

Bacterial Indicators of Pathogens in the Great Miami River Watershed, Southwest Ohio

INTRODUCTION

The aesthetic, recreational, and ecological health of the Great Miami River Watershed in Southwest Ohio may be influenced by a wide variety of point and nonpoint sources of fecal contamination. Chapter 3745-1 of the Ohio Administrative Code designates aquatic life habitat, water supply, and recreation uses for streams in the watershed. Portions of the watershed currently meet Exceptional Warm Water Habitat (EWH) criteria used for waters with unique and unusual assemblages of aquatic life, above average abundance of sensitive species, and above average populations of top carnivores (Ohio Environmental Protection Agency, 1995, 2001, 2007, and 2011).

In 2010, the Great Miami River was designated by the Ohio Department of Natural Resources as an Ohio Water Trail reflecting the rivers popularity with canoeists and kayakers. Many of the streams in the watershed are also designated for primary contact recreation which means; water depths allow full body immersion, the water body is proximal to residential areas, and there is an intermediate potential for exposure to bacteria. Elevated concentrations of pathogens associated with fecal contamination pose a potential health threat to boaters and fisherman and may degrade the quality of aquatic habitats. Pathogens are microbes that cause disease. Pathogens in river water are also a potential water quality concern for public water systems in the Great Miami River Watershed that rely on the process of induced infiltration to recharge shallow production wells located in close proximity to the river.

METHODS OF SITE SELECTION AND DATA COLLECTION

In 2010, MCD staff conducted periodic sampling of surface water at four sites within the Great Miami River Watershed for bacterial indicators of fecal contamination (see Figure 1). The sampling sites are located near the mouths of the four major subwatersheds that comprise the Great Miami River Watershed in close proximity to U.S. Geological Survey (USGS) stream gaging stations equipped with tipping bucket rain gages. MCD and YSI, Inc. staff deployed YSI™ 6-series sondes at each of the sampling locations to measure water temperature, pH, specific conductance, dissolved oxygen, turbidity, chlorophyll, and blue green algae at hourly intervals.

In practice it is difficult to measure pathogens directly in natural waters because the variety and often low concentrations of pathogenic bacteria and viruses make them difficult to test for individually. One of the primary pathways for pathogens to enter natural waters is through fecal contamination from humans and animals. Bacterial indicators of fecal contamination are present in water when bacterial pathogens from fecal contamination are present. Thus, bacterial indicators of fecal contamination may be used as indicators of risk for the occurrence of fecal pathogens.

MCD staff selected fecal coliform and *Escherichia coli* (*E. coli*) bacteria as the indicators of fecal contamination for this investigation. Fecal coliform bacteria are present in large numbers in the feces and intestinal tracts of humans and other warm-blooded animals and can enter water bodies from human and animal waste. *E. coli* is a rod shaped, gram negative bacterium, commonly found in the gastrointestinal tract and feces of warm-blooded animals. It is one species within the fecal-coliform group of bacteria and can be distinguished from other fecal coliforms by biochemical tests. Most strains of *E. coli* are harmless, but some strains can cause illness. Fecal coliform and *E. coli* bacteria indicate the potential presence of water-borne pathogens that may present a public health risk for humans that consume or have bodily contact with contaminated water (Dufour, 1977).

MCD staff grabbed samples at each of the four sampling sites several times per month between January 20 and December 20, 2010. Sampling frequency was targeted to be representative of a wide range of flow conditions at the sampling sites. The samples were collected by wading out as close as possible to the middle of the river channel and filling up sterilized plastic sample containers. The containers were packed in a cooler with ice and transported to the laboratory on the day of sample collection. The laboratory analyzed the water samples for fecal coliform using Standard Method 9222D. All fecal coliform results are reported as a number of colonies per 100 ml of water. The laboratory used an IDEXX Quanti-Tray 2000™ to measure *E. coli* concentrations. All *E. coli* results are reported as a most probable number of colony forming units per 100 ml of water.

WATER QUALITY STANDARDS

The Ohio Environmental Protection Agency (OEPA) set recreational use standards for *E. coli* which apply to streams designated for primary and secondary contact recreation. See Table 1 for statewide numerical standards for *E. coli* based upon the recreation use the state designated for a particular stream. Portions of the Great Miami River Watershed are designated for Class A primary contact recreation. For the purposes of this investigation, the Class A *E. coli* standards are used to evaluate *E. coli* data.

Table 1. Statewide numerical criteria for the protection of recreation uses

Recreation use	E. coli (colony counts per 100 ml)	
	Seasonal geometric mean	Single sample maximum
Bathing water	126	235
Class A primary contact recreation	126	298
Class B primary contact recreation	161	523
Class C primary contact recreation	206	940
Secondary contact recreation	1030	1030

The above criteria apply inside and outside the mixing zone for wastewater treatment plant discharges during the recreation season which runs from May 1 to October 31.

