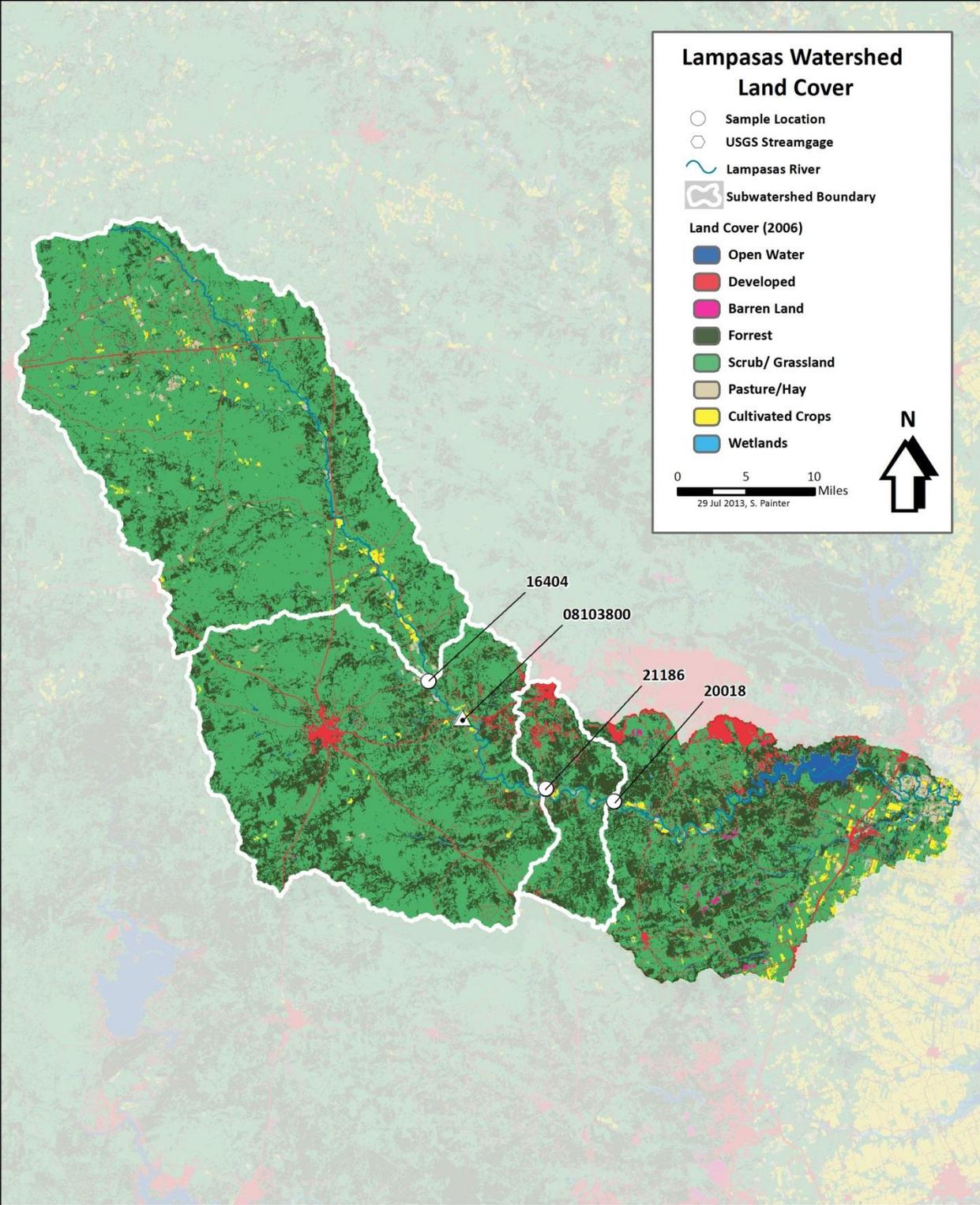


Figure 1-3. Land use and land cover in the Lampasas River watershed and the sub-watersheds of sampling Stations 16404, 21186, and 20018

Table 1-1. Land use and land cover in the sub-basins of Stations 16404, 21186, and 20018 in the Lampasas River watershed

Land Cover Type	Station						Total Acres*
	16404		21186		20018		
	Acres	%	Acres	%	Acres	%	
Scrub/ Grassland	316282	81.2%	203152	69.7%	28086	49.7%	547520
Forrest	53203	13.7%	72764	25.0%	23060	40.8%	149027
Developed	9656	2.5%	12140	4.2%	4449	7.9%	26245
Cultivated Crops	4902	1.3%	889	0.3%	176	0.3%	5967
Pasture/ Hay	3863	1.0%	746	0.3%	167	0.3%	4776
Wetlands	1102	0.3%	1081	0.4%	357	0.6%	2540
Open Water	604	0.2%	487	0.2%	107	0.2%	1198
Barren Land	44	0.0%	176	0.1%	110	0.2%	330
Grand Total	389656		291435		56512		737603

* Total acreage above the most-downstream bridge, Station 20018

The upper Lampasas River watershed (assessment units 1217_04 and 1217_05) was first placed on the Texas 303(d) list for bacteria impairment in 2002 (TCEQ, 2002). It was delisted following publication of the 2008 303(d) list (TCEQ, 2008). Recently, the Texas Water Resources Institute produced a report on bacteria source tracking (BST) for the LRW showing that among bacteria samples collected between February 2011 and January 2012, 14% of the *E. coli* isolates were traceable to avian wildlife (Gregory *et al.*, 2013). Only one of the 15 sampling sites in the TWRI study overlapped with this study (Station 16404; Figure 1-2).

Methods

Field Methods

Three bridges were chosen in the LRW. Stations 16404 and 21186 were treatment bridges occupied by migratory cliff swallows during the spring, and Station 20018 was a control bridge that possessed no bird nests during the spring. The treatment bridges were sampled during the cliff swallow nesting season except for Station 16404 which was also sampled as a temporal control site in January – February, 2013, when cliff swallows were absent. At each bridge crossing three sampling locations were established—at the upstream side of the bridge (“bridge”), 45 m upstream of the bridge (“upstream”), and 45 m downstream of the bridge (“downstream”) in that order (Figure 1-4). During each survey, each of the locations in the vicinity of the bridge crossing was visited three times (three repetitions or “reps”) and 5 samples at each location were collected one minute apart during each rep for *E. coli* analysis. Thus, a total of 45 *E. coli* samples were collected from each station during each survey. Reps were separated by 30-min intervals, beginning at the time of the 5th upstream sample in each rep. This time lapse ensured that any disturbances of the water and sediment during one rep did not carry into subsequent reps. A total of 23 survey events occurred over the 2-year monitoring period. During the first period of monitoring (spring and summer 2012) each of the three bridge stations were sampled four times. During the second year of monitoring (winter 2013 – summer 2013) the control changed from a spatial control (Station 20018) to a temporal control by sampling treatment Station 16404 prior to the arrival of birds in late March to early April. Thus, Station 16404 was sampled seven times during 2013; three times before arrival of birds and four times when birds were present. The other treatment bridge, Station 21186, was sampled four times in 2013 when birds were present. Cumulatively, this sampling regime produced 23 total surveys (12 in 2012 and 11 in 2013) and 1035 *E. coli* samples. All surveys were conducted under low-flow conditions not influenced by stormwater runoff. Low-flow conditions were selected as a criterion for survey conditions because bacteria gathered by runoff from the surrounding landscape can dramatically raise instream *E. coli* during and after rainfall runoff events (Geldreich *et al.*, 1968; Collins *et al.*, 2005; Chu *et al.*, 2011).

During the study, USGS gage 08103800 near Kempner, Texas, was used to monitor flow levels following storm events and to determine days since last significant precipitation. The gage is located on the US 190 crossing of the Lampasas River near Kempner, about six miles east of the City of Lampasas.

Throughout the study, all contact with the water was avoided, except for the sample collection apparatuses when obtaining the water samples for *E. coli* analysis, until all 45 bacteria samples for a survey were collected. Multiple studies have demonstrated that disturbed sediment is a major source of bacteria to the water column (Cho *et al.*, 2010; Pachepsky and Shelton, 2011). To this end, large rocks and cinder blocks were deployed as stepping stones at some sampling locations prior to sampling events to enable sample collection in flowing water without entering the stream. There were only a few minor instances of field personnel slipping from a stepping stone and planting their foot in the sediment and these events were noted, however, no impacts on data were identified. Extension poles with bottle holders were also utilized to ensure collections were made in flowing water without entering the stream. At Station 20018, the large pool beneath the bridge was too wide to allow sample collection in flowing water without entering the stream so a bottle holder was lowered by rope from the bridge and dipped in the middle of the pool.

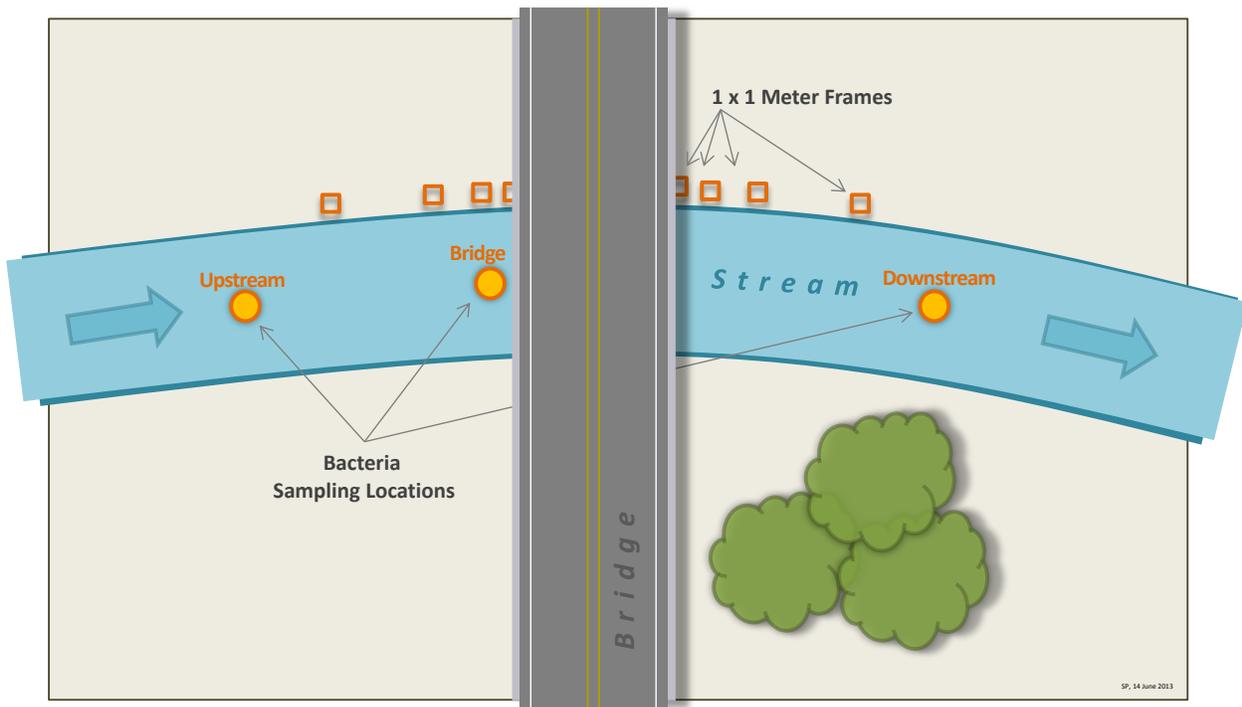


Figure 1-4. Sampling scheme for collection of instream *E. coli* and estimation of bird density using fecal collection frames

In 2013, filamentous algae mats grew increasingly thick around the upstream sampling location at Station 16404. On a day prior to spring sampling, a square-foot patch of algae was removed by hand from the sampling point to ensure the bottle could be dipped without algal interference.

Whether by hand, by pole, or by rope, all sample bottles were dipped fully below the surface and all contact with sediment was avoided. These extensive measures were taken to ensure that only bacterium from the water column was collected.

A streamflow measurement was taken at each bridge during each survey using a SonTek Flow Tracker™ Acoustic Doppler Velocimeter. Physicochemical parameters were measured in the vicinity of each bridge crossing during each survey using a YSI 600 XLM multiprobe for dissolved oxygen (DO), water temperature (T), specific conductivity (SC), and pH. The streamflow and physicochemical measurements were not made until after the collection of all 45 bacteria samples. Days since last precipitation (DSLPP) was determined from the rain gage at USGS 08103800, located near the middle of the three bridge stations on the Lampasas River.

In addition to bacteria samples, multiprobe, and flow measurements, eight frames were deployed for measuring direct fecal matter deposition at various distances from the bridge (Figure 1-4). Fecal deposition was directly quantified by counting bird droppings. To diminish subjectivity in the counting method, the lead field supervisor was the sole counter and the same criteria was applied every time: all separate piles of fecal matter were counted as an individual dropping and counted only as one dropping,

i.e., no effort was made to discern whether a pile was comprised of multiple droppings in the same spot on the frame because of the subjective nature of making such judgments. The plywood frames were 1 m² and four were deployed both upstream of the bridge and downstream of the bridge at 0 m (directly under the bridge faces), 4 m from the bridge face, 10 m, and 30 m. The frames were placed as near the stream as bank slope and other conditions allowed, and at each station all frames were deployed on the same side of the stream. The frames at the bridge faces were deployed immediately below active nests at the treatment bridges and directly under the outer edge of the bridge for the control station. Frames were deployed either the day before the bacteria sample collection or the day of bacteria sampling and retrieved the following day with a total elapsed time of deployment between 21.2 and 25.2 hours.

Bird numbers were estimated from counts of active nests on seven dates at Station 16404 and six dates at Station 21186. The field supervisor performed the nest count on every occasion for consistency. Nests were considered active if a) birds were observed in the nest, b) fresh mud was observed on the nest indicative of active nest maintenance, or c) fresh accumulations of bird feces were observed directly beneath the nests on the bridge beams. Nests built on the bridge beams that spanned the stream were all counted as “over water” though only a percentage of the nests were actually located directly over the stream. For example, the width of the stream at Station 16404 was generally 3 – 5 meters underneath the bridge but the beams above the wetted channel were approximately 20 m long. The results of these counts, though imprecise, produced estimates of relative bird colony size and density between the two treatment bridges.

Statistical Methods

Determining the appropriate statistical tests to apply depends on the study design and the underlying distribution of the data. Parametric tests are considered more powerful than non-parametric tests for detecting significant differences between independent sample groups that are normally distributed. Helsel and Hirsch (2002) said that where “natural structure in the order of observations across groups” of samples was lacking, the groups could be considered independent. A “natural structure” was, however, present among the 15 samples collected at each location on each date in the present study because they were collected from the same reach within 60 minutes of each other, on average. Since these sample groups had a moderately low n (15) and were rarely normally distributed according to Shapiro-Wilkes tests of normality (W), non-parametric tests were determined more powerful tests of location differences than the parametric paired t-test.

Another factor to consider was the influence of time of day on reps collected at a location. The literature is mixed on whether time of day is a factor in *E. coli* concentrations. Sejkora *et al.* (2011) found no significant differences between samples collected in 6-hr intervals during the nesting season. However, Meays *et al.* (2006b) reported considerable diurnal variation among samples collected every 15 minutes over a 24-hr period in three British Columbian streams. The rep sample groups for this study were considered dependent because they were collected from the same location usually within an hour of each other. The sample groups for each rep had an n of five rendering these datasets too small to adequately assess their distribution. To ascertain whether *E. coli* concentrations changed significantly over time between reps, differences between reps at a location were evaluated using non-parametric statistics (signed-rank and sign tests), but also included the parametric t-test for comparison.

In consideration of the lack of independence and normality among location and rep sample groups, non-parametric tests were run to compare location (upstream, bridge, and downstream) and rep (1, 2, and 3) for each bridge on each date to test the null hypothesis of equal medians among the three locations and among the three reps. In all cases, p values < 0.05 were considered significant and values < 0.001 were considered highly significant. Samples were paired in the order they were collected so that in the test of location the first sample of the upstream location was paired with the first sample of the downstream location, and so on. For the test of reps, the first sample of the first rep at a location was paired with the first sample of the second rep at a location, and so forth.

The univariate procedure in SAS was used to produce the tests of normality, skewness, and kurtosis, as well as the parametric and non-parametric tests of differences between locations and reps (SAS 9.3, SAS Institute, 2002-2010). *E. coli* values were log₁₀-transformed prior to running tests of differences in location and rep. This transformation resulted in more normal sample distributions, yet some distributions were still heavy-tailed or contained outliers. Therefore, two non-parametric tests were relied upon in this paper to detect differences in sample groups: the signed-rank test (the S-statistic) and the sign test (the M-statistic). Both test for differences in medians but the sign test is more powerful for detecting differences between skewed sample groups or those with outliers. Results of the parametric paired t-test of population means are included in this report despite parametric assumptions being met in only a few of the location test groups. The *t* statistic is included because in some cases of agreement between parametric and non-parametric tests, the t-test provides additional support for interpretation of the results. Transforming the *E. coli* dataset was done primarily to accommodate the t-test which requires normality.

For each station, a table of test results is presented by survey for location comparisons where BU compares the 15 samples from the bridge and upstream locations; DU compares downstream and upstream samples, and DB compares downstream and bridge samples. The mean difference is the average of the 15 differences between samples at the two locations in each comparison. Thus, a positive mean difference indicates a mean increase in *E. coli* in a downstream direction and a negative value indicates a mean decrease in a downstream direction. These tables are followed by plots of *E. coli* samples organized by survey and location. A reference line representing the geometric mean (geomean) criterion of 126 CFU/100 ml is included for determination of whether fecal inputs from bird colonies were sufficient to push sample geomeans over the criterion. The geomean line is not provided to determine whether the stream segment requires placement on the 303(d) list but is provided to aid interpretation of the results in the context of the statewide bacteria geomean criterion. Tables of geomeans are then presented in a location-rep matrix. Following these tables and sample plots, the results of the tests of sample reps are provided. The rep comparisons cover 1 and 2 (R12), 1 and 3 (R13), and 2 and 3 (R23) and the test results are displayed the same format as for location differences. Tables follow that display fecal frame counts by station and survey, normalized to 24-hr fecal deposition rates. A table of flow and multiprobe data complete the results section.

CHAPTER 2

Results and Discussion

Estimation of Active Bird Nests

Cliff swallow activity at the treatment bridges, Stations 16404 and 21186, followed closely the phases described in published literature. The birds arrived at the bridges in early April 2012 and in late March 2013. Each year the nesting phases progressed the same. After a couple of weeks of foraging away from the bridge and only occasional nest mending, the nest repairs began in earnest. From mid-April to early May eggs were laid and incubated during which time at least one bird was almost always present in the nest while the partner foraged. By mid-May, nestlings were observed at the rims of the nests, on the bridge beams and, occasionally, on the ground. Total active nest counts during the swallow season were higher at Station 21186 than at Station 16404 (Figure 2-1); however, the span of the bridge was several times that of Station 16404, extending approximately 130 m to the northeast beyond the streambank over grassy fields. It was under this extension of the bridge, rather than over the river, that the highest concentration of nests and bird activity was recorded on most dates. Station 16404 actually had the higher density of nests over water between the two treatment bridges owing to the fact that it was only $\frac{1}{4}$ as wide but often had as many birds active in nests and in flight within 10 m of the stream banks as Station 21186. One count was made at Station 16404 during the temporal control collection on 23 January 2013. Only six active nests were counted and these were all over the water. They were occupied by house sparrows which maintained a population at the bridge of about 1 – 2 dozen throughout January and February 2013 while temporal control samples were gathered. No other species were spotted nesting at the bridge during visits in these winter months.

Results of *E. coli* Sampling

Tests of Location

E. coli at the heavily-birded Station 16404 increased significantly ($p < 0.05$) between upstream and downstream sampling locations during all eight treatment surveys (Table 2-1 and Figure 2-2) according to the parametric t-test (t), sign test (M) and signed-rank test (S). The exception was 22 May 2012 when half of the samples were discarded because of lab error. There was strong agreement among parametric and non-parametric tests that the *E. coli* increases were highly significant ($p < 0.001$) in a downstream direction between all three sampling locations on most dates. During late April – May in 2012 and 2013 geomeans at the downstream location exceeded 150 MPN/100 ml, well above the criteria of 126 CFU/100 ml (Table 2-2). Geomeans at the bridge location also exceeded 150 MPN/100 ml during the late bird season in 2013. Variability was higher at bridge and downstream locations than at the upstream location on all dates—often much higher (Figure 2-2 and Appendix B-1).

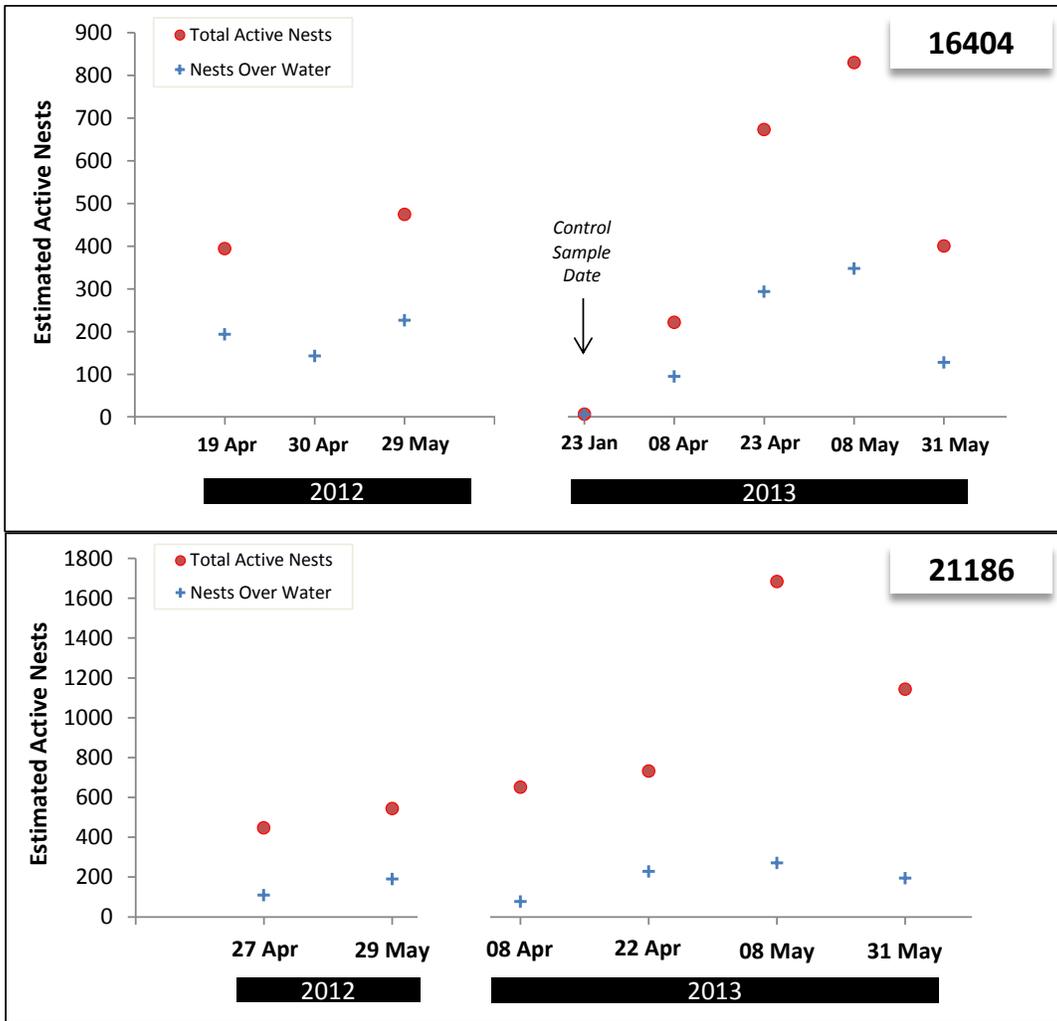


Figure 2-1. Estimated counts of active nests at treatment Stations 16404 and 21186, 2012 – 2013. “Nests over water” refers to all nests on beams that spanned the wetted channel, not only the nests directly above the water. Total count was not recorded at Station 16404 on 30 April 2013.

Table 2-1. Differences between sampling locations at the densely-birded bridge, Station 16404, 2012 – 2013. Significant differences ($p < 0.05$) are highlighted; strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for bridge – upstream (BU), downstream – upstream (DU), and downstream – bridge (DB). Values were \log_{10} -transformed prior to testing.

Location Comparison	BU	DU	DB	BU	DU	DB	BU	DU	DB	BU	DU	DB
Survey	19-Apr-12			30-Apr-12			22-May-12			23-May-12		
<i>n</i>	15	15	15	15	15	15	7	5	5	15	15	15
Mean Difference (MPN/100 ml)	11.5	15.1	3.6	39.9	70.1	30.2	-3.7	135.0	138.8	19.1	220.5	201.4
$Pr \geq t $	<i><0.001</i>	<i><0.001</i>	0.181	0.002	<i><0.001</i>	0.014	0.278	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>
$Pr \geq M $	<i><0.001</i>	<i><0.001</i>	0.424	<i><0.001</i>	<i><0.001</i>	0.007	0.453	0.063	0.063	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>
$Pr \geq S $	<i><0.001</i>	<i><0.001</i>	0.178	<i><0.001</i>	<i><0.001</i>	0.015	0.375	0.063	0.063	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>
Survey	8-Apr-13			23-Apr-13			6-May-13			30-May-13		
<i>n</i>	15	15	15	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	66.9	121.3	54.4	165.7	273.4	107.7	169.5	166.9	-2.7	144.8	208.8	63.9
$Pr \geq t $	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	0.001	<i><0.001</i>	<i><0.001</i>	0.832	<i><0.001</i>	<i><0.001</i>	0.002
$Pr \geq M $	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	0.007	<i><0.001</i>	<i><0.001</i>	0.607	<i><0.001</i>	<i><0.001</i>	0.007
$Pr \geq S $	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>	0.003	<i><0.001</i>	<i><0.001</i>	0.858	<i><0.001</i>	<i><0.001</i>	0.001

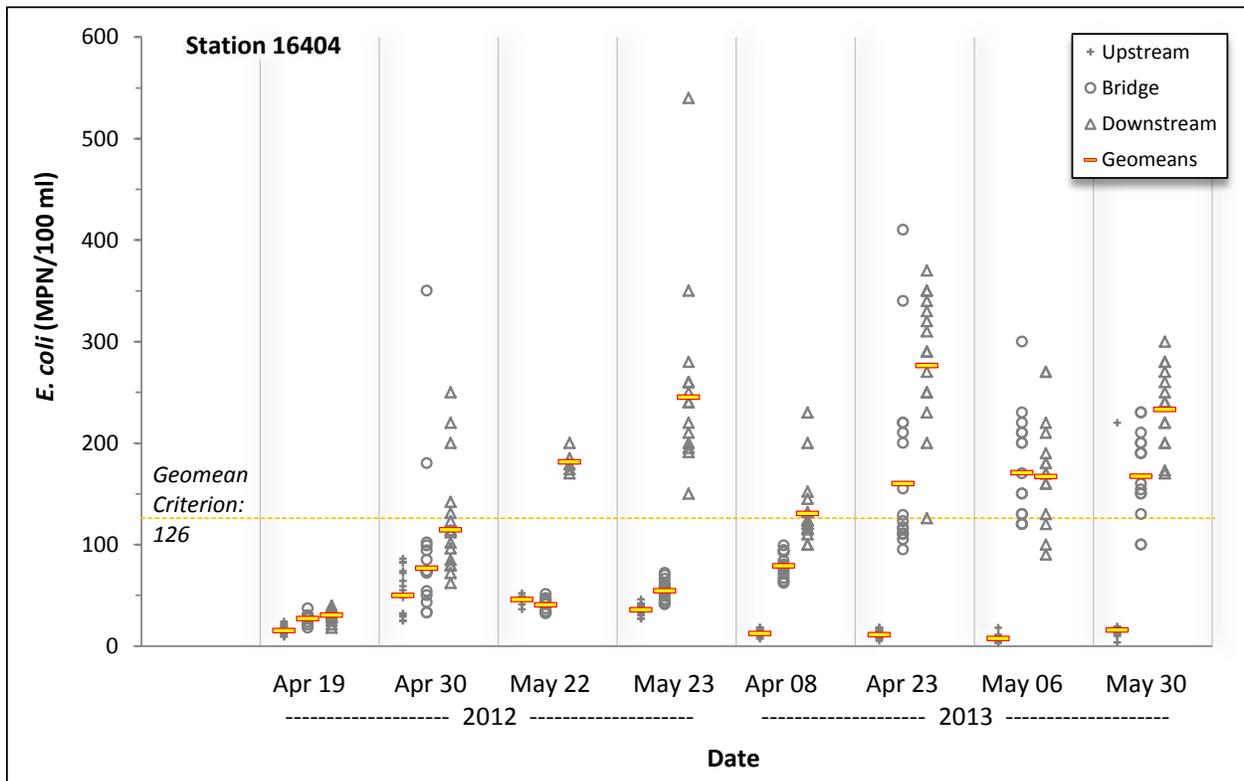


Figure 2-2. Treatment *E. coli* at the densely-birded bridge, Station 16404, 2012 – 2013. State geomean criterion line provided for reference; the *n* for 22 May 2012 was 10 (bridge), 7 (upstream), and 5 (downstream).

Table 2-2. Geomeans of *E. coli* samples from upstream, bridge, and downstream locations at the densely-birded treatment Station 16404, 2012 – 2013. ND = no data.

Station 16404					
Survey	Rep	UPSTREAM	BRIDGE	DOWNSTREAM	Overall
19-Apr-12	1	16.2	30.4	27.6	23.9
	2	15.8	29.0	30.6	24.1
	3	13.3	22.2	32.8	21.3
	Overall	15.1	26.9	30.3	23.1
30-Apr-12	1	79.2	107.7	180.6	115.5
	2	55.1	99.9	110.4	84.7
	3	28.2	41.7	75.2	44.6
	Overall	49.8	76.6	114.4	75.8
22-May-12	1	46.2	42.3	181.3	70.8
	2	43.9	38.4	ND	39.9
	3	ND	ND	ND	ND
	Overall	45.6	40.3	181.3	59.0
23-May-12	1	38.6	63.8	298.8	90.3
	2	31.7	46.0	232.6	69.7
	3	36.4	54.2	211.8	74.8
	Overall	35.5	54.2	245.1	77.8
8-Apr-13	1	13.7	87.1	131.3	53.9
	2	12.9	81.3	112.7	49.1
	3	10.4	69.5	149.7	47.7
	Overall	12.3	78.9	130.4	50.2
23-Apr-13	1	12.4	140.0	332.9	83.3
	2	11.7	121.8	287.1	74.2
	3	8.9	239.8	220.5	77.7
	Overall	10.9	159.9	276.2	78.3
6-May-13	1	7.3	142.6	199.0	59.1
	2	9.1	215.9	196.0	72.7
	3	5.6	161.0	119.0	47.4
	Overall	7.2	170.5	166.8	58.8
30-May-13	1	26.4	194.1	215.7	103.4
	2	13.7	190.2	249.0	86.6
	3	10.4	126.2	235.6	67.6
	Overall	15.6	167.0	233.1	84.6

At the moderately birded bridge, Station 21186, the differences between upstream, bridge, and downstream samples were often significant (Table 2-3 and Figure 2-3), but less consistent and pronounced than the heavily-birded Station 16404 (Table 2-1). In both years, location differences were not strongly significant until later in the bird season. Geomeans exceeded the 126 CFU/100 ml criteria on only two dates: 29 May 2012 (431 and 130 at the bridge and downstream locations, respectively) and 31 May 2013 (190 at the downstream location; Table 2-4). Several minutes prior to sampling at Station 21186 on 29 May 2012, two fishermen waded across the stream about 4-m upstream of the bridge sampling location, stirring sediment as they went. All bridge values that day were higher than the highest value recorded on 24 May 2013 and far exceeded outliers on all other dates at Station 21186 (Figure 2-3 and Appendix B-2). Furthermore, the upstream geomean on 29 May 2012 was only slightly greater than the upstream geomean of the previous survey, suggesting no change in background *E. coli* levels entering the sampling reach. The outliers on 29 May 2013 are thus likely the result of sediment stores of bacteria re-entering the water column and persisting in the sluggish flow for the duration of that day's survey. Downstream increases in *E. coli*, although significant on 22 April 2013 and 07 May 2013, were not nearly as high in magnitude as the increases seen on 31 May 2013. Bird activity above the water was notably higher during this final 2013 survey as parent swallows were busy feeding mature fledglings, making frequent, brief visits to their nests. Variability at bridge and downstream locations was generally higher than at the upstream location (Figure 2-3 and Appendix B-2) but the pattern is not as overt as at the heavily birded Station 16404 (Figure 2-2 and Appendix B-1). Variability at the bridge and downstream locations at Station 21186 increased during the later phases of the bird season in each year, commensurate with geomean increases at these locations.

At the spatial control, Station 20018, bridge and downstream geomeans were sometimes higher, sometimes lower, than the upstream location (Tables 2-4 – 2-6 and Figure 2-4). With only one exception, the geomeans of the upstream location at Station 20018 were all higher than the geomeans at the upstream locations of the other two study bridges (Tables 2-6; see Tables 2-4 and 2-6 for comparisons). Higher background *E. coli* concentrations entering the sampling reach might have resulted from a cliff swallow colony at a bridge approximately 300-m upstream of Station 20018. Much lower values during the last survey when swallows were absent from the upstream bridge corroborates the suspicion that bacteria was persisting in the water column to the sampling station. Large common carp (*Cyprinus carpio*; > 0.5 m) and turtles were seen foraging in the sediment at the start of the 30 May 2012 survey at Station 20018. Multiple plumes of sediment spread several meters through the water in the wide, sluggish run underneath the bridge. This foraging activity likely is the cause of the unusually high values at that location that had an average of 1,134 CFU/100 ml (Table 2-6).

The temporal control Station 16404, which was colonized by 1-2 dozen house sparrows during the winter samples, showed low but significant increases in *E. coli* concentrations from upstream to downstream locations on all three dates (Tables 2-5 – 2-6 and Figure 2-4). Differences between adjacent locations (BU and DB) were not as stark or consistently significant. Comparisons of upstream geomeans between control and treatment surveys at Station 16404 reveal background bacteria concentrations were slightly lower during the control samples; ranging 7 – 19 CFU/100 ml versus 7 – 50 during treatment visits (see Tables 2-2 and 2-6 for comparisons). Lower background bacteria levels may have enhanced the signal coming from roosting sparrows.

Table 2-3. Differences between sampling locations at the moderately-birded bridge, Station 21186, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for bridge – upstream (BU), downstream – upstream (DU), and downstream – bridge (DB). Values were \log_{10} -transformed prior to testing.

Location Comparison	BU	DU	DB	BU	DU	DB	BU	DU	DB	BU	DU	DB
Survey	26-Apr-12			1-May-12			24-May-12			29-May-12		
<i>n</i>	15	15	15	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	-1.2	1.6	2.8	4.1	9.3	5.2	77.3	53.5	-23.8	451.1	100.4	-351.0
$Pr \geq t $	0.985	0.529	0.294	0.308	0.007	0.133	<i><0.001</i>	<i><0.001</i>	0.067	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>
$Pr \geq M $	0.607	1.000	0.791	0.581	0.118	0.035	<i><0.001</i>	<i><0.001</i>	0.118	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>
$Pr \geq S $	0.296	0.893	0.296	0.340	0.010	0.048	<i><0.001</i>	<i><0.001</i>	0.064	<i><0.001</i>	<i><0.001</i>	<i><0.001</i>
Survey	9-Apr-13			22-Apr-13			7-May-13			31-May-13		
<i>n</i>	15	15	15	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	-5.0	-3.2	1.7	0.8	5.5	4.7	2.0	6.6	4.5	13.5	172.9	159.4
$Pr \geq t $	0.001	0.036	0.090	0.158	<i><0.001</i>	0.002	0.303	<i><0.001</i>	0.002	0.006	<i><0.001</i>	<i><0.001</i>
$Pr \geq M $	0.013	0.302	0.791	0.607	<i><0.001</i>	0.007	0.180	<i><0.001</i>	0.007	0.035	<i><0.001</i>	<i><0.001</i>
$Pr \geq S $	0.002	0.066	0.173	0.169	<i><0.001</i>	0.001	0.267	<i><0.001</i>	0.004	0.010	<i><0.001</i>	<i><0.001</i>

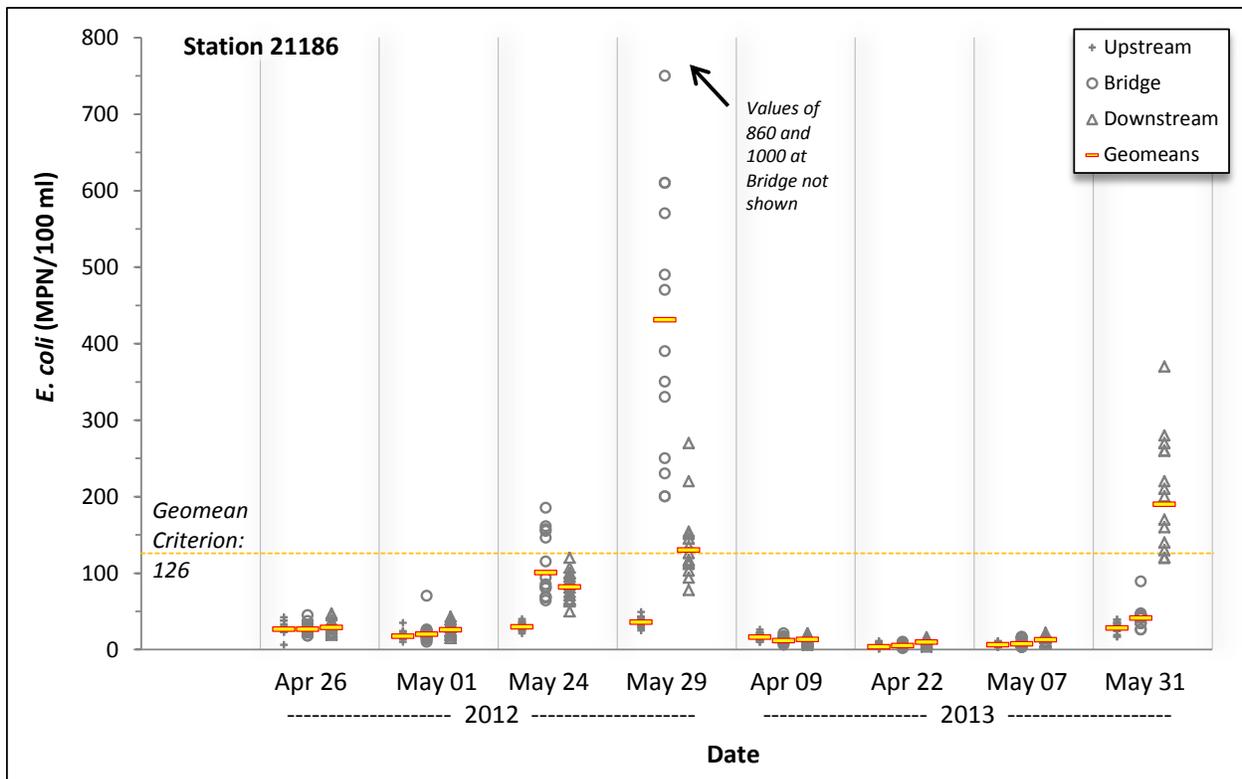


Figure 2-3. Treatment *E. coli* at the moderately-birded bridge, Station 21186, 2012 – 2013. State geomean criterion line provided for reference. Outliers on 29 May 2012 likely attributable to fishermen wading across stream 4 meters above the bridge sampling location at the start of the first sampling rep.

Table 2-4. Geomeans of *E. coli* samples from upstream, bridge, and downstream locations at the moderately-birded treatment Station 21186, 2012 – 2013

Station 21186					
Survey	Rep	UPSTREAM	BRIDGE	DOWNSTREAM	Overall
26-Apr-12	1	30.3	24.5	26.6	27.0
	2	21.2	29.9	30.6	26.9
	3	28.9	25.1	29.5	27.8
	Overall	26.5	26.4	28.8	27.2
1-May-12	1	15.7	23.0	22.3	20.0
	2	15.8	15.4	31.0	19.6
	3	20.5	21.8	24.9	22.3
	Overall	17.2	19.8	25.8	20.6
24-May-12	1	35.0	104.5	78.4	65.9
	2	26.9	94.6	94.0	62.1
	3	27.7	102.3	73.4	59.2
	Overall	29.6	100.4	81.5	62.3
29-May-12	1	35.1	424.3	140.6	127.9
	2	34.7	353.5	127.9	116.2
	3	37.1	533.0	122.0	134.1
	Overall	35.6	430.8	129.9	125.8
9-Apr-13	1	15.9	10.8	12.9	13.0
	2	19.5	14.6	17.6	17.1
	3	13.7	9.5	9.6	10.7
	Overall	16.2	11.4	13.0	13.4
22-Apr-13	1	5.3	6.4	11.5	7.3
	2	4.2	4.9	8.1	5.5
	3	2.0	3.9	9.7	4.2
	Overall	3.5	5.0	9.6	5.5
7-May-13	1	7.9	11.3	14.1	10.8
	2	5.0	8.5	14.0	8.4
	3	5.4	3.8	10.1	5.9
	Overall	6.0	7.1	12.6	8.1
31-May-13	1	26.1	41.0	164.0	56.0
	2	23.6	37.7	269.4	62.1
	3	36.1	44.1	154.7	62.7
	Overall	28.1	40.8	189.8	60.2

Table 2-5. Differences between control sampling locations at the non-birded bridge, Station 20018 (2012) and Station 16404 (2013) when migratory cliff swallows were not present. Significant differences ($p < 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for bridge – upstream (BU), downstream – upstream (DU), and downstream – bridge (DB). Values were \log_{10} -transformed prior to testing.