DATA ANALYSIS

Figure 2 shows time series plots of fecal coliform and *E. coli* concentrations measured in samples collected during this investigation at each of the four sampling sites. The plots show similar up and down trends for each of the sampling sites and a wide range in concentrations for the two bacterial indicators. Concentrations of fecal coliform and *E. coli* ranged from less than 10 to several thousand colonies per 100 ml of water. *E. coli* concentrations exceeded the OEPA single-sample maximum numerous times at each of the sampling sites (see Table 2). Three out of the four sampling sites did not meet the seasonal geometric mean *E. coli* criteria for Class A primary contact recreation (see Table 2).

Table 2. Summary of bacteria data in relation to regulatory criteria

Station	Number of samples	Seasonal geometric mean (May 1 – Oct 31)	Number of samples with <i>E. coli</i> concentration > 298 colonies per 100 ml
Stillwater River	26	219.60	6
Upper Great Miami River	26	136.56	8
Mad River	26	352.44	12
Lower Great Miami River	27	107.44	9

Figure 3 is a time series plot of streamflow and bacteria concentrations measured at the Lower Great Miami River sampling site. The plot shows a tendency for higher bacteria concentrations to occur during higher flows.

Relations among bacteria concentrations, field measured properties of water, and streamflow were evaluated statistically by use of Spearman-rank correlations (Sokal and Rohlf, 1995). The Spearman-rank correlation coefficient or Spearman's rho is a non-parametric measure of statistical dependence between two variables. It assesses how well the relationship between two variables can be described using a monotonic function. Spearman-rank coefficients can range from -1 to 1. A value of 1 or -1 implies that each variable is a monotone function of the other. A value of 0 implies that a monotonic relationship does not exist between the two variables. Positive correlation coefficients between measured variables indicate when one variable increases the other variable also increases. Negative correlation coefficients indicate when one variable decreases the other variable increases. Spearman-rank correlation coefficients for various combinations of measurement variables are shown in Table 3.

Table 3. Spearman-rank correlations (r_s)

[r_s , Spearman Rho coefficient; n, number of samples; p-value, significance value]

Relationship	Lower Great Miami River			Stillwater River			Mad River			Upper Great Miami River		
	r_s	n	p-value	r_s	n	p-value	r_s	n	p-value	r_s	n	p-value
<i>E. coli</i> - streamflow	0.62	25	<0.05	0.29	24	>0.05	-0.04	24	>0.05	0.59	24	<0.05
<i>E. coli</i> - turbidity	NA	NA	NA	0.76	24	<0.05	0.35	16	>0.05	-0.02	22	>0.05
<i>E. coli</i> - water temperature	-0.35	25	>0.05	0.12	24	>0.05	0.28	24	>0.05	-0.42	24	<0.05
<i>E. coli</i> - fecal coliform	0.77	25	<0.05	0.69	24	<0.05	0.91	24	<0.05	0.91	24	<0.05
fecal coliform - streamflow	0.50	25	<0.05	0.15	24	>0.05	0.08	24	>0.05	0.45	24	<0.05

fecal coliform - turbidity	NA	NA	NA	0.63	24	<0.05	0.22	16	>0.05	0.08	21	>0.05
fecal coliform - water temperature	-0.17	25	>0.05	0.16	24	>0.05	0.29	24	>0.05	-0.30	24	>0.05

NA - turbidity data not available

Statistically significant relations (for $p < 0.05$) were found to exist. Spearman-rank correlations significant at the 95-percent confidence interval exist between *E. coli* and fecal coliform for all four sample sites suggesting when one indicator of fecal contamination is present at an elevated concentration the other indicator is also likely to be present at an elevated concentration. Statistically significant positive correlations also exist for the indicators *E. coli* and fecal coliform and streamflow at the Lower Great Miami River and Upper Great Miami River sites. *E. coli* and fecal coliform had statistically significant positive correlations with turbidity at the Stillwater River site. *E. coli* and water temperature had a statistically significant negative correlation at the Upper Great Miami River site.

The distribution of *E. coli* and fecal coliform concentrations for each site are summarized in figure 4. The box plots show that median *E. coli* concentrations for all sites fell below the single-sample maximum standard for Class A primary contact recreation. The Stillwater River and Upper Great Miami River sites had the lowest median *E. coli* concentrations (114.5 colonies per 100 ml of water).

Sample concentrations of *E. coli* and fecal coliform collected at all four sites under base flow conditions are compared with samples collected under storm flow conditions in the box plots shown in figure 5. For the purposes of this report, base flow conditions were defined as flows less than or equal to the 90th exceedance percentile for the nearest streamgaging station. Storm flow conditions were defined as meeting at least one of the following criteria:

1. *Flows equal to or exceeding the 10th exceedance percentile for the nearest streamgaging station occurred at the date and time of sample collection.*
2. *Precipitation as rainfall greater than or equal to 0.5 inches was recorded at the nearest precipitation gage during the 48 hours preceding the date and time of sample collection.*

The plots show that median storm flow sample concentrations of *E. coli* and fecal coliform tended to be higher than median base flow sample concentrations suggesting that some of the bacterial indicators of fecal contamination are derived from wet weather processes such as combined sewer overflows (CSOs) or runoff from agricultural fields fertilized with manure. The median concentration of *E. coli* for all samples collected under baseflow conditions was 89.5 colonies per 100 ml of water. The median concentration of *E. coli* for all samples collected under storm flow conditions was substantially higher at 533.5 colonies per 100 ml of water. Median concentrations of *E. coli* fell below the Class A primary contact single-sample maximum standard for samples collected under base flow conditions. In contrast, median concentrations of *E. coli* for storm flow samples exceeded the standard. While *E. coli* and fecal coliform concentrations measured in samples collected under base flow conditions tend to be lower than storm flow samples, high concentrations are still often present and suggest that dry weather

sources such as wastewater effluent from treatment plants and septic systems are also important sources of fecal contamination in the Great Miami River Watershed.