Station 20018 Spatial Control												
Location Comparison	BU	DU	DB	BU	DU	DB	BU	DU	DB	BU	DU	DB
Survey	27-Apr-12			3-May-12			30-May-12			25-Jun-12		
<i>n</i>	10	13	10	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	-26.3	-42.6	-8.1	6.3	-1.9	-8.2	1388.0	-7.1	-1395.0	12.1	5.6	-6.5
Pr ≥ t	0.005	0.002	0.039	0.124	0.417	0.031	<i><0.001</i>	0.019	<i><0.001</i>	<i><0.001</i>	0.001	0.006
Pr ≥ M	0.022	0.023	0.109	0.607	1.000	0.118	<i><0.001</i>	0.007	<i><0.001</i>	<i><0.001</i>	0.013	0.057
Pr ≥ S	0.004	0.003	0.027	0.135	0.609	0.026	<i><0.001</i>	0.015	<i><0.001</i>	<i><0.001</i>	0.003	0.008
Station 16404 Temporal Control												
Survey	24-Jan-13			6-Feb-13			27-Feb-13					
<i>n</i>	15	15	15	15	15	15	15	15	15			
Mean Difference (MPN/100 ml)	18.3	19.2	0.9	0.9	14.1	13.2	2.9	4.6	1.7			
Pr ≥ t	<i><0.001</i>	<i><0.001</i>	0.869	0.274	<i><0.001</i>	<i><0.001</i>	0.041	0.006	0.167			
Pr ≥ M	<i><0.001</i>	<i><0.001</i>	0.581	0.424	<i><0.001</i>	<i><0.001</i>	0.118	<i><0.001</i>	0.180			
Pr ≥ S	<i><0.001</i>	<i><0.001</i>	0.893	0.358	<i><0.001</i>	<i><0.001</i>	0.041	<i><0.001</i>	0.068			

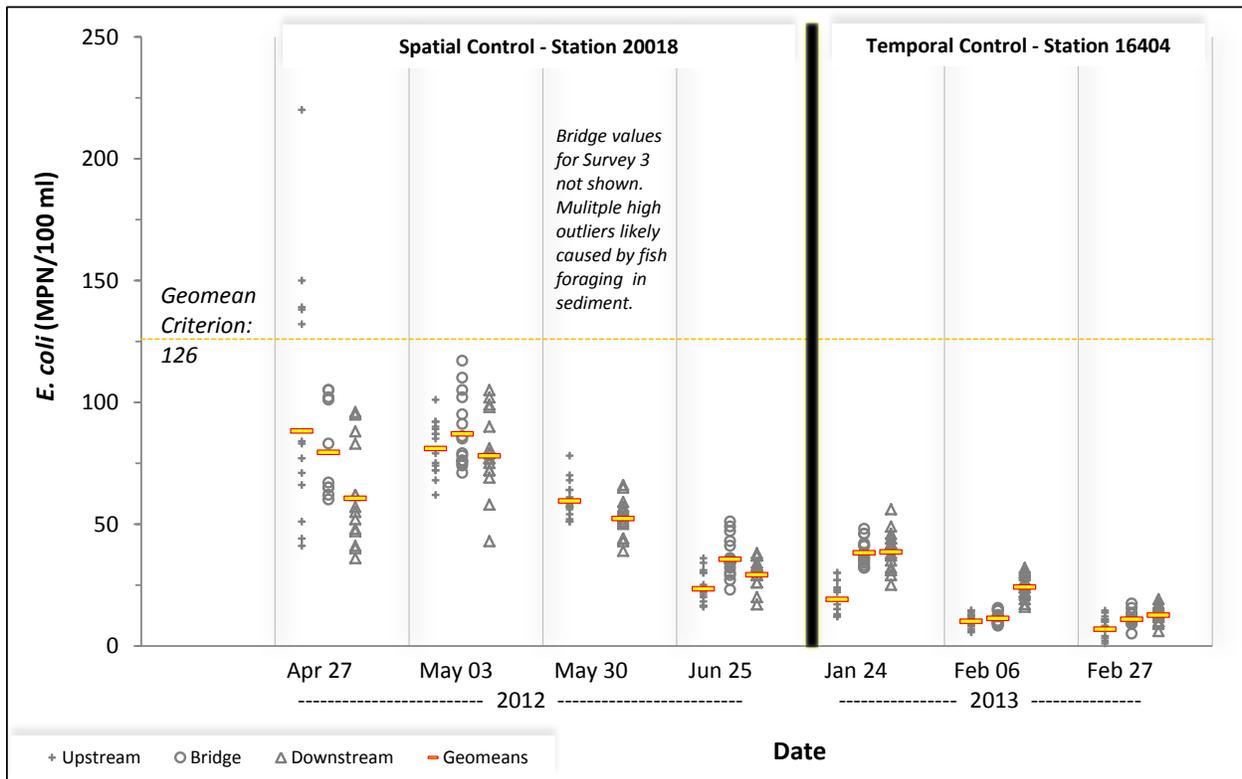


Figure 2-4. Control *E. coli* samples at the non-birded bridge, Station 20018 (2012) and Station 16404 (2013) when migratory cliff swallows were not present. State geomean criterion line provided for reference. The *n* for 27 Apr 2012 was 10 (bridge), 13 (upstream), and 15 (downstream).

Table 2-6. Geomeans of *E. coli* samples from upstream, bridge, and downstream locations at the spatial control Station 20018 (2012) and the temporal control Station 16404 (2013) when cliff swallows were absent

Station 20018 Spatial Control					
Survey	Rep	UPSTREAM	BRIDGE	DOWNSTREAM	Overall
27-Apr-12	1	152.9	98.8	91.3	111.3
	2	75.9	63.8	54.8	64.2
	3	45.1	.	44.2	44.6
	Overall	88.1	79.4	60.5	73.9
3-May-12	1	91.2	100.5	98.7	96.7
	2	83.8	85.0	75.7	81.4
	3	69.5	76.9	63.3	69.7
	Overall	81.0	86.9	77.9	81.9
30-May-12	1	56.4	934.5	58.5	145.6
	2	59.6	1261.9	46.2	151.4
	3	62.5	1235.5	52.5	159.5
	Overall	59.4	1133.7	52.2	152.0
25-Jun-12	1	30.7	43.0	32.4	35.0
	2	23.8	35.5	33.0	30.3
	3	17.5	29.3	23.2	22.8
	Overall	23.4	35.5	29.2	28.9
Station 16404 Temporal Control					
24-Jan-13	1	27.0	37.9	44.4	35.7
	2	19.2	38.1	40.0	30.8
	3	13.3	38.4	32.2	25.5
	Overall	19.0	38.1	38.5	30.4
6-Feb-13	1	11.2	11.9	27.2	15.3
	2	8.7	9.8	24.1	12.7
	3	10.4	12.1	21.3	13.9
	Overall	10.1	11.2	24.1	13.9
27-Feb-13	1	11.9	12.7	14.3	12.9
	2	7.7	11.1	13.7	10.5
	3	3.5	9.0	9.9	6.7
	Overall	6.8	10.8	12.5	9.7

Tests of Reps

The effect of time differences (30 – 60 minutes) between reps at each location were insignificant according to non-parametric sign (M) and signed-rank (S) tests (Tables 2-7 – 2-9). Only paired *t*-tests produced significant results but low *n* (5) render the results of this parametric *t*-test highly questionable. The mean differences between the first and third reps were always negative at Station 16404 including both treatment and control samples (Table 2-7). This suggests small decreases in *E. coli* with time of day, insignificant though they may be. The negative correlation between *E. coli* and time of day did not hold consistently for Stations 21186 and 20018 (Tables 2-8 and 2-9). Although time of day may have an influence on bacteria concentrations, significant patterns could not be discerned for this study.

Table 2-7. Differences between sampling reps at the densely-birded bridge, Station 16404, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for reps 1 – 2 (R12), reps 1 – 3 (R13), and reps 2 – 3 (R23). Values were \log_{10} -transformed prior to testing.

Rep Comparison	R12	R13	R23	R12	R13	R23	R12	R13	R23	R12	R13	R23
Survey	19-Apr-12			30-Apr-12			22-May-12			23-May-12		
<i>n</i>	5	5	5	5	5	5	2	0	0	5	5	5
Mean Difference (MPN/100 ml)	-0.2	-2.8	-2.6	-24.0	-51.0	-27.0	-4.0			-7.0	-2.4	4.6
Pr ≥ t	0.926	0.153	0.602	0.003	<i><0.001</i>	<i><0.001</i>	0.416			0.049	0.589	0.199
Pr ≥ M	1.000	0.375	1.000	0.063	0.063	0.063	0.500			0.125	1.000	0.375
Pr ≥ S	1.000	0.188	0.438	0.063	0.063	0.063	0.500			0.125	0.625	0.188
Survey	8-Apr-13			23-Apr-13			6-May-13			30-May-13		
<i>n</i>	5	5	5	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	-0.6	-3.5	-2.9	-1.0	-4.5	-3.5	2.6	-1.4	-4.0	-42.5	-45.0	-2.4
Pr ≥ t	0.791	0.186	0.079	0.879	0.272	0.278	0.474	0.175	0.140	0.254	0.176	0.457
Pr ≥ M	1.000	0.375	0.125	1.000	0.375	1.000	1.000	0.375	0.375	0.063	1.000	1.000
Pr ≥ S	1.000	0.188	0.125	0.750	0.438	0.313	0.625	0.250	0.188	0.063	0.313	0.625

Table 2-8. Differences between sampling reps at the moderately-birded bridge, Station 21186, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted. Mean differences are presented for reps 1 – 2 (R12), reps 1 – 3 (R13), and reps 2 – 3 (R23). Values were \log_{10} -transformed prior to testing.

Rep Comparison	R12	R13	R23									
Survey	26-Apr-12			1-May-12			24-May-12			29-May-12		
<i>n</i>	5	5	5	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	-6.0	-1.4	4.6	-0.2	5.4	5.6	-8.2	-7.2	1.0	-0.6	2.4	3.0
Pr ≥ t	0.312	0.695	0.486	0.958	0.214	0.124	0.001	0.090	0.759	0.910	0.631	0.653
Pr ≥ M	1.000	0.375	1.000	1.000	0.375	0.063	0.063	0.063	0.375	0.625	1.000	1.000
Pr ≥ S	0.438	0.625	0.875	1.000	0.188	0.063	0.063	0.063	0.625	0.875	1.000	0.813
Survey	9-Apr-13			22-Apr-13			7-May-13			31-May-13		
<i>n</i>	5	5	5	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	3.5	-2.4	-5.9	-2.2	-4.5	-2.3	-2.5	-2.2	0.3	-2.0	9.6	11.6
Pr ≥ t	0.350	0.406	0.116	0.735	0.234	0.045	0.173	0.255	0.732	0.383	0.057	0.053
Pr ≥ M	0.375	0.625	0.375	0.375	0.375	0.063	0.375	1.000	1.000	0.625	0.375	0.063
Pr ≥ S	0.438	0.375	0.125	0.625	0.313	0.063	0.188	0.313	0.875	0.375	0.125	0.063

Table 2-9. Differences between control sampling locations at the non-birded bridge, Station 20018 (2012) and Station 16404 (2013) when migratory cliff swallows were not present. Significant differences ($p = 0.05$) are highlighted. Mean differences are presented for reps 1 – 2 (R12), reps 1 – 3 (R13), and reps 2 – 3 (R23). Values were \log_{10} -transformed prior to testing.

Station 20018 Spatial Control												
Rep Comparison	R12	R13	R23	R12	R13	R23	R12	R13	R23	R12	R13	R23
Survey	27-Apr-12			3-May-12			30-May-12			25-Jun-12		
<i>n</i>	5	3	3	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	-79.6	-124.0	-30.0	-7.4	-21.8	-14.4	3.4	5.8	2.4	-7.0	-13.4	-6.4
Pr ≥ t	0.003	0.015	0.061	0.142	0.004	0.005	0.277	0.110	0.443	0.134	0.009	0.002
Pr ≥ M	0.063	0.250	0.250	1.000	0.063	0.063	0.375	0.375	1.000	0.375	0.063	0.063
Pr ≥ S	0.063	0.250	0.250	0.313	0.063	0.063	0.438	0.125	0.625	0.188	0.063	0.063
Station 16404 Temporal Control												
Survey	24-Jan-13			6-Feb-13			27-Feb-13					
<i>n</i>	5	5	5	5	5	5	5	5	5			
Mean Difference (MPN/100 ml)	-7.4	-13.8	-6.4	-2.8	-0.8	2.0	-3.8	-7.2	-3.3			
Pr ≥ t	0.111	<0.001	0.111	0.209	0.817	0.359	0.131	0.047	0.070			
Pr ≥ M	0.375	0.063	0.375	0.375	1.000	0.375	0.375	0.125	0.063			
Pr ≥ S	0.125	0.063	0.188	0.188	0.875	0.313	0.125	0.125	0.063			

Counts from Fecal Collection Frames

Median counts of bird feces from fecal collection frames at Station 16404 were 2 to 4 times higher at the bridge faces than at the 4-m mark and 6 times higher than at the 30-m mark (Table 2-10). At Station 21186 there is not a discernible pattern in counts with distance from the bridge (Table 2-11). It is notable, however, that during many surveys at this moderately-birded bridge, swallows periodically congregated in tight flocks of > 100 birds near the upstream and downstream sampling points collecting mud from near the water's edge. No such congregating activity was ever observed within the vicinity of the fecal frames. This could partially explain the lack of pattern in fecal counts. As expected, counts at the spatial control Station 20018 were negligible (Table 2-12). Interestingly, counts at the temporal control, Station 16404 during winter months (Table 2-12), were moderate, owing to the presence of 1-2 dozen house sparrows that persisted throughout the two-year study by kleptoparasitizing the cliff swallow nests. Vandalism in the form of footprints and board displacement prevented counts at Stations 21186 on 29 May 2012, and 20018 on 03 May 2012.

Table 2-10. Fecal counts at the densely-birded bridge, Station 16404, 2012 – 2013. Values normalized per 24-hr and rounded to nearest whole number; counts are for downstream (negative distance) to upstream (positive).

Station 16404										
Distance (m)	19Apr12	30Apr12	22May12	23May12	08Apr13	23Apr13	06May13	30May13	Sum	Median
-30	2	2	10	9	0	6	5	2	37	3.6
-10	3	3	11	4	3	5	5	1	36	3.7
-4	4	6	8	7	4	11	12	2	53	6.3
-1	26	5	184	147	8	18	32	0	419	21.7
1	22	10	13	14	21	60	15	14	170	14.5
4	16	7	2	4	14	28	5	0	76	5.9
10	12	3	6	1	14	29	3	0	68	4.6
30	1	1	3	0	2	13	7	3	30	2.5
Sum	86	37	237	185	67	169	83	22		

Table 2-11. Fecal counts at the moderately-birded bridge, Station 21186, 2012 – 2013. Values normalized per 24-hr; counts are for downstream (negative distance) to upstream (positive).

Station 21186										
Distance (m)	26Apr12	01May12	24May12	29May12	09Apr13	22Apr13	07May13	31May13	Sum	Median
-30	6	9	3	7	1	17	4	4	51	4.9
-10	2	13	4	*	4	8	5	4	40	4.1
-4	3	15	11	*	2	7	4	2	44	3.9
-1	2	12	11	7	3	18	6	13	73	9.0
1	0	1	11	2	4	27	7	2	54	3.0
4	2	7	4	5	1	24	10	4	57	4.5
10	2	4	11	5	1	23	20	2	68	4.5
30	4	2	6	8	2	11	15	4	51	5.0
Sum	20	64	61	34	18	135	70	36		

* No counts due to vandalism

Table 2-12. Fecal counts at the non-birded bridge, Station 20018 (2012) and Station 16404 (2013) when migratory cliff swallows were not present. Values normalized per 24-hr; counts are for downstream (negative distance) to upstream (positive).

		Station 20018 <i>Spatial Control</i>					
Distance (m)	27Apr12	03May12	30May12	25Jun12	Sum	Median	
-30	0	0	0	0	0	0.0	
-10	0	0	0	0	0	0.0	
-4	0	0	0	0	0	0.0	
-1	0	*	0	0	0	0.0	
1	0	0	0	1	1	n/a	
4	0	0	0	0	0	0.0	
10	0	0	0	0	0	0.0	
30	0	*	0	0	0	0.0	
Sum	0	0	0	1			
		Station 16404 <i>Temporal Control</i>					
Distance (m)	24Jan13	06Feb13	27Feb13	Sum	Median		
-30	0	0	1	1	0.0		
-10	0	0	0	0	0.0		
-4	0	0	0	0	0.0		
-1	0	1	3	4	1.0		
1	9	13	1	23	8.9		
4	0	0	1	1	0.0		
10	0	0	1	1			
30	0	0	2	2	0.0		
Sum	9	14	9				

* No counts due to vandalism

Flow and Water Chemistry

Drought conditions drastically reduced streamflow at the treatment bridges between 2012 and 2013 (Table 2-13). Diminished rainfall contributions to instream flow probably raised the relative contributions of groundwater to the system causing the increases of SC seen in 2013 at Station 21186. Elevated DO in 2013 at the treatment bridges is the result of sampling an hour or two later in the mornings during 2013 and a greater abundance of dense algae at both stations, while pH was stable across the two years of sampling at all three stations.

Table 2-13. Multi-probe readings and flow measurements from all stations, 2012 – 2013. ND = no data.

Station	Date	Time	Flow (cfs)	DSL/P	Water Temperature (°C)	Specific Conductivity (µS/cm)	DO (mg/L)	pH
<i>Treatment</i>								
16404	19-Apr-12	12:34	25	>7	23.8	526	9.0	8.2
16404	30-Apr-12	8:25	5.6	>7	24.2	529	7.9	8.2
16404	22-May-12	9:52	9.2	6	27.2	471	8.0	8.1
16404	23-May-12	8:53	8.3	7	26.0	476	7.8	8.1
16404	8-Apr-13	11:29	0.6	5	20.4	498	10.3	8.2
16404	23-Apr-13	12:33	0.1	5	21.6	536	10.9	8.0
16404	6-May-13	11:44	0.1	>7	21.1	546	10.1	8.1
16404	30-May-13	12:52	0.3	5	26.3	443	10.3	8.2
21186	26-Apr-12	9:25	56	>7	24.3	877	8.7	8.2
21186	1-May-12	9:29	44	>7	24.3	952	8.0	8.1
21186	24-May-12	9:33	37	>7	26.5	909	7.6	8.1
21186	29-May-12	11:17	27	>7	29.8	922	9.0	8.1
21186	9-Apr-13	10:24	11	6	20.3	1788	9.2	8.2
21186	22-Apr-13	12:46	8.8	4	24.6	ND	12.4	8.4
21186	7-May-13	11:51	5.6	>7	22.9	2048	11.5	8.3
21186	31-May-13	11:52	9.4	6	28.7	1404	10.3	8.3
<i>Spatial Control</i>								
20018	27-Apr-12	ND	66	>7	ND	ND	ND	ND
20018	3-May-12	10:48	48	>7	25.1	867	9.3	8.1
20018	30-May-12	10:03	26	>7	27.4	885	6.7	8.0
20018	25-Jun-12	10:15	16	5	29.7	1015	6.8	8.1
<i>Temporal Control</i>								
16404	24-Jan-13	10:50	0.7	>7	16.2	513	13.0	8.1
16404	6-Feb-13	9:42	0.1	>7	14.7	533	7.9	7.8
16404	27-Feb-13	10:22	0.2	>7	13.3	534	10.2	8.1

Discussion

This study was concerned with quantifying the impact of bridge-dwelling cliff swallow colonies on instream *E. coli* concentrations above, adjacent to, and below road crossings to determine whether swallow colonies introduced bias to samples collected upstream of bridges. Cliff swallows were chosen over bats and other bird species because they are among the most common species colonizing Texas bridges, their population numbers are more easily quantified than bats, and the timing of their migration and nesting phases are fairly predictable. Project resources were directed towards dry-weather sampling to

construct a robust dataset for addressing *E. coli* contributions from bird colonies under the flow conditions most common in Texas. Since it is common practice to collect stream samples immediately upstream of the upstream bridge face, collecting samples from this location in addition to 45-m upstream and 45-m downstream of the bridge enabled testing of whether sampling a few meters upstream of a bridge is sufficient distance to avoid the influence of nesting cliff swallows. To our knowledge no previous studies have explored this question with such study design and sample intensity. The only published study on cliff-swallow contributions to instream *E. coli* was Sejkora *et al.* (2011) and they collected a total of 78 samples, none at the bridgefaces. Collecting 360 samples from each of the treatment bridges plus 315 spatial and temporal control samples provided for a more robust analysis of spatial and temporal trends in *E. coli* in relation to bridge-dwelling bird colonies than has been previously produced.

Since this research was focused on bridges inhabited predominately by migratory cliff swallows, further study needs to be conducted on bridges possessing colonies of non-migratory birds to determine whether their continuous presence and defecation habits result in perennially higher instream bacteria concentrations. The small colony of house sparrows that occupied swallow nests at Station 16404 were the only vertebrates observed at the bridge during control surveys. If they represent the source of bacteria inputs causing the small—but nonetheless significant—increases in *E. coli* downstream of the bridge, a reasonable assumption, then the results of these winter samples support the hypothesis that colony size is a factor in the magnitude of differences in bacterial concentrations between upstream and downstream locations.

Colony size alone cannot explain variation in instream bacteria concentrations, it must be considered in the context of stream surface area and flow. The present study was located on a typical central Texas stream in reaches prone to sluggish flow in the late spring and summer months when rain events become less common. He *et al.* (2007) found that ponded waters contained higher levels of total coliform, fecal coliform, and enterococcus than flowing waters. Greater streamflow in higher-order rivers presumably dilutes bacteria inputs from bird bridge colonies. However, if higher streamflow is coursing through a wide, shallow channel then the river would capture more bird droppings and the greater surface area of the river would counteract the dilution effect.

Testing for rep differences in the present study accomplished two things. First, the results confirm that the efforts employed to diminish contamination of samples in reps 2 and 3 by avoiding contact with the water during sampling and spacing each rep by 30 minutes were successful. Secondly, though the three reps at a location were collected within about 60 minutes of each other and not diurnally, the results lend moderate support to the diurnal studies of Meays *et al.* (2006b) and Sejkora *et al.* (2011), both of whom found no significant correlations between *E. coli* concentrations and time of day. The tests of rep presented herein cannot confirm the absence of a time-of-day effect, but neither do they demonstrate an overt time-of-day effect that would contradict the findings of Meays *et al.* (2006b) and Sejkora *et al.* (2011). Desai and Rifai (2013) recorded significant differences in *E. coli* at 2-hour intervals during their diurnal study. It is possible that the time gap between reps was simply too small to capture variation caused by such factors as increased radiation.

The significant increases in *E. coli* downstream of the bridges in this study are not the only evidence of bird colony impacts on instream bacteria. The degree of variation among samples collected from a single

location was also generally greater for bridge and downstream locations (Figures 2-2 – 2-4). Furthermore, the variation in downstream samples was much greater during peak periods of bird activity. This increased variation is not surprising given that increased numbers of bird droppings into the stream lead to a greater likelihood of collecting a pocket of suspended bacteria.

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CHAPTER 3

Summary and Conclusions

The null hypothesis that there is no significant increase in *E. coli* between upstream, bridge, and downstream samples at a birded bridge during low and baseflow was firmly rejected by the results of this study. The consistent and strongly significant downstream increases in *E. coli* at the heavily birded Station 16404, as well as the weaker but significant increases at the moderately birded Station 21186, were the predicted outcomes if bird colonies are, in fact, the primary drivers of increases in bacteria concentrations between samples collected upstream and downstream of a bridge. During winter samples at Station 16404, 1-2 dozen sparrows maintained an active presence at the bridge. The low number of birds produced significant but weaker differences than samples collected at the same station when hundreds of swallows were present. The lack of downstream increases of *E. coli* at the non-birded station 20018 completes the picture: more birds means more bacteria inputs to the stream and stronger, more significant differences between upstream and downstream sampling locations.

Our findings demonstrate that multiple factors determine the magnitude of impacts on instream bacteria from colonies of cliff swallows at bridges. First, the density of birds over the stream must be high enough to produce significant increases in instream bacteria. One to two dozen sparrows over the narrow stream channel at Station 16404 were sufficient to produce significant increases between upstream and downstream locations. Second, samples taken during peak activity periods of the nesting cycle are likely to produce higher bacteria values. Defecation events around the nests are more frequent when both parent swallows are busy with nest construction and feeding nestlings. Spikes in fecal frame counts and higher *E. coli* values both coincided with fledging season in late May when chicks were roosting on the nest edge and lower lip of the beam. The emergence of fledglings effectively doubled the number of birds at each nest. Third, the background *E. coli* in a stream segment must be high enough that supplemental inputs from bird colonies are sufficient to bump station geomeans beyond assigned criterion. Only during peak cliff swallow activity at the birded bridges were differences between upstream and downstream concentrations greater than the geomean criterion of 126 CFU/100 ml. At all other times, the bird influence was not enough, by itself, to cause exceedence of the criterion downstream of the bridge. Fourth, the sampling distance upstream of a bridge required to avoid bias from bird colonies is variable and little specific guidance can be provided based on the results of this study. Fecal frame counts at Station 16404 showed a marked decrease in droppings merely 4-m upstream of the bridgeface. However, at Station 21186, birds were often seen foraging for mud in the banks about 45-m upstream and downstream of the bridge, and fecal counts were fairly uniform throughout the sampling reach. Fifth, hydromorphology governs both the likelihood of fecal matter hitting the stream surface area and the extent of bacteria dilution.

Considerable resources have been spent by government agencies in TMDL and WPP programs to assess instream bacteria and develop abatement strategies. These efforts have recently been called into question by stakeholders because of sampling protocols that some have contended are biased because many samples are collected immediately upstream of bridges where bird colonies could influence bacteria levels. Until now, data were scarce to inform this debate. This report corroborates the findings of Sejkora

et al. (2011) for dry-weather samples but provides more robust results on account of larger sample populations at both treatment and control bridges. Our findings demonstrate that bird colonies, depending on hydromorphological factors, can contribute significantly to instream *E. coli* concentrations within common sampling distances both upstream and downstream of bridges.

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CHAPTER 4

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Withers, P. C. 1977. Energetic aspects of reproduction by the cliff swallow. *The Auk* 94 (4):718-725.

Appendix A
Annotated Bibliography of Published Studies
Relevant to This Report, *Instream Bacteria*
Influences from Bird Habitation of Bridges

Each record includes reference information, abstract (when available), and annotations.

Barrett, M. E., J. F. Malina, and R. J. Charbeneau. 1995. Characterization of highway runoff in the Austin, Texas area Center for Research in Water Resources, The University of Texas, Austin, Texas.

NO ABSTRACT AVAILABLE

ANNOTATION:

Barrett *et al.* examined a variety of water quality parameters in rainfall runoff from bridges of varying traffic intensity. Bacteria constituents examined were total coliform, fecal coliform, and fecal streptococcus. The median event mean concentrations (cfu/100 ml) from runoff samples ranged 4200 – 189,000 for total coliform, 1000 – 116,000 for fecal coliform, and 3800 – 89,000 for fecal streptococcus. The authors note the results are consistent with a nationwide survey of highway runoff conducted by Driscoll *et al.*, 1990. Because this report is concerned with rainfall events, its immediate relevance to the bird bridge bacteria report is negligible. However, it implicates bridges as points of FIB accumulation during dry weather that flush into the underlying water body during runoff events. Follow-up investigations, if they involve wet-weather sampling, should address sources of FIB from bridge runoff to determine the percentage attributable to avian feces.

Bashar, R. 2010. Contamination of surface water associated with birds nesting on highway bridges, College of Engineering. The University of Texas at San Antonio, San Antonio, Texas.

ABSTRACT:

Highway bridge structures provide habitat to many different species of birds. To investigate the significance of the bacterial loadings from direct dropping of bridge nesting migratory birds, fecal coliform, *E. coli*, and Enterococci concentrations were measured in the Guadalupe River at Kerrville, TX. Samples were collected at three locations, such as, upstream, downstream and below the bridge for one and a half year which included two nesting seasons. The dry and wet weather samples were analyzed separately to obtain the combined effect of runoff and bird defecation on the pathogenic concentration of the river water. To obtain the diurnal variation of bacterial concentration, samples were collected every 2-4 hours in a day. The bacterial loadings under the bridge increased as soon as the birds started nesting on the bridge. The samples from downstream and below the bridge showed remarkably higher bacterial concentrations than that of the upstream in dry weather. In wet weather, *E. coli*, fecal Coliform and Enterococci concentrations went up to 1200, 1300, and 940 cfu/100 ml that are much higher than the bacterial water quality standards of 394, 400, and 89 cfu/100 ml respectively for contact recreational water designated by the State of Texas. Statistical analysis showed the concentrations of *E. coli* and fecal coliform remained significantly higher below the bridge than the concentrations upstream of the bridge. Also, the wet weather and dry weather samples for all three bacteria concentrations were significantly different. The probability distribution plots constructed for below the bridge location showed that the concentrations of *E. coli*, fecal coliform and Enterococci are likely to remain within the TCEQ specified contact recreation limit in 72, 65 and 35 percent of the days respectively.

ANNOTATION:

Although the details are inconclusive based on the information presented in the thesis, the general hypothesis that cliff swallows impact bacteria levels in streams seems supported by the data. Insufficient information regarding sampling technique, specific sampling dates, weather on the sampling dates, etc., render this a difficult paper to reference with any confidence.

Benton, C., F. Khan, P. Monaghan, W. N. Richards, and C. B. Shedden. 1983. The contamination of a major water supply by gulls (*Larus sp.*): A study of the problem and remedial action taken. *Water Research* 17: 789-798.

ABSTRACT:

In recent years one of the storage reservoirs supplying Loch Katrine water to Glasgow exhibited a serious deterioration in bacterial quality. This was associated with the winter nocturnal roosting habits of gulls (*Larus sp.*) on the reservoir. Investigations showed a significant correlation between the numbers of gulls roosting and the numbers of *E. coli* present in the water. Salmonellae organisms of identical serotypes were isolated from the gulls, from the untreated water and, on three occasions, from the treated water. Broadcasting species specific distress calls of *Larus* gulls proved to be an effective measure for discouraging gulls from roosting on the reservoir. The routine use of this bio-acoustic method of gull scaring reduced the bacteria in the water to numbers typical of an upland unpolluted reservoir.

ANNOTATION:

The authors examined the impact on *E. coli* in a Scotland reservoir by colonies of gulls. They found a highly significant relationship between the number of birds and the number of *E. coli*. This study supports the underlying assumption of the bird bridge bacteria report that cliff swallows, in fact, have the potential to significantly impact *E. coli* levels in streams. It is also relevant to interpreting results between bridges with greater and lesser densities of birds.

Brown, C. R., and M. B. Brown. 1995. Cliff swallow (*Petrochelidon pyrrhonota*). *The Birds of North America Online*. <http://bna.birds.cornell.edu/bna/species/149/articles/introduction>. Accessed 29 March 2013.

NO ABSTRACT AVAILABLE

ANNOTATION:

Birds of North America Online is a strictly electronic journal. No other resources on the natural history of the cliff swallow provided the breadth and depth of information covered by Brown and Brown. Important information from the article includes time spent at the nest during different phases of nesting and fledging and defecation habits (e.g., "Adults fly out from nest several meters to defecate").

Cho, K. H., Y. A. Pachepsky, J. H. Kim, A. K. Guber, D. R. Shelton, and R. Rowland. 2010.
Release of *Escherichia coli* from the bottom sediment in a first-order creek: experiment and reach-specific modeling. *Journal of Hydrology* 391: 322-332.

ABSTRACT:

Escherichia coli release from streambed sediments may substantially affect microbial water quality. Models of *E. coli* release and transport commonly use a single set of parameters for the whole stream or reservoir, yet little is known about the magnitude and sources of the variability of parameters of the streambed bacteria release. The objectives of this work were: (a) to obtain and compare parameters of streambed *E. coli* resuspension in three stream reaches with distinctly different bottom sediment textures, and (b) to see whether the modeling of streambed *E. coli* resuspension with reach-specific parameters could provide substantially better accuracy than modeling with a single set of parameters. Sediment particle size distributions and the streambed *E. coli* concentrations were measured along a first-order creek in the USDA-ARS OPE3 experimental watershed in Maryland. Afterwards, 80 m³ of water were released into the creek at a rate of 60 L per second in four equal allotments separated by 1–3 min intervals. Flow rates and *E. coli* concentrations were monitored with automated samplers at the ends of the three reaches with a total length of 630 m. A high concentration of streambed *E. coli* (“hotspot”) resuspended within the first reach caused a pulse of high *E. coli* concentrations that propagated along the creek without substantial attenuation; inputs of sediment-borne *E. coli* from the next two reaches were relatively small. The *E. coli* transport model included one-dimensional Saint–Venant and advective–dispersive equations. The calibrated roughness coefficient values were comparable for the three reaches, whereas the critical stress and the entrainment rate differed among reaches by a half order and an order of magnitude, respectively. Overall, better accuracy was observed when the model contained reach specific parameters. Additional research is needed to understand which and how sediment properties affect parameters of streambed *E. coli* release into the water column.

ANNOTATION:

Cho *et al.* present results demonstrating that resuspension of *E. coli* from benthic sediments can be “the major factor of microbiological water quality in streams.” Although the resuspension driver was hydrological (an artificial high-flow simulation that excluded runoff), it is reasonable to assume that resuspension caused by biological disturbance of benthic sediment has a similar effect on *E. coli* values.

Chu, Y., C. Salles, M.-G. Tournoud, P. Got, M. Troussellier, C. Rodier, and A. Caro. 2011.
Faecal bacterial loads during flood events in Northwestern Mediterranean coastal rivers. *Journal of Hydrology* 405:501-511.

ABSTRACT:

In Mediterranean coastal rivers, floods last often less than a few hours but supply large amounts of contaminants to transitional and coastal waters. Estimating flood loads requires appropriate sampling strategies. We applied flood-scale sampling for the survey of two rivers flowing into the Thau lagoon (France). Two bacterial indicators were considered, thermotolerant coliforms (TTC) and faecal streptococci (FC). During floods, concentrations of indicator bacteria associated with non-mineral suspended solids increased quickly with the rising flow, their decrease during the recession period was

slow and erratic. Statistical analysis was performed on total bacterial flood loads measured during 20 floods, versus hydrological variables and land-use characteristics. The analysis highlighted the significant impacts of human pollution sources together with the magnitude of the flood. Regarding the results, the best linear regression models linked total bacterial flood loads to peak discharge for both TTC and FS, reinforcing the assumption that in-stream bacterial stores play an important role in the level of bacterial flood loads in Mediterranean coastal rivers. At an annual scale, between 13.9 and 16.6 \log_{10} cfu of TTC could be supplied depending on the hydrological conditions during the year. Over the 12 year period, from 1994 to 2006 it was shown that the flood loads were responsible for at least 98% of the TTC total annual load and in 8 of 12 years the floods contributed to at least 99.9% of the annual loads. Over the same period on average the single major flood represents 74% of the total annual load. The contribution of in-stream bacterial stores was demonstrated but spatial variations in total flood loads showed that this contribution is difficult to evaluate. Bacteria from land stores appeared to be negligible in both catchments.

ANNOTATION:

Supports the growing evidence that in-stream and near-shore sediment stores of bacteria are a major component of the total bacteria in the early stages of high flow events. It is cited as a supporting reference for the decision to sample only during dry-weather conditions.

Collins, R., S. Elliott, and R. Adams. 2005. Overland flow delivery of faecal bacteria to a headwater pastoral stream. *Journal of Applied Microbiology* 99:126-132.

ABSTRACT:

AIMS: To quantify and derive statistical relationships with which to predict the delivery of faecal bacteria (*Escherichia coli*) to a pastoral stream, by overland flow.

METHODS AND RESULTS: A large-scale (1050 m²) rainfall simulator, located upon a steep (18 degrees) grazed hillside in New Zealand, was used to simulate 11 heavy rainfall events. Overland flow was generated and sampled throughout each event, before discharging to a headwater stream. The samples were subsequently analyzed to determine the concentration of *E. coli*. Statistical analysis showed that the time elapsed since the last period of grazing was a statistically significant predictor of both the total number (load) and concentrations of *E. coli* in overland flow. Between 10⁵ and 10⁸ *E. coli* per m² of hillside were delivered to the stream within overland flow during each event, and peak concentrations ranged between 10³ and 10⁷ most probable number per 100ml.

CONCLUSIONS: Under heavy rainfall on steep pastoral land, overland flow can transport substantial levels of faecal bacteria to streams. Under such conditions, it is unlikely that vegetated buffer strips will be particularly effective at attenuating bacteria within overland flow.

SIGNIFICANCE AND IMPACT OF THE STUDY: This work has improved understanding of the importance of overland flow as a process contributing to the contamination of pastoral streams by faecal bacteria. In addition, the predictive relationships derived can be incorporated within catchment models.

ANNOTATION:

Cited as a recent example of quantified loads of bacteria washing into a stream from overland flow. It was in light of the thesis of this paper, and others, that samples in the bird bridge bacteria study were collected under dry-weather conditions.

Desai, A. M., and H. S. Rifai. 2013. *Escherichia coli* concentrations in urban watersheds exhibit diurnal sag: implications for water-quality monitoring and assessment. *Journal of the America Water Resources Association* 49:766-779.

ABSTRACT:

The variability of indicator bacteria over a fine resolution time scale on the order of minutes has yet to be fully understood. In this study, we collected more than 700 *Escherichia coli* samples at a 10- and 30-min resolution in an urban watershed in Houston. A Bacteria Diurnal Sag (BDS) marked with daytime exponential decay followed by an exponential nighttime regeneration was observed. This pattern was observed during all sampled events but varied depending on other variables. The concentrations during a 24-h period varied 1 to 5 orders of magnitude and the fecal load was at least 10 times lower than what would be obtained using a single morning *E. coli* measurement, the typical sampling scheme in most monitoring programs. Decay rates, ranging from 3.67 to 24.7/day, decreased *E. coli* concentrations to below the water-quality standards from 14:00 to 18:00 h and were strongly influenced by water temperatures and solar radiation intensities. Rapid regeneration occurred on the order of 9.41 to 64.1/day allowing *E. coli* concentrations to return to their pre-decay levels. The data indicated that four to six samples taken between 06:00 and 18:00 h may be sufficient to define the BDS depending on stream conditions, and that a threshold concentration of approximately 100 CFU/dl (most probable number in a deciliter) existed for the studied urban watershed. These findings have significant implications for water-quality monitoring, regulation, and compliance.

ANNOTATION:

The authors recorded daytime decay and nightly increases in *E. coli* concentrations covering several orders of magnitude. They attribute the day-night disparities to a host of environmental factors but sunlight radiation was chief among them. They found significant differences in *E. coli* series collected 2 hours apart and recommended that 4-6 samples be collected between 10:00 and 16:00 to capture variation in the window of time when temporal variation is at its peak. It is possible that the lack of significant differences between reps in the current bird study is because the time gap between reps was simply too short to account for diurnal variation in *E. coli* that might only be detectable at larger (i.e., multi-hour) time scales.

Emlen, J. T., Jr. 1954. Territory, nest building, and pair formation in the cliff swallow. *The Auk* 71: 16-35.

NO ABSTRACT AVAILABLE

ANNOTATION:

This paper provides detailed observations regarding breeding and nest-building among cliff swallows. Not cited directly, but a useful paper for understanding cliff swallow behavior in the field.

Fleming, R., and H. Fraser. 2001. The impact of waterfowl on water quality--Literature review. University of Guelph, Ridgetown, Ontario, Canada.

NO ABSTRACT AVAILABLE

ANNOTATION:

This paper appears to have been prepared for a college class and is unsuitable for citation. However, it collects and summarizes various papers relevant to the current study.

Fogerty, L. R., S. K. Haack, M. J. Wolcott, and R. L. Whitman. 2003. Abundance and characteristics of the recreational water quality indicator bacteria *Escherichia coli* and enterococci in gull faeces. *Journal of Applied Microbiology* 94: 865-878.

ABSTRACT:

Aims: To evaluate the numbers and selected phenotypic and genotypic characteristics of the faecal indicator bacteria *Escherichia coli* and enterococci in gull faeces at representative Great Lakes swimming beaches in the United States.

Methods and Results: *E. coli* and enterococci were enumerated in gull faeces by membrane filtration. *E. coli* genotypes (rep-PCR genomic profiles) and *E. coli* (Vitek® GNI+) and enterococci (API® rapid ID 32 Strep and resistance to streptomycin, gentamicin, vancomycin, tetracycline and ampicillin) phenotypes were determined for isolates obtained from gull faeces both early and late in the swimming season. Identical *E. coli* genotypes were obtained only from single gull faecal samples but most faecal samples yielded more than one genotype (median of eight genotypes for samples with 10 isolates). *E. coli* isolates from the same site that clustered at ≥85% similarity were from the same sampling date and shared phenotypic characteristics, and at this similarity level there was population overlap between the two geographically isolated beach sites. Enterococcus API® profiles varied with sampling date. Gull enterococci displayed wide variation in antibiotic resistance patterns, and high-level resistance to some antibiotics.