Seasonality in the Great Miami River Watershed affects bacterial indicator concentrations through variations in streamflow, precipitation, water temperature, soil drainage characteristics, and the timing of agricultural applications of fertilizers such as manure. Box plots of concentration ranges of *E. coli* and fecal coliform for samples collected at all four sites during each season are shown in figure 6. The plots show *E. coli* and fecal coliform median concentrations tended to be lowest during the summer months. Median *E. coli* concentrations fell below the Class A primary contact recreation single-sample maximum standard during the winter, summer, and fall seasons but rose above the standard during the spring season.

FINDINGS AND CONCLUSIONS

This investigation showed concentrations of the bacterial indicator *E. coli* often exceeded the OEPA Class A primary contact recreation single-sample maximum standard of 298 colonies per 100 ml in the Great Miami River and its tributaries. The OEPA Class A primary contact recreation seasonal geometric mean standard of 126 colonies per 100 ml was also exceeded on the Stillwater River, Upper Great Miami River, and Mad River sampling sites. Sampling results showed that *E. coli* and fecal coliform concentrations have strong positive correlations at all sampling sites. When *E. coli* concentrations are high fecal coliform concentrations also tend to be high and vice versa.

Median concentrations of *E. coli* fell below the OEPA Class A single-sample maximum standard at all four sampling sites. The Mad River site had the highest median concentrations of *E. coli* and fecal coliform out of the four sampling sites used in this investigation.

In general, *E. coli* and fecal coliform concentrations tended to be higher during storm flow events than during base flow conditions. However elevated concentrations of both bacterial indicators occurred during storm flow and base flow conditions. The data suggests wet weather processes are an important source of fecal contamination in the Great Miami River Watershed. However wet weather processes alone are not the only factor influencing fecal contamination as evidenced by the presence of elevated bacterial indicator concentrations in samples collected during base flow conditions.

Seasonality also plays a role in determining *E. coli* and fecal coliform concentrations in the watershed. Both bacterial indicators tend to be present at higher concentrations in the spring and fall seasons. Median concentrations of *E. coli* for all samples collected during the spring seasons fell above the OEPA Class A primary contact single-sample maximum standard. Median concentrations of *E. coli* samples collected during the winter, summer, and fall seasons fell below the standard.

The data collected as part of this sampling project suggest that wet weather or runoff events play a significant role in contributing to fecal contamination of natural waters in the Great Miami River Watershed. Elevated concentrations of fecal indicators were more pronounced during the spring and late fall seasons and often corresponded to large runoff events. Analysis of samples

collected during base flow conditions occasionally detected elevated bacterial indicator concentrations showing that dry weather processes also play a role in introducing fecal contamination to the Great Miami River Watershed.

Future sampling of bacterial indicators of pathogens should focus on trying to identify which sources of contaminants are most significant, so management strategies to reduce fecal contaminants can be appropriately targeted.

REFERENCES

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- Ohio Environmental Protection Agency (1995). *Biological and water quality study of the Upper Great Miami River and selected tributaries: OEPA Technical Report MAS/1995-12-13*.
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Figure 1. Sampling sites in the Great Miami River Watershed.

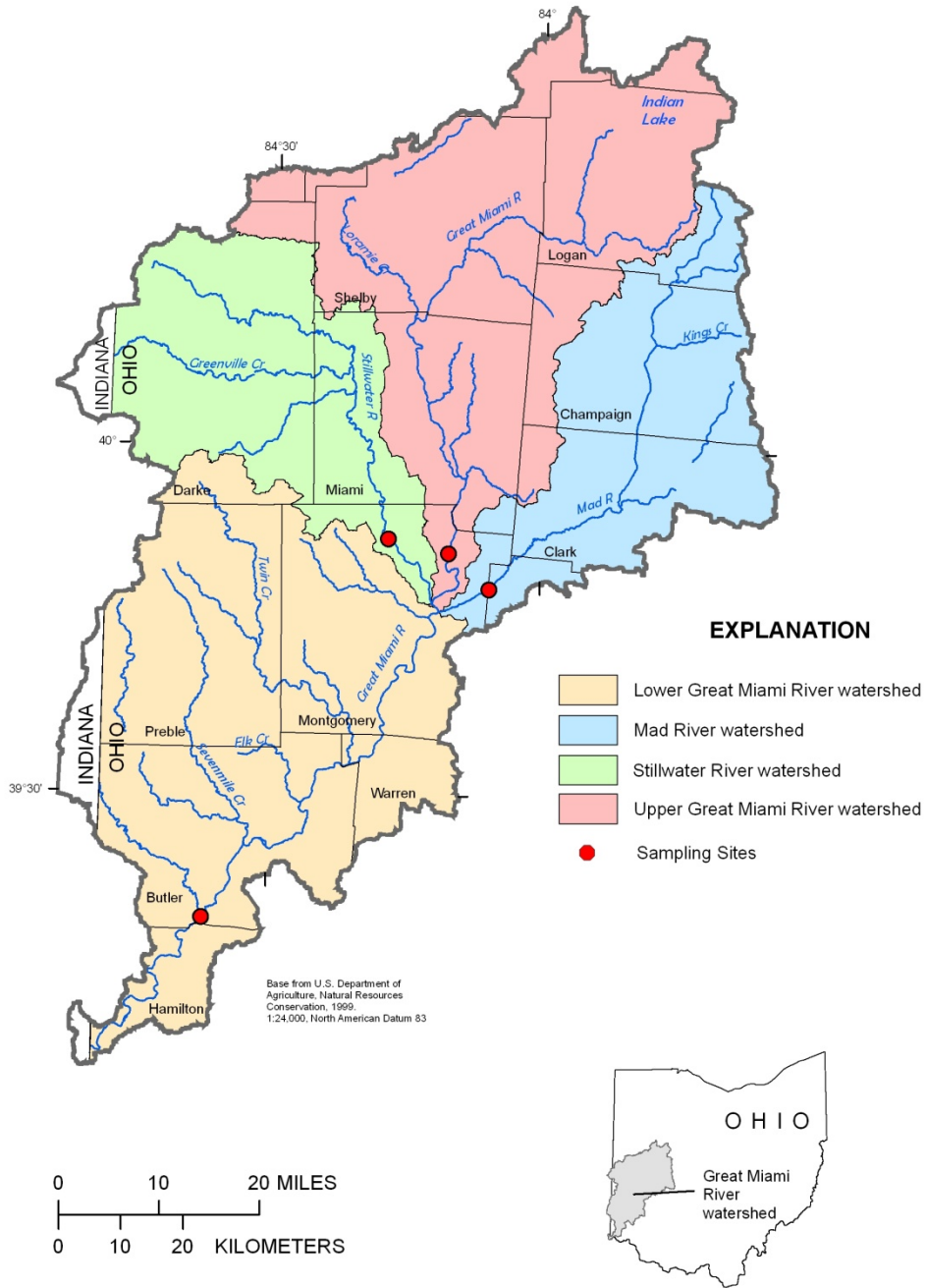


Figure 2. Time series plots showing fecal coliform and *E. coli* concentrations measured at each sampling site in 2010.