Conclusions: Gull faeces could be a major contributor of *E. coli* (10^5 – 10^9 cfu g⁻¹) and enterococci (10^4 – 10^8 cfu g⁻¹) to Great Lakes recreational waters. *E. coli* and enterococci in gull faeces are highly variable with respect to their genotypic and phenotypic characteristics and may exhibit temporal or geographic trends in these features.

Significance and Impact of the Study: The high degree of variation in genotypic or phenotypic characteristics of *E. coli* or enterococci populations within gull hosts will require extensive sampling for adequate characterization, and will influence methods that use these characteristics to determine faecal contamination sources for recreational waters.

ANNOTATION:

This study on gulls in the Great Lakes corroborates the assertions of Benton *et al.* (1983) that gulls are a major source of enteric bacteria to water bodies. It is not unreasonable to assume that other avian species such as cliff swallows, depending on colony density and hydrologic factors, could also contribute significantly to *E. coli* loads in streams.

Fujioka, R. S., and O. T. Narikawa. 1982. Effect of sunlight on enumeration of indicator bacteria under field conditions. *Applied and Environmental Microbiology* 44: 395-401.

ABSTRACT:

The effect of sunlight on the enumeration of fecal coliform (FC) and fecal streptococcal (FS) bacteria when water samples are collected in containers and brought back to the laboratory for analysis or when the water samples are filtered through membranes on site was determined. FC and FS in raw sewage stored in clear glass or translucent polyethylene containers were resistant to the effects of sunlight. However, under the same conditions of storage and exposure to sunlight, 90% of FC and FS in sewage diluted 1:100 in seawater were inactivated within 13 to 32 min. When sewage was similarly diluted in stream water and exposed to sunlight, 90% of FC were inactivated after 28 to 38 min, whereas 90% of FS were not inactivated even after a 2-h exposure to sunlight. Other experiments showed that 90 to 99% of FC and FS retained on membranes were inactivated when these membranes were exposed to sunlight for 10 to 15 min. FS were inherently more resistant to sunlight inactivation than were FC. Finally, evidence was obtained to show that sunlight initially stresses the bacteria but eventually causes cell death.

ANNOTATION:

The authors convincingly demonstrate the lethal and semi-lethal effect of sunlight radiation on fecal coliform and fecal streptococcal bacteria suspended in both water and membranes. The results support the stashing of field samples on ice in a closed cooler as quickly as possible. The conclusions also have implications for *insitu* transport of bacteria. The persistence of suspended FIB from bird droppings deposited directly into a stream is regulated not only by hydrology and TSS but also by meteorological conditions, namely sunny versus cloudy days.

Geldreich, E. E., L. C. Best, B. A. Kenner, and D. J. Van Donsel. 1968. The bacteriological aspects of stormwater pollution. *Water Environment Federation* 40 (11):1861-1872.

NO ABSTRACT AVAILABLE.

ANNOTATION:

One of the early papers demonstrating that overland runoff carries a large and significant load of bacteria into streams regardless of landuse. Cited as reason to sample under dry-weather conditions and avoid the confounding effects of stormwater loads.

Gorenzel, W. P., and T. P. Salmon. 1982. The cliff swallow--biology and control, *Proceedings of the Tenth Vertebrate Pest Conference*.

ABSTRACT:

Cliff swallows (*Petrochelidon pyrrhonota*) nesting in colonies on man-made structures can cause aesthetic problems and health hazards. Cliff swallows are migratory, wintering in South America and breeding throughout most of North America. Cliff swallows have a homing tendency to old colonies and are attracted to the gourd-shaped mud nests. Egg laying begins before nest construction is finished; clutch size averages 3 or 4 eggs. Re-nesting is common if a nest fails and some pairs may raise 2 broods in 1 nesting season. Cliff swallows may be present at a colony for up to 132 days. Cliff swallows are protected by the Migratory Bird Treaty Act of 1918, and a permit from the United States Fish and Wildlife Service is required for certain control activities. Successful control methods include nest removal by water hose or a pole, and exclusion using netting, poultry wire, or strip doors. Nest substrate modification is successful in some instances. Methods employed with little success or that remain unproven include metal spines, repellents, frightening devices, predator models, taped alarm calls, and a fresh coat of paint. Attention to architectural design may alleviate cliff swallow nesting problems.

ANNOTATION:

Provides general information on the biology and ecology of the cliff swallow including migration schedules, typical durations for nesting and brooding phases, and feeding and defecation habits.

Graczyk, T. K., A. C. Majewska, and K. J. Schwab. 2008. The role of birds in dissemination of human waterborne enteropathogens. *Trends Parasitol* 24: 55-9.

ABSTRACT:

Cryptosporidiosis, giardiasis and microsporidiosis are serious human diseases of waterborne origin; their etiologic agents and a substantial fecal coliform load can enter surface, drinking and recreational water resources from aquatic birds. The aim of this article is to present interactions between waterfowl and these waters that imply a negative public health impact, reinforcing the need for either better water-quality indicators or for water monitoring specifically for *Cryptosporidium*, *Giardia* and microsporidia. Where justifiable, the presence of waterfowl should be supported;

however, management of drinking and recreational water resources needs to be improved by incorporating effective protection measures for pathogens linked to these birds.

ANNOTATION:

Graczyk *et al.* present useful data and references connecting various avian species to human pathogens. Of particular interest is Table 1 which summarizes reported concentrations of pathogens by bird species and pathogen group. The authors take the works of Benton *et al.* (1983) and Fogarty *et al.* (2003), which connect bird populations to high amounts of fecal matter, and connects the avian fecal matter described in those papers to human health concerns.

Gupta, M. K., R. W. Agnew, D. Gruber, and W. Kreuzberger. 1981. Constituents of highway runoff vol. IV: characteristics of runoff from operating highways. *FHWA-RD-81-45*. Federal Highway Administration, Washington, D.C.

ABSTRACT:

This report relates to the identification and quantification of the constituents of highway runoff. It includes the details of monitoring site selection, field monitoring procedures, analysis of accumulated data, conclusions, significant findings and limitations. A total of 159 storm events were monitored at six sites between Spring of 1976 to September, 1977. The data were evaluated for: rainfall/runoff relationships; highway runoff pollutants loadings and variations with time; differences in pollutant characteristics from paved and non-paved areas; correlation of pollutants amongst measured parameters as well as with highway operation related factors. This is the fourth volume of a six volume document series.

ANNOTATION:

Cited as an example of an early study of bridge runoff pollutants.

He, L.-M., J. Lu, and W. Shi. 2007. Variability of fecal indicator bacteria in flowing and ponded waters in southern California: Implications for bacterial TMDL development and implementation. *Water Research* 41: 3132-3140.

ABSTRACT:

Recreational water quality is assessed by using water quality objectives for fecal indicator bacteria (FIB) including total coliform, fecal coliform (or *E. coli*), and/or Enterococcus. It is required under the Clean Water Act that a TMDL be developed for a bacteria-impaired water body. The development and implementation of bacterial TMDLs has proven challenging and often difficult due to unknown source(s) of FIB. This study found that FIB levels varied significantly in flowing water, ponded water, and associated sediment. FIB levels in isolated ponded water in waterways were significantly higher than in flowing water. Sediment under ponded water contained a great amount of FIB. Furthermore, FIB concentrations in ponded water tended to increase with increasing

water temperature and to decrease with increasing water salinity. The result provides the field evidence of survival/growth of FIB in water and sediment under ambient conditions in southern California. A holistic approach including natural sources (e.g., a reference system) should be considered for practical and applicable purposes while developing and implementing bacterial TMDLs for pathogen-impaired waterbodies.

ANNOTATION:

He *et al.* (2007) found that ponded waters contained higher levels of FIB (total coliform, fecal coliform, and enterococcus) than flowing waters. They noted along with Stephenson and Rychert (1982) and Cho *et al.* (2010), that hydrological disturbance of sediment can create pulses of FIB in the water column through entrainment of in-channel bacteria stores. At station 20018, the dramatic spikes in *E. coli* at the ponded bridge location may be the result of benthic feeding behavior by carp (*Cyprinus carpio*) and turtles which, on the two dates in question, were observed disturbing sediment throughout the deeper pool under the bridge such that large plumes of suspended sediment extended many meters downstream from the points of disturbance where the animals were rummaging through the sediment. This has implications for bacteria sampling techniques such that field technicians should seek flowing water free of turbidity caused by recent sediment disturbances. Additionally, results might indicate that the pooled conditions common to central Texas intermittent streams may exacerbate FIB proliferation.

Hussong, D., J. M. Damaré, R. J. Limpert, W. J. L. Sladen, R. M. Weiner, and R. R. Colwell. 1979. Microbial impact of Canada geese (*Branta canadensis*) and whistling swans (*Cygnus columbianus columbianus*) on aquatic ecosystems. *Applied and Environmental Microbiology* 37: 14-20.

ABSTRACT:

Quantitative and qualitative analyses of the intestinal bacterial flora of Canada geese and whistling swans were carried out with the finding that wild birds harbor significantly more fecal coliforms than fecal streptococci. The reverse was typical of captive and fasting birds. Neither *Salmonella spp.* nor *Shigella spp.* were isolated from 44 migratory waterfowl that were wintering in the Chesapeake Bay region. Enteropathogenic *Escherichia coli* were detected in seven birds. Geese eliminated 107 and swans 109 fecal coliforms per day. Results of in situ studies showed that large flocks of waterfowl can cause elevated fecal coliform densities in the water column. From the data obtained in this study, it is possible to predict the microbial impact of migratory waterfowl upon aquatic roosting sites.

ANNOTATION:

Hussong *et al.* demonstrate that migratory geese and swans increased the sediment levels of FC in Chesapeake Bay waters, including in oyster beds, during the bird season from 13 to 170 FC per 100 ml. Given recent research substantiating the relative contribution of sediment FIB to aquatic systems, the presence of cliff swallows may have both short and long-term impacts by their FIB settling into the channel sediment and shoreline until it is disturbed by human or animal activity or rain events. In other words, direct deposition of fecal matter to the water column may elevate readings substantially in short-term measures but a larger, broader impact is felt when disturbances unsettle FIB accumulated in the shore and instream sediment during dry weather.

Jamieson, R. C., D. M. Joy, H. Lee, R. Kostaschuk, and R. J. Gordon. 2005. Resuspension of sediment-associated *Escherichia coli* in a natural stream. *Journal of Environmental Quality* 34: 581-589.

ABSTRACT:

In this study, a tracer bacteria was used to investigate the resuspension and persistence of sediment-associated bacteria in a small alluvial stream. The study was conducted in Swan Creek, located within the Grand River watershed of Ontario, Canada. A 1.1-m² section of the bed was seeded with a strain of *Escherichia coli* resistant to nalidixic acid (*E. coli* NAR). The survival, transport, and redistribution of the tracer bacteria within a 1.7-km river section downstream of the source cell was assessed for a 2-mo period following the introduction of the tracer bacteria. This study has illustrated that enteric bacteria can survive in bed sediments for up to 6 wks. and that inactivation of the tracer bacteria resembled typical first-order decay. Critical conditions for resuspension, as well as resuspension rates, of sediment-associated bacteria were determined for several storm events. The critical shear stress for *E. coli* NAR resuspension in Swan Creek ranged from 1.5 to 1.7 N m⁻², which is comparable with literature values for critical shear stresses for erosion of cohesive sediments. Bacteria resuspension was primarily limited to the rising limb of storm hydrographs implying that a finite supply of sediment-associated bacteria are available for resuspension during individual storm events. The information presented in this paper will further the development of representative microbial water quality models.

ANNOTATION:

Jamieson *et al.* demonstrate that enteric bacteria can persist in stream sediments for 6 weeks. This supports the hypothesis presented in Sejkora *et al.* (2011) that the impact of nesting birds at bridges on instream bacteria can linger for more than a month after the birds have migrated away.

Kirschner, A. K. T., T. C. Zechmeister, G. G. Kavka, C. Beiwl, A. Herzig, R. L. Mach, and A. H. Farnleitner. 2004. Integral strategy for evaluation of fecal indicator performance in bird-influenced saline inland waters. *Applied and Environmental Microbiology* 70: 7396-7403.

ABSTRACT:

Wild birds are an important nonpoint source of fecal contamination of surface waters, but their contribution to fecal pollution is mostly difficult to estimate. Thus, to evaluate the relation between feces production and input of fecal indicator bacteria (FIB) into aquatic environments by wild waterfowl, we introduced a new holistic approach for evaluating the performance of FIB in six shallow saline habitats. For this, we monitored bird abundance, fecal pellet production, and the abundance of FIB concomitantly with a set of environmental variables over a 9-month period. For estimating fecal pellet production, a new protocol of fecal pellet counting was introduced, which was called fecal taxation (FTX). We could show that, over the whole range of investigated habitats, bird abundance, FTX values, and FIB abundance were highly significantly correlated and could demonstrate the good applicability of the FTX as a meaningful surrogate parameter for recent bird abundances and fecal contamination by birds in shallow aquatic ecosystems. Presumptive enterococci (ENT) were an excellent surrogate parameter of recent fecal contamination in these saline environments for samples collected at biweekly to monthly sampling intervals while presumptive *Escherichia coli* and fecal coliforms (FC) were often undetectable. Significant negative correlations with salinity indicated that *E. coli* and FC survival was hampered by osmotic stress. Statistical analyses further revealed that fecal pollution-associated parameters represented one system component independent from other environmental variables and that, besides feces production, rainfall, total suspended solids (direct), and trophy (indirect) had significant positive effects on ENT concentrations. Our holistic approach of linking bird abundance, feces production, and FIB detection with environmental variables may serve as a powerful model for application to other aquatic ecosystems.

ANNOTATION:

The study was carried out in Neusiedler See-Seewinkel National Park, Eastern Austria. Its primary contribution to the current cliff swallow project is that it helps quantify the magnitude of fecal contributions and associated FIB from waterfowl to waterbodies. Notably, fecal pellet production was strongly and positively correlated with FIB abundance, primarily enterococci in this case, because of the moderately saline waters of their study location.

Leasure, D. R., R. Kannan, and D. A. James. 2010. House sparrows associated with reduced cliff swallow nesting success. *The Wilson Journal of Ornithology* 122: 135-138.

ABSTRACT:

We quantified the impact of nesting and roosting House Sparrows (*Passer domesticus*) on nesting success of Cliff Swallows (*Petrochelidon pyrrhonota*) in colonies in western Arkansas in 2007 and 2008. Two sections of a large swallow colony under a bridge with House Sparrows were compared in 2007 to two sections with little House Sparrow usage. Nesting success of Cliff Swallows (percent of nests yielding at least 1 chick) was 61% in sections with low House Sparrow activity, significantly higher than the 30% in

sections with high House Sparrow activity. House Sparrows defended a broad zone surrounding their nests from Cliff Swallow nesting attempts. We compared the proportion of nests used, clutch sizes, and brood sizes of Cliff Swallows in two colonies in 2008, one with and one without House Sparrow activity. In the colony without House Sparrow activity, 48% of old and new nests were used by swallows versus only 8% in the colony with House Sparrows. Swallow clutch sizes were similar in the two colonies, but swallow brood sizes in the colony with no House Sparrows were significantly higher, mean 5 2.3 nestlings per nest (mode 5 2; 75th percentile 5 3) compared to 0.8 nestlings (mode 5 0; 75th percentile 5 1) in the colony with House Sparrows. This suggests Cliff Swallows are less successful when House Sparrows are present in colonies.

ANNOTATION:

This paper pertains only weakly to the current study but it provides details on the kleptoparasitism of cliff swallow nests by house sparrows (a.k.a. English sparrows), a phenomenon prevalent at the bridges in the current cliff swallow investigation.

McDonald, A., and D. Kay. 1981. Enteric bacterial concentrations in reservoir feeder streams: baseflow characteristics and response to hydrograph events. *Water Research* 15: 961-968.

ABSTRACT:

This paper reports on work carried out from June 1976 to May 1978 on feeder streams to Thruscross Reservoir. The study examined short term changes in total coliform and *Escherichia coli* concentrations during five diurnal phases and 11 hydrograph events. Statistical analysis of the results show that highly significant increases in enteric bacterial concentrations occurred in nine of the 11 hydrograph events examined. The results suggest that a flushing mechanism may operate in the transport of enteric bacteria into upland reservoirs.

ANNOTATION:

A foundational paper from the early 1980s substantiating the flushing of enteric bacteria early in the rising limb of a hydrograph. It supports both the decision to restrict bacteria sampling in the current study to >7 DSLP and to limit sediment disturbance during sampling.

McDonald, A., D. Kay, and A. Jenkins. 1982. Generation of fecal and total coliform surges by stream flow manipulation in the absence of normal hydrometeorological stimuli. *Applied and Environmental Microbiology* 44: 292-300.

ABSTRACT:

The response of *Escherichia coli* and total coliform concentration to increases in river discharge was investigated. Artificial hydrographs were generated on eight occasions between 21 October 1979 and 3 March 1981 by releasing water from Thruscross Reservoir in North Yorkshire into Fewston Reservoir. The majority of the releases were made after rainless periods to isolate the effects of stream channel entrainment from those induced by rainfall on the land surface. In the absence of rainfall, bacterial

concentrations are shown to increase more than 10-fold in response to stage increases. It is suggested that two stores of bacteria must exist on the catchment, the first being a land store and the second a channel or near-channel store. Movement from the land to the channel store must relate to hill slope hydrological processes, whereas movement between stores in the channel fluvial system may be closely allied to sedimentary processes. Some consideration is given to bacterial levels in relation to European Economic Communities guidelines for contact recreation.

ANNOTATION:

The authors present some of the early research on instream sediment stores of enteric bacteria and the results support efforts to reduce sediment disturbance during *E. coli* sampling. The paper also aids interpretation of *E. coli* spikes at Station 20018 of this study on dates when plumes of sediment were seen emanating from benthic foraging by turtles and fish.

Meays, C. L., K. Broersma, R. Nordin, A. Mazumder, and M. Samadpour. 2006. Diurnal variability in concentrations and sources of *Escherichia coli* in three streams. *Canadian Journal of Microbiology* 52: 1130-1135.

ABSTRACT:

Microbial contamination is a major concern for drinking water worldwide. Many monitoring protocols that use one or very few samples are inadequate and introduce a very large margin of error. An intensive sampling program needs to be conducted to characterize the *Escherichia coli* concentrations of a source water stream prior to establishing a monitoring program so that the sample frequency can be determined statistically, based on an acceptable margin of error. Developing meaningful monitoring programs for managing bacterial water quality is dependent on scientific data that determine the bacterial sources. In this study, three streams from drinking water watersheds were sampled every 15 min over a 24 h period on three different days to determine the concentrations of *E. coli* and to identify their sources, using ribosomal RNA finger printing (ribotyping). The concentrations of *E. coli* varied throughout the day in each of the three streams. Ribotyping identified many different animal sources of *E. coli* in the samples. The sources of *E. coli* varied significantly with stream ($P < 0.001$, $df = 16$). The development of monitoring programs for watersheds needs to consider the watershed, and care needs to be taken in selecting appropriate sample sites, sampling regime, and number of samples taken during each sampling period. This note provides a prescription for the development of monitoring programs for watersheds.

ANNOTATION:

Meays *et al.* present diurnal data but, because of lack of replication, the results are indeterminate regarding diurnal patterns. A useful application of their results is that drizzle, without runoff, occurred during one of their diurnal studies without a significant difference between pre- and post-drizzle samples. This corroborates the findings of the present study at Station 16404 on 6 February 2013 when light drizzle fell without significant impact on our samples.

Meays, C. L., K. Broersma, R. Nordin, A. Mazumder, and M. Samadpour. 2006. Spatial and annual variability in concentrations and sources of *Escherichia coli* in multiple watersheds. *Environmental Science Technology* 40: 5289-5296.

ABSTRACT:

Nonpoint source fecal contamination is a concern for drinking water supplies worldwide. In this study, 4812 *E. coli* isolates were classified to source. Results of this experiment show that the fecal coliform (FC) counts varied by year, month, and site, for each of the watersheds sampled. For both years, the lowest FC counts tended to be at the highest elevation sites followed by the drinking water intake sites at the lowest elevation. The highest FC counts tended to be at the mid-elevation sites on BX, Deer, and Duteau Creeks. The sources of *E. coli* varied significantly with stream for 2003 and 2004 ($P < 0.001$, $df=39$), although the main sources of *E. coli* (avian, deer/elk, canine, rodent, bovine, and bear) tended to be similar between watersheds. The dominant sources of *E. coli* changed from 2003 (avian, deer/elk, and canine) to 2004 (avian, bovine, and rodent). It is important to look at the results of more than 1 year of source tracking data to get a better picture of the dominant sources within a watershed. Overall, wildlife was the largest contributor of *E. coli* to the watersheds in both 2003 (> 84%) and 2004 (>73%).