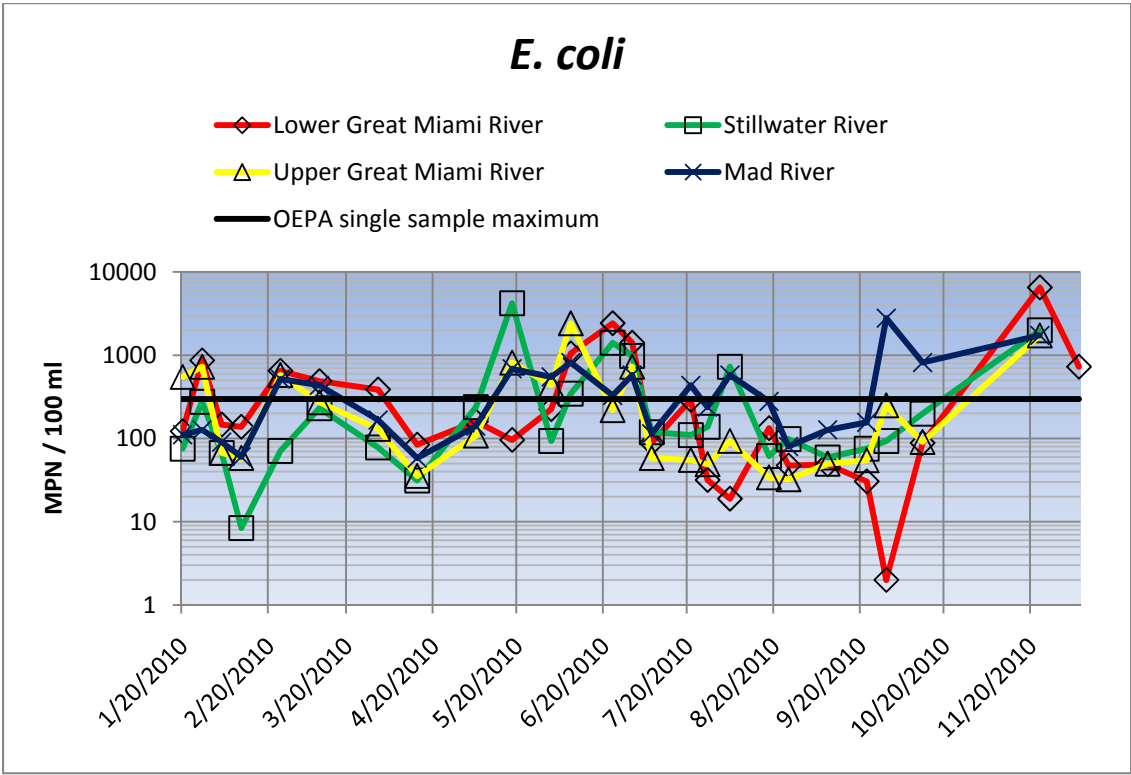
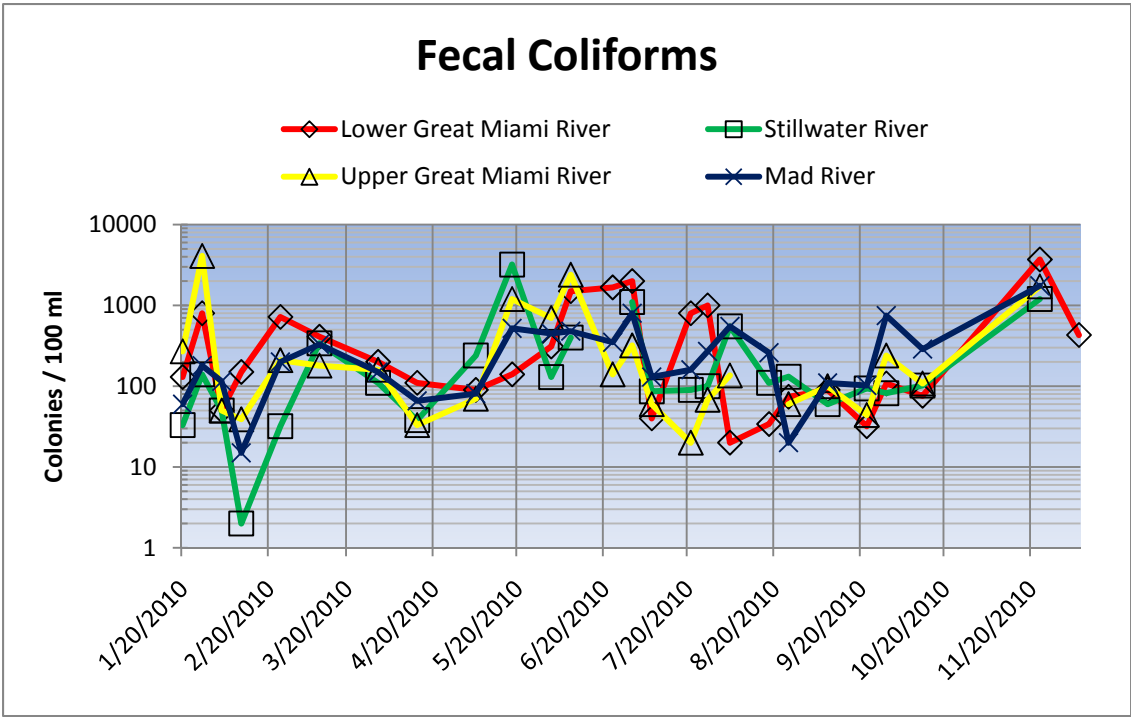


Figure 3. Time series plot showing streamflow and bacteria concentrations measured in the Lower Great Miami River.

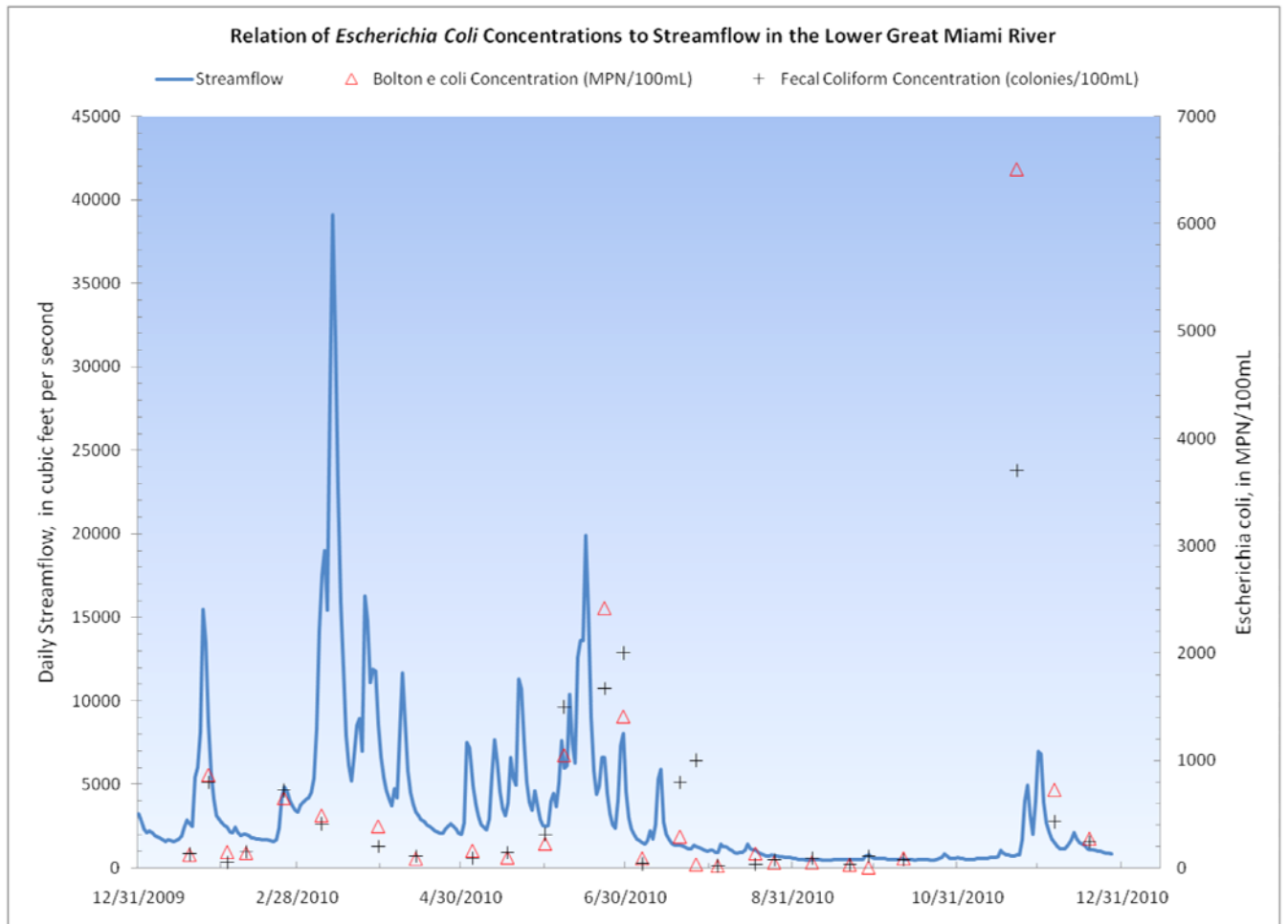


Figure 4. Distribution fecal coliform and *E. coli* concentrations