ANNOTATION:

This BST study corroborates many others that birds rank among the largest contributors to instream FIB. It supports an important assumption at the front of the bird bridge bacteria report: that birds have the potential to contribute significantly to instream bacteria concentrations.

Muirhead, R. W., R. J. Davies-Colley, A. M. Donnison, and J. W. Nagels. 2004. Faecal bacteria yields in artificial flood events: quantifying in-stream stores. *Water Research* 38: 1215-1224.

ABSTRACT:

Stream sediments have been recognized as an in-channel store of faecal contamination that can be mobilized during floods or other sediment-disturbing events. We studied this store of faecal contamination by creating artificial floods during dry weather when, in the absence of overland flow from the catchment, the only source of faecal bacteria was stores within the channel. Artificial floods, created by releasing water from a supply reservoir, increased the *E. coli* concentration in the water column by two orders of magnitude, from a background level of 102 cfu per 100 ml to over 104 cfu per 100 ml. The bacterial peak concentrations and yields declined systematically through a triplicate flood series. The size of the total in-channel store, calculated as the sum of yields of an infinite series of artificial floods, was approximately 108 cfu m⁻² of streambed area. Direct measurements of sediment *E. coli* found few sites (only those associated with cattle crossings) with areal concentrations as high as 108 cfu m⁻², consistent with flood yields. Concentrations of *E. coli* in the biofilms on exposed rocks were orders of magnitude lower, indicating that exposed rocks were not a source of *E. coli* released by the artificial floods.

ANNOTATION:

The authors build on the work of McDonald *et al.* (1982) and attempt to quantify *E. coli* in sediment stores through artificial flooding on three successive days at two sites and disturbance of the benthic substrates in a New Zealand stream. Their results clearly demonstrate a flushing effect with a drop of 53-59% in *E. coli* with each successive flood. Their results point to the major impact sediment disturbance can have on *E. coli* results if care is not taken to reduce the disturbance during sampling. Disturbance of benthic sediments by aquatic organisms (e.g., carp) could kick up a significant amount of bacteria deposited from upstream sources, including non-swallow avian species.

Nayamatullah, M. M. M., R. Bashar, S. Bin-Shafique, and H. Sharif. 2011. Effect of direct droppings from bridge nesting birds on bacterial concentration of underneath surface water, pp. 1850-1857, World Environmental and Water Resources Congress 2011: Bearing Knowledge for Sustainability. ASCE.

ABSTRACT:

Concentrations of indicator bacteria, such as *E. coli*, fecal coliform, and Enterococci were measured from upstream, underneath, and downstream of a bridge over the Guadalupe River near Kerrville, TX, to evaluate the effect of direct droppings of birds residing under the bridge structures on the bacterial loading in the underneath water. The bacterial loadings in all sampling locations increased as soon as the migratory birds started nesting on the bridge. The concentrations of bacteria in water beneath the bridge were significantly higher than those in upstream water. The concentrations of bacteria in downstream water were slightly higher than those in upstream water and significantly lower than those in water underneath the bridge due to dilution and decay. The probability analysis of the concentration of bacteria in water underneath the bridge showed that the concentrations of *E. coli*, fecal coliform, and Enterococci are likely to remain within the Texas Commission on Environmental Quality specified contact recreation limit for 71, 68, and 35 percent of the time, respectively, during the nesting season.

ANNOTATION:

See note on Bashar's thesis (Bashar, 2010) upon which this publication is based.

O'Keefe, B., B. J. D'Arcy, J. Davidson, B. Barbarito, and B. Clelland. 2005. Urban diffuse sources of faecal indicators. *Water Science & Technology* 51: 183-190.

ABSTRACT:

Increasing concern about bathing water quality in Scotland has led to renewed interest in diffuse sources, as well as the already closely monitored municipal sewage effluents and combined sewer overflows (CSOs) that have been the subject of multi-million pound capital expenditure schemes for several years. Early investigations of diffuse sources focused on rural land uses. This paper is an initial effort to consider the possible significance of urban diffuse sources. A review of the potential for diffuse urban sources includes consideration of sewage pollution in surface water sewers, as well as non-human sources such as pigeon and other bird roosts, and faecal material from pets such

as dogs and cats. Portobello beach in Edinburgh is the case study selected, because of earlier work done by Scottish Water and SEPA. The Figgate Burn crosses Edinburgh to discharge onto the beach at Portobello, and pollution sources in its catchment are described. Additional information is reported from Dunfer line, where the sewer network has provided examples of three ways in which sewage pollution can occur in urban streams, and also Scottish examples of measures to control some non-human sources (e.g. SUDS).

ANNOTATION:

The authors review several case studies involving FIB in variously used waterways. They report fecal coliform values as high as 500,000 CFU/100 ml in the Figgate Burn catchment (Edinburgh, Scotland), values they associated with twice-weekly power washings of a walkway beneath a bridge occupied by pigeons. This suggests impacts on *E. coli* values by birds both from direct deposition and indirect through runoff from soiled surfaces.

Pachepsky, Y. A., and D. R. Shelton. 2011. *Escherichia coli* and fecal coliforms in freshwater and estuarine sediments. Critical Reviews in Environmental Science and Technology 41: 1067-1110.

ABSTRACT:

It has been known for some time that substantial populations of fecal coliforms and *E. coli* are harbored in freshwater bottom sediments, bank soils, and beach sands. However, the relative importance of sediments as bacterial habitats and as a source of waterborne fecal coliforms and *E. coli* has not been recognized until recently, when a large number of publications have shown that in many cases the resuspension of sediment, rather than runoff from surrounding lands, can create elevated *E. coli* concentrations in water. This review is an attempt to develop the first comprehensive single source of existing information about fecal coliforms and *E. coli* in sediments and adjacent soils and to outline the knowledge gaps and research needs. The authors summarize available information on variability and environmental correlations of *E. coli* and FC concentrations in sediments, genetic diversity of *E. coli* in sediments, survival of *E. coli* and FC in sediments, release with resuspended sediment and settling of *E. coli* and FC, modeling of sediment effects on fate and transport of *E. coli* in surface waters, and implications for monitoring and management of microbiological water quality. The demonstrated role of pathogenic *E. coli* strains in food and water quality challenges reinforces the need in better understanding ecological and hydrological factors that affect functioning of sediments as *E. coli* reservoirs.

ANNOTATION:

This literature review is a useful clearinghouse of information and references pertaining to *E. coli* in stream sediments. Notable points from the paper relevant to the bird bridge bacteria report include:

- High variability in sediment *E. coli* concentrations.
- The vast majority of *E. coli* sediment stores are found in the surface 1-cm of sediment.
- Sediment stores are much, much higher than the water column.

- There is no correlation between sediment and water column bacteria in short timescale experiments but according to Shelton (2008) and Erkenbrecher (1981), water and sediment *E. coli* patterns were correlated at annual and seasonal time scales.
- Proximity to wildlife, including birds, could increase sediment bacteria. They cite Hussong *et al.* (1979) and Niewolak (1989).
- Shoreline *E. coli* is a substantial source as indicated by DNA fingerprinting showing strong similarity between instream and beach Ecoli genetics. Since a large proportion of swallow feces can be seen on the shoreline during their season at the bridges, their contributions to instream bacteria may be felt most strongly during rainfall runoff events that not only wash in shoreline stores but entrain sediment-bound bacteria.
- Inactivation rates are an order of magnitude lower in sediment than in the water column. Pachepsky and Shelton do not address sunlight as an inactivator but see Sinton *et al.* (2007) whose work suggests that higher inactivation rates in the water column could be the result of increased exposure to sunlight in addition to other factors.

Palmer, M. D. 1983. Fecal coliform loadings from birds on bridges. Canadian Journal of Civil Engineering 10: 241-247.

ABSTRACT:

The fecal coliform loadings to the Rideau River from bird populations resident on bridges were measured for dry weather, summer, low river flows (8.5–17 m³ s⁻¹). The Rideau River has typical velocities to 0.216 km h⁻¹. The loadings were estimated by an intensive river sampling program over 4 weeks at river cross sections upstream and downstream of a bridge with birds on the bridge and without birds on the bridge. The birds were temporarily displaced from the bridge with bird nets. Statistical testing of the river cross section geometric means showed the birds do have a significant effect on river fecal coliform levels. From bird census, river water sampling data, and river flow data, a pigeon was estimated to generate on the average a daily loading of between 0.88 and 1.3 x 10¹⁰ fecal coliform organisms with a standard deviation of approximately 0.39 x 10¹⁰. An independent check on the validity of the estimated loadings from resident bird populations at another bridge produced results within 40% of the measured values.

ANNOTATION:

The study examined fecal coliform loadings above and below a bridge in Ottawa, Canada, inhabited mostly by pigeons. They sampled transects daily for one month in July - August and used netting to exclude birds for several days to create a control for comparisons of birded and non-birded samples. Significant differences in fecal coliform loading were found between upstream and downstream samples and that the bird defecation at the bridge accounted for 17 - 35% of the total loadings during dry weather. Important differences between Palmer and the bird bridge bacteria study include: only one bridge was investigated, the control was the same bridge during the same season with birds excluded with netting, and pigeons and fecal coliform were considered rather than cliff swallows and *E. coli*.

Sejkora, P., M. J. Kirisits, and M. Barrett. 2011. Colonies of cliff swallows on highway bridges: a source of *Escherichia coli* in surface waters. *Journal of the American Water Resources Association*. doi: 10.1111/j.1752-1688.2011.00566.x.

ABSTRACT:

Animals, such as birds, are a source of fecal indicator bacteria and pathogens in the environment. Our objective was to determine whether a colony of cliff swallows nesting underneath a bridge would yield a measurable increase in fecal indicator bacteria (specifically *Escherichia coli*) in the underlying creek. When the swallows were absent, dry-weather concentrations of *E. coli* upstream and downstream of the bridge (in Austin, Texas) were below the Texas contact recreation criteria. When the swallows were present, dry-weather geometric-mean *E. coli* concentrations increased significantly from upstream (43 most probable number [CFU]/100 ml) to downstream (106 CFU/100 ml) of the bridge. One exceedance and one near-exceedance of the Texas single sample contact recreation criterion were observed during the swallows' nesting phase. When the swallows were present, the downstream *E. coli* geometric-mean concentration in storm events (875 CFU/100 ml) was significantly higher than the upstream concentration (356 CFU/100 ml), suggesting that runoff flushes swallow feces from the ground into the creek. Although the loading of *E. coli* from cliff swallows nesting under bridges can be significant (e.g., dry-weather loading of 3.1×10^8 CFU/day/ nest), the zoonotic potential of the cliff swallow must be examined to determine the risk to human health from contact recreation in waters contaminated with cliff swallow feces.

ANNOTATION:

This paper is the primary reference for the current cliff swallow study. It represents the only known paper to directly test the impact of bridge-roosting swallows on *E. coli* samples above and below a bridge. Sejkora *et al.* examined *E. coli* contributions from cliff swallows at bridges above Bull Creek in Austin, TX. They compared upstream samples to downstream samples under wet and dry conditions when swallows were present and absent. Under both dry and wet conditions there were only significant differences between upstream and downstream samples when swallows were present. A diel study also led Sejkora *et al.* to conclude that "time of day for sampling does not substantially affect the results" because downstream samples maintained significantly higher *E. coli* concentrations than the upstream samples throughout the day while flow remained constant. The authors also observed that accumulated feces on terrestrial surfaces and bridges washed into the stream during rain events likely contributed to wet weather spikes in *E. coli* concentrations that exceeded both geometric mean and single-sample criteria. In summary, cliff swallows under bridges above Bull Creek contributed significant loadings of *E. coli* to the creek, especially during nesting and fledging when bird activity around the nests was most intense.

Silva, V. L., J. R. Nicoli, T. C. Nascimento, and C. G. Diniz. 2009. Diarrheagenic *Escherichia coli* strains recovered from urban pigeons (*Columba livia*) in Brazil and their antimicrobial susceptibility patterns. *Current Microbiology* 59: 302-308.

ABSTRACT:

Urban pigeons (*Columba livia*) come into close contact with humans and animals, and may contribute to the spread of infectious agents. These may include human pathogens

such as diarrheagenic *Escherichia coli* strains, which are able to survive in pigeon feces, thus creating potential for human exposure and infection. Our objectives were to determine the occurrence of diarrheagenic *E. coli* strains in fresh feces from urban pigeons and their drug susceptibility patterns. *E. coli* strains were isolated from 100 fresh feces samples and presumptive phenotypic species identification was carried out, confirmed by amplification of specific 16S ribosomal RNA encoding DNA. Multiplex PCR was performed to characterize pathogenic strains. Drug susceptibility patterns were determined by the agar dilution method. Enteroinvasive *E. coli*, Shiga toxin-producing *E. coli*, enteropathogenic *E. coli*, and enterotoxigenic *E. coli* were detected at an overall rate of 12.1%. Among the isolated *E. coli* strains, 62.1% were susceptible to all tested drugs, whereas 37.9% were resistant to at least one of the antimicrobials tested. Amikacin was the less effective drug (36.8% resistance), followed by ampicillin (7.8%). No resistance was detected to gentamicin, ceftriaxone, and ceftazidime and almost all the isolates were susceptible to ampicillin-sulbactam (98.4%), levofloxacin (97.8%), and trimethoprim-sulfamethoxazole (96.1%). Since these pigeons may harbor multidrug-resistant pathogens, their presence in an urban environment could be an important component of infection spread, with impact on public health.

ANNOTATION:

This paper connects *E. coli* from pigeon feces to human diarrheagenic illnesses. It serves a small but not insignificant role in the opening arguments in the present study because it demonstrates the real impact on human health that bird feces in freshwater can exert.

Sinton, L., C. Hall, and R. Braithwaite. 2007. Sunlight inactivation of *Campylobacter jejuni* and *Salmonella enterica*, compared with *Escherichia coli*, in seawater and river water. *Journal of Water and Health* 05: 357-365.

ABSTRACT:

The inactivation of *Campylobacter jejuni* and *Salmonella enterica*, compared with *Escherichia coli*, was determined in 100 l chambers of seawater and river water located at an outdoor site. The chambers (paired with dark controls) were seeded with waste stabilization pond effluent and laboratory-cultured pathogens, and exposed to sunlight in summer and winter experiments. All sunlight inactivation (k_s) rates, as a function of cumulative global solar radiation (insolation), were far higher than the corresponding dark (k_D) rates, with a ranking (and average k_s rates for seawater and river water, respectively) of: *C. jejuni* (3.23; 2.34) > *S. enterica* (0.51; 0.37) > *E. coli* (0.34; 0.26). All the T_{90} (time to 90% inactivation) values were higher in winter than in summer, but there was far greater similarity between the summer and winter S_{90} (insolation needed for 90% inactivation) values. The rapid inactivation of *C. jejuni* was attributed to a high susceptibility to photooxidative damage. The results suggest that, in sunlight-exposed waters, *E. coli* will be a more conservative indicator for *C. jejuni* than for *S. enterica*, and *C. jejuni* transmission as a pathogenic agent is less likely than for *S. enterica*.

ANNOTATION:

The authors define T_{90} and S_{90} values as the time taken to achieve a 90% reduction in CFUs (T_{90}) and the insolation needed to achieve a 90% reduction in CFUs (S_{90}). They then present experimentally derived values of these parameters for *E. coli* in stream water during winter (17.3 h) and summer (3.85 h). The summer values might indicate

that the time-of-day decay apparent in some of the samples in the bridge study is attributable to sunlight radiation which kicks in around the time of our arrival at the bridge and increases during the 2 hours of sampling. Since cliff swallows tend to forage during late morning, their departure from the bridge coincides with several hours of sunlight exposure and these factors may combine to produce very different *E. coli* results between sunrise and high noon.

Standridge, J. H., J. J. Delfino, L. B. Kleppe, and R. Butler. 1979. Effect of waterfowl (*Anas platyrhynchos*) on indicator bacteria populations in a recreational lake Madison, Wisconsin. *Applied and Environmental Microbiology* 38: 547-550.

ABSTRACT:

A public swimming beach in Madison, Wis., experienced intermittent high fecal coliform counts during the late summer and early fall of 1978. Public health officials closed the beach on a number of occasions. A public health survey identified a combination of waterfowl wastes and meteorological events as the explanation for the high bacteria counts. Fecal coliform bacteria were deposited by mallard ducks and multiplied in the beach sands. The bacteria were subsequently transported into the lake and resulted in high fecal coliform counts in the swimming area.

ANNOTATION:

Standridge *et al.* demonstrate that FC associated with mallard feces can survive and even multiply in beach sands and be washed into a water body through rain events or physical sediment disturbance. The implications of this research are similar to Hussong *et al.* (1979) in as much as the results support the supposition that swallow impacts around bridges can be captured in water samples for some days after the birds have migrated away.

Stephenson, G. R., and R. C. Rychert. 1982. Bottom sediment: a reservoir of *Escherichia coli* in rangeland streams. *Journal of Range Management* 35: 119-123.

ABSTRACT:

Escherichia coli concentrations of bottom sediment and overlying water were determined from a variety of streams in southwestern Idaho by a one-step most probable number technique. Results show *E. coli* concentrations of bottom sediments to be from 2 to 760 times greater than from the overlying water. *E. coli* concentrations of bottom sediment were found to be resuspended following disturbance simulation and a rainstorm event, contributing to pollution of the overlying waters. It is, therefore, suggested that microbial analysis of bottom sediments be considered a part of water-quality evaluations for rangeland streams.

ANNOTATION:

This study presents ratios of sediment to water column *E. coli* that range 2 - 760 and corroborates Muirhead *et al.* (2004) showing an initial spike in *E. coli* in the opening minutes of flood pulses with rapidly diminishing values thereafter suggesting a flushing effect.

Traister, E., and S. C. Anisfeld. 2006. Variability of indicator bacteria at different time scales in the Upper Hoosic River watershed. *Environmental Science Technology* 40:4990-4995.

ABSTRACT:

Accurately evaluating whether a water body is meeting water quality criteria for indicator bacteria requires an understanding of the spatial and temporal variability in concentrations of these indicators. We have collected data on concentrations of *Escherichia coli* at 12 sites within the upper Hoosic River Basin, spanning a range of land uses and levels of development. Sampling was conducted with the goal of assessing the variation in *E. coli* levels over different time scales: seasonal, storm-related, and diurnal. General linear models were constructed to describe the factors contributing to *E. coli* concentrations at a given location and time. We found that bacterial levels were higher in more developed watersheds; in summer rather than winter; in storms rather than baseflow; and in the early morning rather than afternoon. Seasonal and storm sampling captured different portions of the range of *E. coli* concentrations, but the levels of variability at these two scales were similar. Diurnal sampling produced concentrations intermediate between seasonal and storm sampling. Compared to a pristine stream, a more urbanized stream exhibited greater diurnal variability, but less variation from baseflow to stormflow. We recommend collecting both seasonal and storm data, but not necessarily diurnal data, in assessment of stream bacterial quality.

ANNOTATION:

The authors found small but significant diurnal variation in instream *E. coli* from a watershed in Massachusetts with mixed landuse. The results are contrary to Meays *et al.* (2006b) and Sejkora *et al.* (2011) who found no significant diurnal variation in *E. coli*. This paper is cited in discussions of the effect of time-of-day on samples collected only 60 minutes apart.

Withers, P. C. 1977. Energetic aspects of reproduction by the cliff swallow. *The Auk* 94: 718-725.

ABSTRACT:

Energy budgets were constructed for the Cliff Swallow during the nest building, incubation, and nestling periods using time budgets and aerodynamic theory. Mean dally energy expenditures during these periods were 1.55, 1.23, and 1.28 watts respectively, with required food harvest rates of at least 3.95, 4.42, and 4.07 watts. The cost of constructing an average size nest (600 g) was approximately 122 kilojoules expended over about 7 days, but the multiple use of the nest makes the cost per brood considerably more than this. The ecological advantages accruing from the nest apparently are related primarily to physical protection from predators and reduction of intraspecific aggression, rather than microclimatic conditions established within the nest.

ANNOTATION:

This study provides estimates of hours per day spent by cliff swallows in California foraging, constructing the nest, and sitting in the nest. This information is valuable for determining what time of year the birds can be expected to deposit the largest amount of fecal material under the nesting area.

Appendix B

Full *E. coli* Results

Results of bacteria samples collected in 2012 – 2013 at Stations 16404, 21186, and 20018.

Appendix B-1.