by subwatershed.

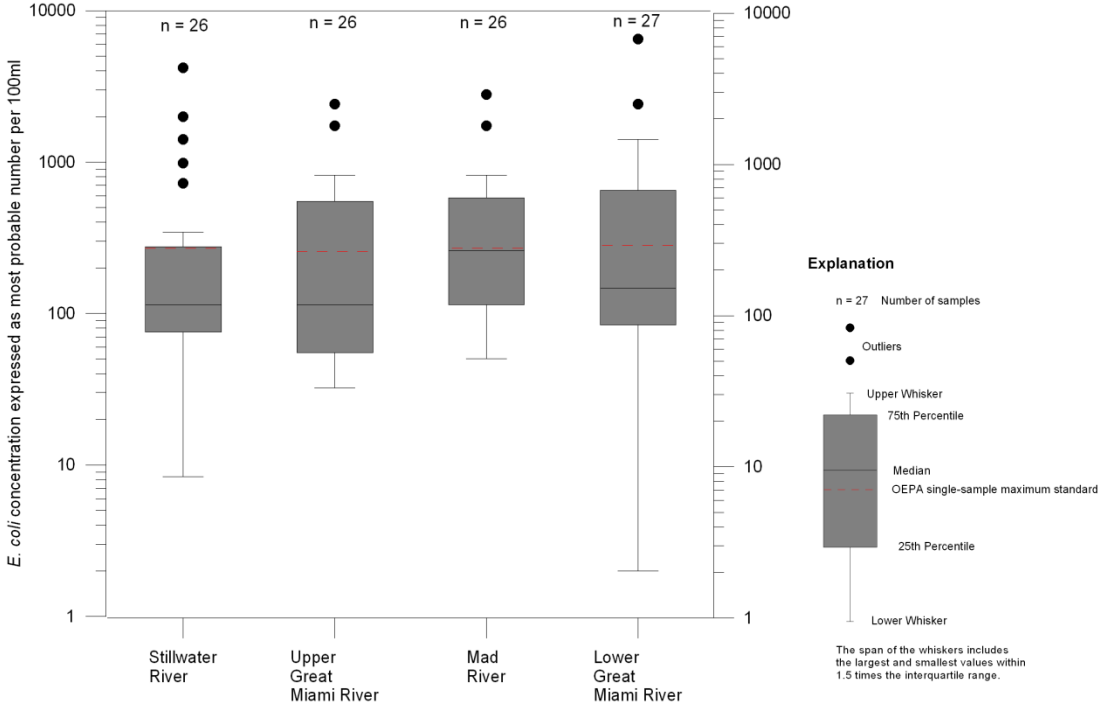
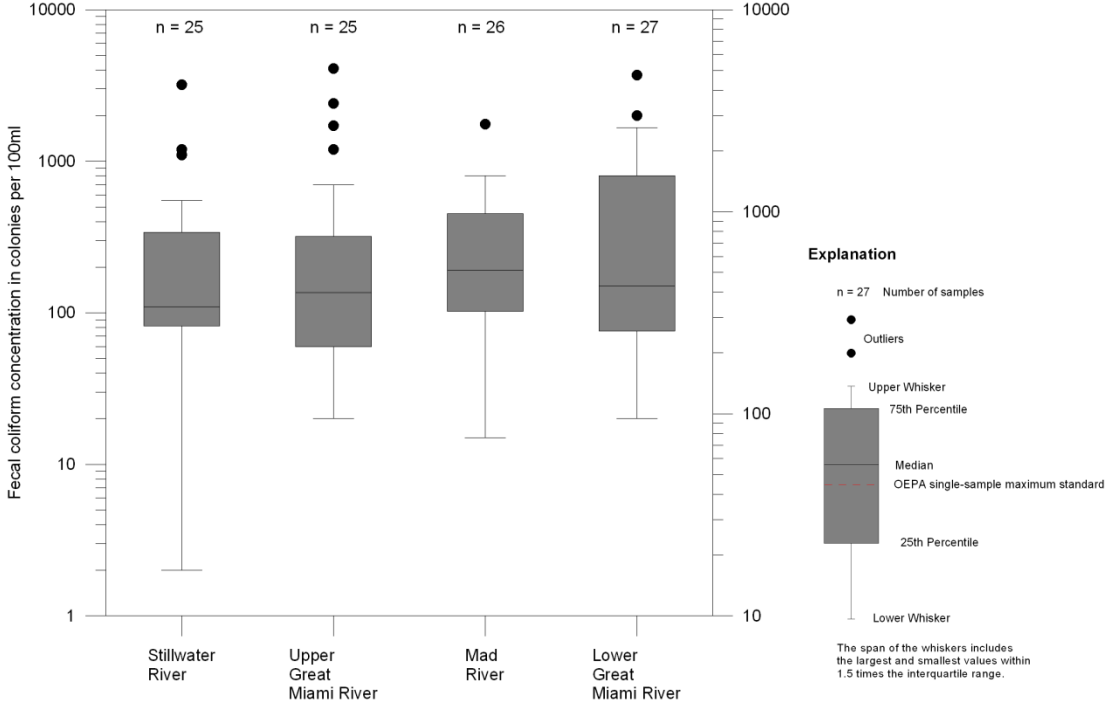