Results of *E. coli* samples from Station 16404, 2012 – 2013

16404 Survey 1 19-Apr-2012				16404 Survey 2 30-Apr-2012				16404 Survey 3 22-May-2012			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	10:57	29	1	Bridge	6:30	102	1	Bridge	7:57	43
		10:58	29			6:31	99			7:58	47
		10:59	30			6:32	180			7:59	51
		11:00	37			6:33	85			8:00	41
		11:01	28			6:34	94			8:01	32
	Upstream	11:05	24		Upstream	6:38	82		Upstream	8:06	48
		11:06	13			6:39	83			8:07	48
		11:07	20			6:40	86			8:08	49
		11:08	12			6:41	74			8:09	36
		11:09	15			6:42	72			8:10	52
	Downstream	11:13	21		Downstream	6:48	220		Downstream	8:18	170
		11:14	38			6:49	142			8:19	179
		11:16	36			6:50	123			8:20	185
		11:17	31			6:51	200			8:21	174
		11:18	18			6:52	250			8:22	200
2	Bridge	11:32	29	2	Bridge	7:04	74	2	Bridge	8:28	37
		11:33	30			7:05	73			8:29	44
		11:34	27			7:06	72			8:30	33
		11:35	29			7:07	73			8:31	46
		11:36	30			7:08	350			8:32	34
	Upstream	11:37	10		Upstream	7:09	64		Upstream	8:37	41
		11:38	12			7:10	51			8:38	47
		11:39	19			7:11	55			8:39	*
		11:40	19			7:12	48			8:40	*
		11:41	23			7:13	59			8:41	*
	Downstream	11:46	25		Downstream	7:17	131		Downstream	8:53	*
		11:47	37			7:18	112			8:54	*
		11:48	36			7:19	114			8:55	*
		11:49	29			7:20	96			8:56	*
		11:50	28			7:21	102			8:57	*
3	Bridge	12:09	21	3	Bridge	7:58	54	3	Bridge	9:19	*
		12:10	28			7:59	33			9:20	*
		12:11	21			8:00	33			9:21	*
		12:12	24			8:01	43			9:22	*
		12:13	18			8:02	50			9:23	*
	Upstream	12:14	21		Upstream	8:04	31		Upstream	9:27	*
		12:15	16			8:05	32			9:28	*
		12:16	14			8:06	29			9:29	*
		12:17	9			8:07	25			9:30	*
		12:18	10			8:08	25			9:31	*
	Downstream	12:22	33		Downstream	8:11	85		Downstream	9:40	*
		12:23	30			8:12	79			9:41	*
		12:24	40			8:13	62			9:42	*
		12:25	28			8:14	72			9:43	*
		12:26	34			8:15	80			9:44	*

**Samples discarded because of lab processing error*

Appendix B-1. Cont.

16404 Survey 4 23-May-2012				16404 Survey 5 08-Apr-2013				16404 Survey 6 23-Apr-2013			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	7:05	54	1	Bridge	9:32	78	1	Bridge	10:17	129
		7:06	59			9:33	79			10:18	200
		7:07	66			9:34	92.8			10:19	117
		7:08	70			9:35	94.5			10:20	115
		7:09	72			9:36	92.7			10:21	155
	Upstream	7:12	46		Upstream	9:38	18		Upstream	10:25	15
		7:13	34			9:39	18			10:26	14
		7:14	41			9:40	12			10:27	18
		7:15	42			9:41	9			10:28	5
		7:16	32			9:42	13.6			10:29	15.5
	Downstream	7:21	210		Downstream	9:46	124		Downstream	10:35	320
		7:22	540			9:47	145			10:36	370
		7:23	350			9:48	152			10:37	350
		7:24	250			9:49	123			10:38	340
		7:25	240			9:50	116			10:39	290
2	Bridge	7:37	44	2	Bridge	10:14	80.2	2	Bridge	11:07	111
		7:38	59			10:15	99.1			11:08	110
		7:39	42			10:16	71			11:09	105
		7:40	41			10:17	75			11:10	95.1
		7:41	46			10:18	83.8			11:11	220
	Upstream	7:44	38		Upstream	10:21	15.5		Upstream	11:16	18.2
		7:45	34			10:22	11.8			11:17	7
		7:46	34			10:23	7.3			11:18	9
		7:47	27			10:24	15			11:19	17.3
		7:48	27			10:25	18			11:20	11
	Downstream	7:53	280		Downstream	10:29	100		Downstream	11:25	330
		7:54	200			10:30	120			11:26	250
		7:55	260			10:31	120			11:27	270
		7:56	240			10:32	115			11:28	250
		7:57	195			10:33	110			11:29	350
3	Bridge	8:25	51	3	Bridge	10:56	79	3	Bridge	12:01	340
		8:26	62			10:57	67			12:02	410
		8:27	54			10:58	62			12:03	220
		8:28	49			10:59	64			12:04	210
		8:29	56			11:00	77			12:05	123
	Upstream	8:31	39		Upstream	11:02	10.9		Upstream	12:10	7.3
		8:32	39			11:03	10.9			12:11	7.9
		8:33	30			11:04	7.3			12:12	12
		8:34	35			11:05	13			12:13	10
		8:35	40			11:06	11			12:14	8
	Downstream	8:40	191		Downstream	11:13	200		Downstream	12:20	200
		8:41	260			11:14	100			12:21	310
		8:42	220			11:15	124			12:22	290
		8:43	260			11:16	132			12:23	230
		8:44	150			11:17	230			12:24	126

Appendix B-1.

Cont.

16404 Survey 7 06-May-2013				16404 Survey 8 30-May-2013			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	9:52	150	1	Bridge	10:41	210
		9:53	130			10:42	200
		9:54	210			10:43	230
		9:55	120			10:44	150
		9:56	120			10:45	190
	Upstream	9:58	8		Upstream	10:50	220
		9:59	5			10:51	17
		10:00	7			10:52	19
		10:01	9			10:53	13
		10:02	8			10:54	14
	Downstream	10:06	210		Downstream	11:00	240
		10:07	180			11:01	220
		10:08	170			11:02	200
		10:09	180			11:03	260
10:10		270	11:04	170			
2	Bridge	10:33	170	2	Bridge	11:25	230
		10:34	230			11:26	190
		10:35	200			11:27	200
		10:36	200			11:28	150
		10:37	300			11:29	190
	Upstream	10:41	9		Upstream	11:35	16
		10:42	11			11:36	15.4
		10:43	18			11:37	18
		10:44	5			11:38	10
		10:45	7			11:39	11
	Downstream	10:49	220		Downstream	11:45	280
		10:50	160			11:46	240
		10:51	160			11:47	270
		10:52	190			11:48	220
10:53		270	11:49	240			
3	Bridge	11:16	210	3	Bridge	12:16	160
		11:17	220			12:17	100
		11:18	120			12:18	100
		11:19	150			12:19	130
		11:20	130			12:20	154
	Upstream	11:24	7		Upstream	12:30	12.7
		11:25	3			12:31	3.6
		11:26	9			12:32	10.9
		11:27	7			12:33	16
		11:28	4			12:34	15
	Downstream	11:33	100		Downstream	12:41	173
		11:34	170			12:42	200
		11:35	130			12:43	280
		11:36	120			12:44	300
11:37		90	12:45	250			

Appendix B-2.

Results of *E. coli* samples from Station 21186, 2012 – 2013

21186 Survey 1 26-Apr-2012				21186 Survey 2 01-May-2012				21186 Survey 3 24-May-2012			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	7:00	18	1	Bridge	7:43	16	1	Bridge	7:47	68
		7:01	27			7:44	26			7:48	146
		7:02	25			7:45	17			7:49	157
		7:03	29			7:46	70			7:50	94
		7:04	25			7:47	13			7:51	85
	Upstream	7:05	42		Upstream	7:52	10		Upstream	7:54	29
		7:06	29			7:53	16			7:55	40
		7:07	28			7:54	23			7:56	36
		7:08	26			7:55	16			7:57	37
		7:09	29			7:56	16			7:58	34
	Downstream	7:10	30		Downstream	8:02	31		Downstream	8:03	65
		7:11	23			8:03	33			8:04	85
		7:12	35			8:04	15			8:05	100
		7:13	29			8:05	15			8:06	85
7:14		19	8:06	24		8:07	63				
2	Bridge	7:35	25	2	Bridge	8:14	17	2	Bridge	8:19	82
		7:36	32			8:15	20			8:20	81
		7:37	18			8:16	18			8:21	155
		7:38	45			8:17	14			8:22	115
		7:39	37			8:18	10			8:23	64
	Upstream	7:40	23		Upstream	8:23	14		Upstream	8:27	24
		7:41	38			8:24	13			8:28	29
		7:42	27			8:25	16			8:29	26
		7:43	6			8:26	16			8:30	31
		7:44	30			8:27	21			8:31	25
	Downstream	7:45	23		Downstream	8:33	19		Downstream	8:39	95
		7:46	34			8:34	39			8:40	66
		7:47	27			8:35	43			8:41	107
		7:48	27			8:36	23			8:42	91
7:49		47	8:37	39		8:43	120				
3	Bridge	8:15	22	3	Bridge	8:56	21	3	Bridge	8:57	69
		8:16	25			8:57	17			8:58	68
		8:17	28			8:58	24			8:59	80
		8:18	26			8:59	24			9:00	161
		8:19	25			9:00	24			9:01	185
	Upstream	8:20	33		Upstream	9:04	15		Upstream	9:14	27
		8:21	23			9:05	17			9:15	21
		8:22	27			9:06	17			9:16	29
		8:23	38			9:07	35			9:17	34
		8:24	26			9:08	24			9:18	29
	Downstream	8:25	21		Downstream	9:14	35		Downstream	9:25	99
		8:26	45			9:15	21			9:26	50
		8:27	38			9:16	25			9:27	81
		8:28	31			9:17	29			9:28	71
8:29		20	9:18	18		9:29	75				

Appendix B-2.

Cont.

21186 Survey 4 29-May-2012				21186 Survey 5 09-Apr-2013				21186 Survey 6 22-Apr-2013			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	9:45	610	1	Bridge	8:16	10	1	Bridge	10:48	9
		9:46	230			8:17	14			10:49	6
		9:47	200			8:18	14			10:50	5
		9:48	570			8:19	8.2			10:51	8.2
		9:49	860			8:20	9			10:52	5
	Upstream	9:50	31		Upstream	8:25	21		Upstream	10:57	1
		9:51	33			8:26	10			10:58	9.1
		9:52	43			8:27	21			10:59	9
		9:53	43			8:28	12.7			11:00	4.5
		9:54	28			8:29	18			11:01	11
	Downstream	10:00	94		Downstream	8:35	17		Downstream	11:07	7
		10:01	78			8:36	13.6			11:08	12.7
		10:02	126			8:37	12.7			11:09	11.8
		10:03	220			8:38	13.6			11:10	12
10:04		270	8:39	9.1		11:11	16				
2	Bridge	10:16	390	2	Bridge	9:00	15.5	2	Bridge	11:32	6
		10:17	490			9:01	21			11:33	4.5
		10:18	330			9:02	11			11:34	3
		10:19	350			9:03	17.1			11:35	5
		10:20	250			9:04	10.9			11:36	7
	Upstream	10:21	37		Upstream	9:09	26		Upstream	11:39	8.2
		10:22	35			9:10	23			11:40	6
		10:23	43			9:11	21.6			11:41	2
		10:24	30			9:12	17			11:42	3
		10:25	30			9:13	12.7			11:43	4.5
	Downstream	10:31	116		Downstream	9:20	14		Downstream	11:48	9
		10:32	113			9:21	19.1			11:49	7
		10:33	117			9:22	21			11:50	9
		10:34	145			9:23	21			11:51	7.3
10:35		154	9:24	14.5		11:52	8.2				
3	Bridge	10:47	750	3	Bridge	9:45	12.7	3	Bridge	12:22	2
		10:48	1000			9:46	6			12:23	4
		10:49	470			9:47	10.9			12:24	2
		10:50	610			9:48	9.1			12:25	5.5
		10:51	200			9:49	10			12:26	10
	Upstream	10:55	25		Upstream	9:52	12.7		Upstream	12:30	5
		10:56	49			9:53	10			12:31	1
		10:57	40			9:54	19.1			12:32	1
		10:58	42			9:55	18			12:33	2
		10:59	34			9:56	10.9			12:34	3
	Downstream	11:06	151		Downstream	10:05	11.8		Downstream	12:38	14
		11:07	112			10:06	6			12:39	9
		11:08	103			10:07	8.2			12:40	14
		11:09	135			10:08	10.9			12:41	4
11:10		115	10:09	12.7		12:42	12				

Appendix B-2.

Cont.

21186 Survey 7 07-May-2013				21186 Survey 8 31-May-2013			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	9:40	15	1	Bridge	9:35	45
		9:41	16.4			9:36	45
		9:42	16			9:37	41
		9:43	3			9:38	41
		9:44	15.5			9:39	34
	Upstream	9:59	11		Upstream	9:40	24
		10:00	7.3			9:41	27
		10:01	10			9:42	19
		10:02	6			9:43	33
		10:03	6.4			9:44	30
	Downstream	10:08	22		Downstream	9:50	280
		10:09	18			9:51	120
10:10		10.9	9:52	170			
10:11		14.5	9:53	130			
10:12		9	9:54	160			
2	Bridge	10:33	7	2	Bridge	10:15	42
		10:34	6			10:16	26
		10:35	10			10:17	45
		10:36	9.1			10:18	44
		10:37	11.8			10:19	35
	Upstream	10:38	3		Upstream	10:20	17
		10:39	5			10:21	34
		10:40	9			10:22	17
		10:41	8			10:23	25
		10:42	3			10:24	30
	Downstream	10:48	15		Downstream	10:37	370
		10:49	10.9			10:38	210
10:50		14.5	10:39	260			
10:51		14.5	10:40	270			
10:52		15.5	10:41	260			
3	Bridge	11:31	3.6	3	Bridge	10:55	47
		11:32	5			10:56	34
		11:33	3			10:57	26
		11:34	4			10:58	89
		11:35	3.6			10:59	45
	Upstream	11:36	3		Upstream	11:00	36
		11:37	10			11:01	35
		11:38	6			11:02	39
		11:39	7			11:03	32
		11:40	3.6			11:04	39
	Downstream	11:46	8.2		Downstream	11:10	220
		11:47	16			11:11	200
11:48		8	11:12	140			
11:49		10	11:13	120			
11:50		10	11:14	120			

Appendix B-3.
(2013)

Results of *E. coli* samples from control Stations 20018 (2012) and 16404

20018 Survey 1 27-Apr-2012				20018 Survey 2 03-May-2012					
Rep	Location	Time (cst)	<i>E. coli</i> (MPN / 100mL)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN / 100mL)		
1	Bridge	6:56	105	1	Bridge	8:38	117		
		6:58	101			8:39	91		
		7:00	83			8:40	86		
		7:02	102			8:41	102		
		7:04	105			8:42	110		
	Upstream	7:08	150		Upstream	8:49	92		
		7:09	138			8:50	101		
		7:10	220			8:51	89		
		7:11	139			8:52	85		
		7:12	132			8:53	90		
	Downstream	7:13	83		Downstream	8:55	98		
		7:14	95			8:56	99		
		7:15	96			8:57	102		
		7:16	88			8:58	90		
		7:17	95			8:59	105		
	2	Bridge	7:28		67	2	Bridge	9:09	105
			7:29		62			9:10	76
7:30			65	9:11	78				
7:31			60	9:12	75				
7:32			65	9:13	95				
Upstream		7:34	66	Upstream	9:19		79		
		7:35	83		9:20		87		
		7:36	77		9:21		92		
		7:37	84		9:22		87		
		7:38	71		9:23		75		
Downstream		7:48	57	Downstream	9:26		81		
		7:49	41		9:27		77		
		7:50	55		9:28		72		
		7:51	62			9:29	77		
		7:52	62			9:30	72		
3	Bridge	8:14	*	3	Bridge	9:47	71		
		8:15	*			9:48	74		
		8:16	*			9:49	76		
		8:17	*			9:50	79		
		8:18	*			9:51	85		
	Upstream	8:26	*		Upstream	9:59	74		
		8:27	*			10:00	68		
		8:28	51			10:01	72		
		8:29	41			10:02	72		
		8:30	44			10:03	62		
	Downstream	8:34	48		Downstream	10:06	69		
		8:35	36			10:07	43		
		8:36	40			10:08	58		
		8:41	52			10:09	75		
		8:38	47			10:10	79		

*Samples discarded because of lab processing error

Appendix B-3.

Cont.

20018 Survey 3 30-May-2012				20018 Survey 4 25-Jun-2012			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	8:11	2000	1	Bridge	8:34	47
		8:12	2000			8:35	29
		8:13	2000			8:36	49
		8:14	680			8:37	43
		8:15	131			8:38	51
	Upstream	8:22	57		Upstream	8:45	36
		8:23	51			8:46	30
		8:24	54			8:47	34
		8:25	52			8:48	24
		8:26	70			8:49	31
	Downstream	8:30	56		Downstream	8:55	29
		8:31	66			8:56	29
		8:32	52			8:57	34
		8:33	65			8:58	33
		8:34	55			8:59	38
2	Bridge	8:42	200	2	Bridge	9:05	34
		8:43	2000			9:06	41
		8:44	2000			9:07	34
		8:45	2000			9:08	34
		8:46	2000			9:09	35
	Upstream	8:56	51		Upstream	9:18	20
		8:57	58			9:19	23
		8:58	57			9:20	22
		8:59	57			9:21	30
		9:00	78			9:22	25
	Downstream	9:05	50		Downstream	9:28	37
		9:06	44			9:29	31
		9:07	39			9:30	32
		9:08	43			9:31	37
		9:09	57			9:32	29
3	Bridge	9:32	840	3	Bridge	9:45	36
		9:33	1020			9:46	30
		9:34	2000			9:47	27
		9:35	840			9:48	23
		9:36	2000			9:49	32
	Upstream	9:43	61		Upstream	9:55	16.4
		9:44	64			9:56	18.2
		9:45	56			9:57	16.4
		9:46	64			9:58	21
		9:47	68			9:59	16
	Downstream	9:50	44		Downstream	10:05	20
		9:51	53			10:06	26
		9:52	51			10:07	17
		9:53	57			10:08	29
		9:54	59			10:09	26

Appendix B-3. Cont.

16404 Temporal Control 1 24-Jan-2013				16404 Temporal Control 2 06-Feb-2013				16404 Temporal Control 3 27-Feb-2013			
Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)	Rep	Location	Time (cst)	<i>E. coli</i> (MPN/100 ml)
1	Bridge	9:15	36	1	Bridge	7:47	10	1	Bridge	8:25	11
		9:16	46			7:48	12.7			8:26	10
		9:17	41			7:49	10			8:27	13.5
		9:18	36			7:50	12			8:28	17.3
		9:19	32			7:51	15.5			8:29	13
	Upstream	9:26	27		Upstream	7:53	14.5		Upstream	8:30	12
		9:27	30			7:54	10.9			8:31	14.5
		9:28	22			7:55	6			8:32	10
		9:29	30			7:56	12.7			8:33	13.6
		9:30	27			7:57	14.5			8:34	10
	Downstream	9:36	56		Downstream	8:01	30		Downstream	8:40	14.5
		9:37	42			8:02	32			8:41	12
		9:38	38			8:03	29			8:42	19.1
		9:39	46			8:04	31			8:43	14.5
		9:40	42			8:05	17.1			8:44	12.6
2	Bridge	9:48	42	2	Bridge	8:28	12	2	Bridge	9:05	11
		9:49	33			8:29	10			9:06	10
		9:50	37			8:30	8.2			9:07	15.5
		9:51	46			8:31	9.1			9:08	10
		9:52	34			8:32	10			9:09	10
	Upstream	9:57	17		Upstream	8:34	8.2		Upstream	9:13	8
		9:58	12			8:35	7			9:14	4
		9:59	24			8:36	8.2			9:15	10.9
		10:00	23			8:37	12			9:16	11
		10:01	23			8:38	9			9:17	7
	Downstream	10:07	44		Downstream	8:47	27		Downstream	9:27	11.8
		10:08	49			8:48	19.1			9:28	16.4
		10:09	37			8:49	24			9:29	17.3
		10:10	40			8:50	22			9:30	14.5
		10:11	32			8:51	30			9:31	10
3	Bridge	10:19	32	3	Bridge	9:09	15	3	Bridge	9:47	13
		10:20	35			9:10	14.5			9:48	9
		10:21	48			9:11	9			9:49	5
		10:22	41			9:12	14.5			9:50	9.1
		10:23	38			9:13	9.1			9:51	11
	Upstream	10:28	15		Upstream	9:19	5.5		Upstream	9:55	3
		10:29	15			9:20	11.8			9:56	2
		10:30	12			9:21	13.6			9:57	10
		10:31	13			9:22	12.7			9:58	8.2
		10:32	12			9:23	10.9			9:59	1
	Downstream	10:39	25		Downstream	9:29	21		Downstream	10:07	6
		10:40	35			9:30	16			10:08	10
		10:41	44			9:31	22			10:09	13.6
		10:42	31			9:32	25			10:10	9
		10:43	29			9:33	24			10:11	13

Appendix C

Full Results of Univariate Procedure on *E. coli*

Samples 2012 - 2013

Results of descriptive statistics for bacteria sampling distributions by location and rep.

Appendix C-1. Differences between sampling locations at the densely-birded bridge, Station 16404, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for bridge – upstream (BU), downstream – upstream (DU), and downstream – bridge (DB). Skewness values $> |0.5|$ are highlighted orange, values $> |1.0|$ are bold italic. Values were \log_{10} -transformed prior to testing.