Figure 5. Distribution of E. coli and fecal coliform bacteria concentrations during base flow and storm flow for all sampling sites.

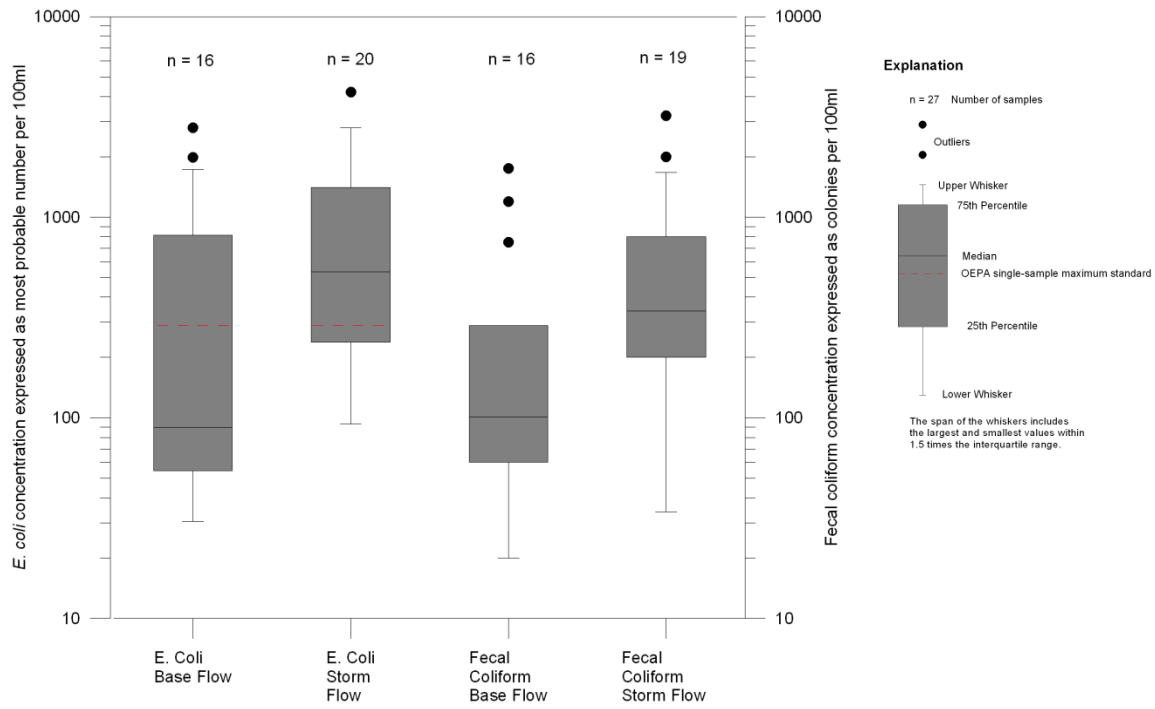
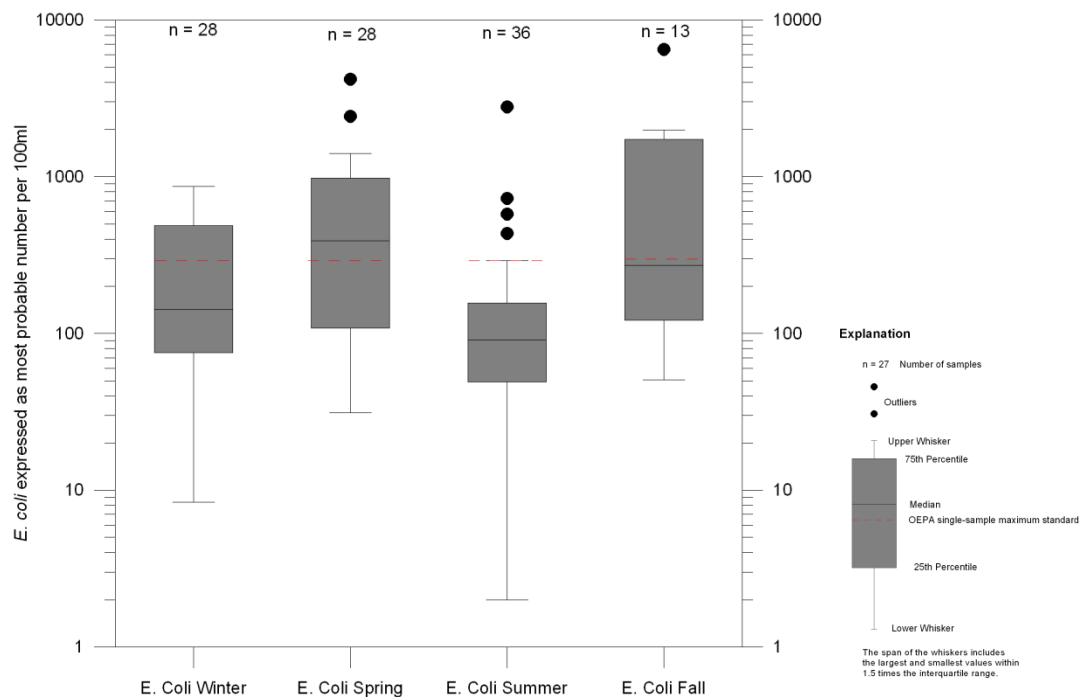
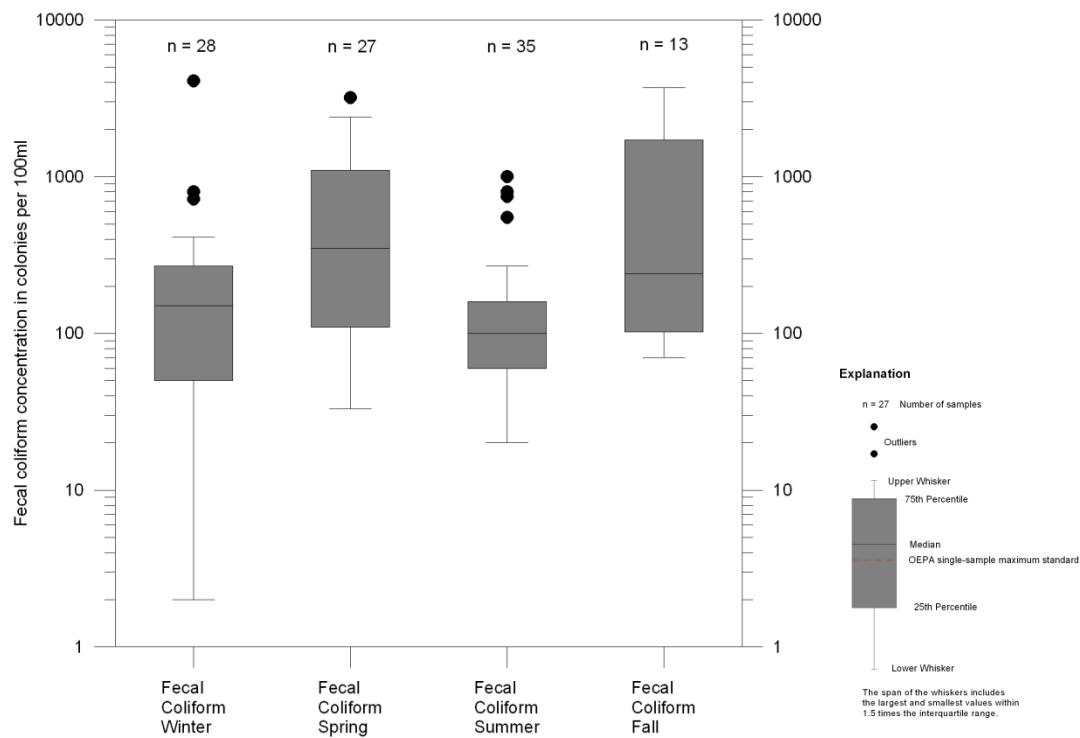


Figure 6. Seasonal variations in fecal coliform and *E. coli* concentrations for all sampling sites.





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