Location Comparison	BU	DU	DB	BU	DU	DB	BU	DU	DB	BU	DU	DB
Survey	19-Apr-12			30-Apr-12			22-May-12			23-May-12		
<i>n</i>	15	15	15	15	15	15	7	5	5	15	15	15
Mean Difference (MPN/100 ml)	11.5	15.1	3.6	39.9	70.1	30.2	-3.7	135.0	138.8	19.1	220.5	201.4
Skewness	0.17	-0.51	0.06	2.41	-0.16	-2.12	-1.58	1.87	1.87	0.19	0.57	0.58
<i>W</i>	0.96	0.93	0.97	0.74	0.98	0.77	0.85	0.78	0.79	0.96	0.92	0.96
Pr< <i>W</i>	0.710	0.296	0.919	<0.001	0.939	0.001	0.130	0.060	0.065	0.619	0.199	0.700
<i>t</i>	6.68	6.53	1.41	3.87	13.00	2.81	-1.19	25.28	14.91	9.02	22.53	18.38
Pr ≥ <i>t</i>	<0.001	<0.001	0.181	0.002	<0.001	0.014	0.278	<0.001	<0.001	<0.001	<0.001	<0.001
<i>M</i>	7	6.5	2	7.5	7.5	5.5	-1.5	2.5	2.5	7.5	7.5	7.5
Pr ≥ <i>M</i>	<0.001	<0.001	0.424	<0.001	<0.001	0.007	0.453	0.063	0.063	<0.001	<0.001	<0.001
<i>S</i>	52.5	59	22	60	60	42	-6	7.5	7.5	60	60	60
Pr ≥ <i>S</i>	<0.001	<0.001	0.178	<0.001	<0.001	0.015	0.375	0.063	0.063	<0.001	<0.001	<0.001
Survey	8-Apr-13			23-Apr-13			6-May-13			30-May-13		
<i>n</i>	15	15	15	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	66.9	121.3	54.4	165.7	273.4	107.7	169.5	166.9	-2.7	144.8	208.8	63.9
Skewness	0.12	0.29	0.87	0.36	0.93	-0.79	0.52	-0.12	0.38	-2.81	-2.27	0.64
<i>W</i>	0.94	0.94	0.92	0.95	0.95	0.90	0.97	0.98	0.96	0.66	0.73	0.95
Pr< <i>W</i>	0.329	0.389	0.187	0.568	0.524	0.090	0.814	0.943	0.736	<0.001	<0.001	0.598
<i>t</i>	24.54	22.78	7.24	15.52	31.40	4.04	23.89	24.87	-0.22	12.63	12.71	3.86
Pr ≥ <i>t</i>	<0.001	<0.001	<0.001	<0.001	<0.001	0.001	<0.001	<0.001	0.832	<0.001	<0.001	0.002
<i>M</i>	7.5	7.5	7.5	7.5	7.5	5.5	7.5	7.5	-1.5	6.5	7.5	5.5
Pr ≥ <i>M</i>	<0.001	<0.001	<0.001	<0.001	<0.001	0.007	<0.001	<0.001	0.607	<0.001	<0.001	0.007
<i>S</i>	60	60	60	60	60	50	60	60	-3.5	59	60	52
Pr ≥ <i>S</i>	<0.001	<0.001	<0.001	<0.001	<0.001	0.003	<0.001	<0.001	0.858	<0.001	<0.001	0.001

Appendix C-2. Differences between sampling locations at the densely-birded bridge, Station 21186, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for bridge – upstream (BU), downstream – upstream (DU), and downstream – bridge (DB). Skewness values $> |0.5|$ are highlighted orange, values $> |1.0|$ are bold italic. Values were \log_{10} -transformed prior to testing.

Location Comparison	BU	DU	DB	BU	DU	DB	BU	DU	DB	BU	DU	DB
Survey	26-Apr-12			1-May-12			24-May-12			29-May-12		
<i>n</i>	15	15	15	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	-1.2	1.6	2.8	4.1	9.3	5.2	77.3	53.5	-23.8	451.1	100.4	-351.0
Skewness	2.56	1.64	-0.15	0.94	-0.14	-1.33	0.80	0.68	0.26	0.12	1.10	-0.22
W	0.70	0.86	0.98	0.93	0.95	0.89	0.89	0.93	0.98	0.97	0.89	0.97
Pr< W	<0.001	0.024	0.978	0.259	0.604	0.075	0.076	0.231	0.970	0.889	0.070	0.782
t	-0.02	0.65	1.09	1.06	3.16	1.60	14.63	13.81	-1.99	17.28	12.47	-9.23
Pr ≥ t	0.985	0.529	0.294	0.308	0.007	0.133	<0.001	<0.001	0.067	<0.001	<0.001	<0.001
M	-1.5	-0.5	1	1.5	3.5	4.5	7.5	7.5	-3.5	7.5	7.5	-7.5
Pr ≥ M	0.607	1.000	0.791	0.581	0.118	0.035	<0.001	<0.001	0.118	<0.001	<0.001	<0.001
S	-19	2.5	17.5	14.5	44	35	60	60	-33	60	60	-60
Pr ≥ S	0.296	0.893	0.296	0.340	0.010	0.048	<0.001	<0.001	0.064	<0.001	<0.001	<0.001
Survey	9-Apr-13			22-Apr-13			7-May-13			31-May-13		
<i>n</i>	15	15	15	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	-5.0	-3.2	1.7	0.8	5.5	4.7	2.0	6.6	4.5	13.5	172.9	159.4
Skewness	0.60	-0.21	0.58	0.39	0.79	0.65	0.08	0.78	-0.41	-0.10	0.56	-1.10
W	0.91	0.94	0.93	0.96	0.92	0.93	0.93	0.93	0.97	0.96	0.95	0.92
Pr< W	0.153	0.363	0.248	0.639	0.194	0.283	0.280	0.255	0.882	0.665	0.570	0.163
t	-4.12	-2.32	1.82	1.49	4.98	3.81	1.07	6.36	3.87	3.27	12.86	11.85
Pr ≥ t	0.001	0.036	0.090	0.158	<0.001	0.002	0.303	<0.001	0.002	0.006	<0.001	<0.001
M	-5	-2.5	1	1.5	7.5	5.5	3	7.5	5.5	4.5	7.5	7.5
Pr ≥ M	0.013	0.302	0.791	0.607	<0.001	0.007	0.180	<0.001	0.007	0.035	<0.001	<0.001
S	-47	-33	22.5	25	60	53	18.5	60	48	44	60	60
Pr ≥ S	0.002	0.066	0.173	0.169	<0.001	0.001	0.267	<0.001	0.004	0.010	<0.001	<0.001

Appendix C-3. Differences between control sampling locations at the non-birded bridge, Station 20018 (2012) and Station 16404 (2013) when migratory cliff swallows were not present. Significant differences ($p = 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for bridge – upstream (BU), downstream – upstream (DU), and downstream – bridge (DB). Skewness values $> |0.5|$ are highlighted orange, values $> |1.0|$ are bold italic. Values were \log_{10} -transformed prior to testing.

Location Comparison	BU	DU	DB	BU	DU	DB	BU	DU	DB	BU	DU	DB
Survey	27-Apr-12			3-May-12			30-May-12			25-Jun-12		
<i>n</i>	10	13	10	15	15	15	15	15	15	15	15	15
Mean Difference (MPN/100 ml)	-26.3	-42.6	-8.1	6.3	-1.9	-8.2	1388.0	-7.1	-1395.0	12.1	5.6	-6.5
Skewness	-1.92	0.03	-0.28	-0.05	-0.77	-0.90	-1.65	0.83	1.48	-0.50	-0.25	0.15
W	0.79	0.97	0.97	0.92	0.96	0.94	0.76	0.92	0.81	0.96	0.98	0.98
Pr< W	0.011	0.856	0.920	0.215	0.738	0.430	0.001	0.192	0.006	0.702	0.981	0.975
t	-3.66	-3.95	-2.41	1.64	-0.84	-2.40	12.65	-2.66	-13.00	7.09	4.01	-3.26
Pr ≥ t	0.005	0.002	0.039	0.124	0.417	0.031	<0.001	0.019	<0.001	<0.001	0.001	0.006
M	-4	-4.5	-3	1.5	-0.5	-3.5	7.5	-5.5	-7.5	6.5	5	-4
Pr ≥ M	0.022	0.023	0.109	0.607	1.000	0.118	<0.001	0.007	<0.001	<0.001	0.013	0.057
S	-27	-40	-22	27	-9.5	-39	60	-42	-60	59	44.5	-41
Pr ≥ S	0.004	0.003	0.027	0.135	0.609	0.026	<0.001	0.015	<0.001	<0.001	0.003	0.008
Station 16404 Temporal Control												
Survey	24-Jan-13			6-Feb-13			27-Feb-13					
<i>n</i>	15	15	15	15	15	15	15	15	15			
Mean Difference (MPN/100 ml)	18.3	19.2	0.9	0.9	14.1	13.2	2.9	4.6	1.7			
Skewness	0.24	0.90	0.67	0.81	-0.14	-1.07	1.11	1.74	-0.17			
W	0.96	0.89	0.91	0.95	0.99	0.84	0.90	0.80	0.92			
Pr< W	0.633	0.075	0.140	0.514	0.998	0.012	0.083	0.004	0.173			
t	7.10	8.15	0.17	1.14	8.66	8.57	2.25	3.25	1.46			
Pr ≥ t	<0.001	<0.001	0.869	0.274	<0.001	<0.001	0.041	0.006	0.167			
M	7.5	7.5	-1.5	2	7.5	7.5	3.5	6.5	3			
Pr ≥ M	<0.001	<0.001	0.581	0.424	<0.001	<0.001	0.118	<0.001	0.180			
S	60	60	-2.5	15.5	60	60	36	56.5	29.5			
Pr ≥ S	<0.001	<0.001	0.893	0.358	<0.001	<0.001	0.041	<0.001	0.068			

Appendix C-4. Differences between sampling reps at the densely-birded bridge, Station 16404, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted, strongly significant differences ($p < 0.001$) are bold italic. Mean differences are presented for reps 1 – 2 (R12), reps 1 – 3 (R13), and reps 2 – 3 (R23). Skewness values $> |0.5|$ are highlighted orange, values $> |1.0|$ are bold italic. Values were \log_{10} -transformed prior to testing.

Event Comparison	E12	E13	E23	E12	E13	E23	E12	E13	E23	E12	E13	E23
Survey	19-Apr-12			30-Apr-12			22-May-12			23-May-12		
<i>n</i>	5	5	5	5	5	5	2	0	0	5	5	5
Mean Difference (MPN/100 ml)	-0.2	-2.8	-2.6	-24.0	-51.0	-27.0	-4.0			-7.0	-2.4	4.6
Skewness	-1.09	1.41	0.50	0.58	0.52	0.18	-1.08			-0.71	0.39	-0.07
<i>W</i>	0.87	0.87	0.92	0.85	0.83	0.97	1.00	1.00	1.00	0.90	0.90	0.99
Pr< <i>W</i>	0.284	0.263	0.514	0.194	0.139	0.894	1.000	1.000	1.000	0.389	0.401	0.991
<i>t</i>	-0.10	-1.76	-0.57	-6.25	-36.20	-10.50	-1.31			-2.81	-0.59	1.54
Pr ≥ <i>t</i>	0.926	0.153	0.602	0.003	<0.001	<0.001	0.416			0.049	0.589	0.199
<i>M</i>	-0.5	-1.5	-0.5	-2.5	-2.5	-2.5	-1			-2	-0.5	1.5
Pr ≥ <i>M</i>	1.000	0.375	1.000	0.063	0.063	0.063	0.500			0.125	1.000	0.375
<i>S</i>	-0.5	-5.5	-3.5	-7.5	-7.5	-7.5	-1.5			-5	-2.5	5.5
Pr ≥ <i>S</i>	1.000	0.188	0.438	0.063	0.063	0.063	0.500			0.125	0.625	0.188
Survey	8-Apr-13			23-Apr-13			6-May-13			30-May-13		
<i>n</i>	5	5	5	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	-0.6	-3.5	-2.9	-1.0	-4.5	-3.5	2.6	-1.4	-4.0	-42.5	-45.0	-2.4
Skewness	0.41	1.72	-0.59	1.31	2.02	-0.15	-0.06	0.44	0.10	-2.20	-0.94	-0.92
<i>W</i>	0.91	0.74	0.93	0.85	0.73	0.96	0.94	0.98	0.99	0.63	0.90	0.93
Pr< <i>W</i>	0.486	0.023	0.621	0.201	0.019	0.823	0.676	0.929	0.976	0.001	0.425	0.591
<i>t</i>	-0.28	-1.59	-2.35	-0.16	-1.27	-1.25	0.79	-1.65	-1.84	-1.33	-1.64	-0.82
Pr ≥ <i>t</i>	0.791	0.186	0.079	0.879	0.272	0.278	0.474	0.175	0.140	0.254	0.176	0.457
<i>M</i>	-0.5	-1.5	-2	-0.5	-1.5	-0.5	0.5	-1.5	-1.5	-2.5	-0.5	-0.5
Pr ≥ <i>M</i>	1.000	0.375	0.125	1.000	0.375	1.000	1.000	0.375	0.375	0.063	1.000	1.000
<i>S</i>	-0.5	-5.5	-5	-1.5	-3.5	-4.5	2.5	-5	-5.5	-7.5	-4.5	-2.5
Pr ≥ <i>S</i>	1.000	0.188	0.125	0.750	0.438	0.313	0.625	0.250	0.188	0.063	0.313	0.625

Appendix C-5. Differences between sampling reps at the densely-birded bridge, Station 21186, 2012 – 2013. Significant differences ($p = 0.05$) are highlighted. Mean differences are presented for reps 1 – 2 (R12), reps 1 – 3 (R13), and reps 2 – 3 (R23). Skewness values $> |0.5|$ are highlighted orange, values $> |1.0|$ are bold italic. Values were \log_{10} -transformed prior to testing.

Event Comparison	E12	E13	E23	E12	E13	E23	E12	E13	E23	E12	E13	E23
Survey	26-Apr-12			1-May-12			24-May-12			29-May-12		
<i>n</i>	5	5	5	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	-6.0	-1.4	4.6	-0.2	5.4	5.6	-8.2	-7.2	1.0	-0.6	2.4	3.0
Skewness	-1.26	1.64	1.62	-0.10	-0.35	1.85	0.59	-1.91	-2.18	-1.69	0.58	-0.86
<i>W</i>	0.88	0.82	0.85	0.93	0.96	0.76	0.77	0.76	0.65	0.83	0.96	0.90
Pr< <i>W</i>	0.317	0.117	0.200	0.610	0.838	0.039	0.046	0.039	0.003	0.134	0.797	0.394
<i>t</i>	-1.16	-0.42	0.77	0.06	1.48	1.94	-7.94	-2.23	0.33	-0.12	0.52	0.49
Pr ≥ <i>t</i>	0.312	0.695	0.486	0.958	0.214	0.124	0.001	0.090	0.759	0.910	0.631	0.653
<i>M</i>	-0.5	-1.5	0	0	1.5	2.5	-2.5	-2.5	1.5	1	-0.5	0.5
Pr ≥ <i>M</i>	1.000	0.375	1.000	1.000	0.375	0.063	0.063	0.063	0.375	0.625	1.000	1.000
<i>S</i>	-3.5	-2.5	1	0	5.5	7.5	-7.5	-7.5	2.5	1	0.5	1.5
Pr ≥ <i>S</i>	0.438	0.625	0.875	1.000	0.188	0.063	0.063	0.063	0.625	0.875	1.000	0.813
Survey	9-Apr-13			22-Apr-13			7-May-13			31-May-13		
<i>n</i>	5	5	5	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	3.5	-2.4	-5.9	-2.2	-4.5	-2.3	-2.5	-2.2	0.3	-2.0	9.6	11.6
Skewness	0.40	0.36	-0.46	1.63	1.48	-2.03	-0.36	-0.44	0.80	0.59	0.37	0.30
<i>W</i>	0.97	0.90	0.87	0.84	0.84	0.71	0.99	0.93	0.97	0.96	0.96	0.88
Pr< <i>W</i>	0.902	0.430	0.250	0.153	0.163	0.011	0.980	0.630	0.848	0.793	0.809	0.318
<i>t</i>	1.06	-0.93	-2.00	-0.36	-1.40	-2.87	-1.65	-1.33	0.37	-0.98	2.65	2.72
Pr ≥ <i>t</i>	0.350	0.406	0.116	0.735	0.234	0.045	0.173	0.255	0.732	0.383	0.057	0.053
<i>M</i>	1.5	-1	-1.5	-1.5	-1.5	-2.5	-1.5	-0.5	0	-1	1.5	2.5
Pr ≥ <i>M</i>	0.375	0.625	0.375	0.375	0.375	0.063	0.375	1.000	1.000	0.625	0.375	0.063
<i>S</i>	3.5	-3	-6.5	-2.5	-4.5	-7.5	-5.5	-4.5	1	-3	6.5	7.5
Pr ≥ <i>S</i>	0.438	0.375	0.125	0.625	0.313	0.063	0.188	0.313	0.875	0.375	0.125	0.063

Appendix C-6. Differences between control sampling locations at the non-birded bridge, Station 20018 (2012) and Station 16404 (2013) when migratory cliff swallows were not present. Significant differences ($p = 0.05$) are highlighted. Mean differences are presented for reps 1 – 2 (R12), reps 1 – 3 (R13), and reps 2 – 3 (R23). Skewness values $> |0.5|$ are highlighted orange, values $> |1.0|$ are bold italic. Values were \log_{10} -transformed prior to testing.

Event Comparison	E12	E13	E23	E12	E13	E23	E12	E13	E23	E12	E13	E23
Survey	27-Apr-12			3-May-12			30-May-12			25-Jun-12		
<i>n</i>	5	3	3	5	5	5	5	5	5	5	5	5
Mean Difference (MPN/100 ml)	-79.6	-124.0	-30.0	-7.4	-21.8	-14.4	3.4	5.8	2.4	-7.0	-13.4	-6.4
Skewness	-0.95	-1.28	0.97	0.54	-0.46	1.47	-1.82	0.17	-0.82	0.99	1.43	-0.56
<i>W</i>	0.88	0.93	0.96	0.79	0.85	0.82	0.80	0.91	0.93	0.94	0.87	0.96
Pr< <i>W</i>	0.306	0.472	0.620	0.062	0.188	0.126	0.080	0.438	0.623	0.655	0.260	0.840
<i>t</i>	-6.70	-8.17	-3.85	-1.83	-5.87	-5.68	1.26	2.05	0.85	-1.87	-4.79	-6.91
Pr ≥ <i>t</i>	0.003	0.015	0.061	0.142	0.004	0.005	0.277	0.110	0.443	0.134	0.009	0.002
<i>M</i>	-2.5	-1.5	-1.5	-0.5	-2.5	-2.5	1.5	1.5	0.5	-1.5	-2.5	-2.5
Pr ≥ <i>M</i>	0.063	0.250	0.250	1.000	0.063	0.063	0.375	0.375	1.000	0.375	0.063	0.063
<i>S</i>	-7.5	-3	-3	-4.5	-7.5	-7.5	3.5	6.5	2.5	-5.5	-7.5	-7.5
Pr ≥ <i>S</i>	0.063	0.250	0.250	0.313	0.063	0.063	0.438	0.125	0.625	0.188	0.063	0.063
Station 16404 Temporal Control												
Survey	24-Jan-13			6-Feb-13			27-Feb-13					
<i>n</i>	5	5	5	5	5	5	5	5	5			
Mean Difference (MPN/100 ml)	-7.4	-13.8	-6.4	-2.8	-0.8	2.0	-3.8	-7.2	-3.3			
Skewness	-0.80	-0.16	0.97	1.06	-0.04	-0.87	-1.43	0.29	-1.09			
<i>W</i>	0.96	0.88	0.85	0.87	0.97	0.91	0.87	0.94	0.93			
Pr< <i>W</i>	0.841	0.328	0.203	0.262	0.902	0.439	0.278	0.683	0.567			
<i>t</i>	-2.04	-13.90	-2.04	-1.50	-0.25	1.04	-1.89	-2.84	-2.46			
Pr ≥ <i>t</i>	0.111	<0.001	0.111	0.209	0.817	0.359	0.131	0.047	0.070			
<i>M</i>	-1.5	-2.5	-1.5	-1.5	0	1.5	-1.5	-2	-2.5			
Pr ≥ <i>M</i>	0.375	0.063	0.375	0.375	1.000	0.375	0.375	0.125	0.063			
<i>S</i>	-6.5	-7.5	-5.5	-5.5	-1	4.5	-6.5	-5	-7.5			
Pr ≥ <i>S</i>	0.125	0.063	0.188	0.188	0.875	0.313	0.125	0.125	0.063			