

ASSESSING COLIFORM RATIOS IN DEPRESSIONAL WETLANDS AS POTENTIAL INDICATORS OF HYDROLOGIC CONNECTIVITY

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Abstract. Depressional wetlands occur in dense clusters on the Dougherty Plain, a physiographic province of Georgia dominated by agricultural land use. These wetlands represent a major potential storage of surface water in southwest Georgia. Hydrologically, these systems are primarily driven by precipitation and evapotranspiration, but a substantial subset are connected to episodic storm flows. The water quality of wetlands connected to episodic flows (recurrence interval <2 years) differ from those that are more isolated from flows (recurrence interval >2 years). However, the extent and / or source water of wetland connectivity is unknown. It may be possible to determine these wetland characteristics through water quality methods. In ecological water quality studies, *E. coli* and total coliform ratios can indicate wildlife presence or the influence of agricultural stormwater / anthropogenic contamination depending on landscape context. The objectives of this study are to: 1) analyze coliform ratios from an episodic flow event for potentially identifying source waters; 2) analyze coliform ratios in a subset of depressional wetlands with regard to episodic flow connectivity; and 3) determine additional water quality variables that may be associated with coliform ratios in depressional wetlands. Water quality data were collected from seven sites along an episodic flow path during an episodic flow event (February-March 2014). Data were also collected through monthly monitoring of 31 depressional wetlands on the Dougherty Plain (March 2014 - March 2015). A suite of biological (coliforms), chemical (nutrients, dissolved carbon), and physical (conductivity, turbidity, dissolved solids) water quality parameters were measured. Preliminary analysis indicates that greater ratios of *E. coli* to total coliforms were observed in wetlands connected to episodic flows and in depressional wetlands isolated from flows. Results are directly applicable to determining depressional wetland source waters as well as source water signals in episodic flows.

INTRODUCTION

Depressional wetlands are surface water features which occur in dense clusters on the Dougherty Plain of southwest Georgia (Hicks et al. 1987, Martin et al. 2012). This karstic physiographic province of Georgia is dominated by agricultural land use and these clusters of wetlands represent a major potential storage of surface water in southwest Georgia. Hydrologically, these systems are primarily

driven by precipitation and evapotranspiration. However, a substantial subset is connected to episodic storm flows.

Episodic flows connect some depressional wetlands to nearby surface waters after intense storm events (Hicks et al. 1987). Runoff generated on agricultural lands flows through distinct or semi-distinct channels often intersecting small depressional wetlands. These wetlands then sequester sediments, reduce nutrient concentrations, and decrease pathogen indicator (coliforms) levels (Deemy and Rasmussen 2017).

Water quality parameters may be a viable method for determining wetland connectivity and/or source waters. Pathogen indicators may also be particularly useful for such analyses. In ecological water quality studies, proportions of *E. coli* and total coliforms can be used to estimate influence of agricultural stormwater / anthropogenic contamination depending on landscape context. An understudied aspect of coliform / pathogen indicator water quality is the ratio of *E. coli* to total coliforms.

OBJECTIVES

Our objectives are to: 1) analyze coliform ratios from an episodic flow event for potentially identifying source waters; 2) analyze coliform ratios in a subset of depressional wetlands with regard to episodic flow connectivity; and 3) determine additional water quality variables that may be associated with coliform ratios in depressional wetlands.

METHODS

Research Site

Research was completed at the J.W. Jones Ecological Research Center within the Dougherty Plain of Georgia (Figure 1). The Dougherty Plain is a physiographic region of Georgia with approximately 11,600 isolated wetlands, characterized by deep sands and interbedded clay lenses over highly permeable, vuggy limestone bedrock (Figure 2) (Hicks et al. 1987, Martin et al. 2012, Rugel et al. 2016).

Sampling Design

Water quality data were collected from two episodic flow paths during a flow event that occurred February-March 2014 (Figure 2). The first flow path was sampled where flow crossed into the Ichuaway property boundary and at the inflows and outflows of two depressional wetlands

located in series along the flow path. The second flow path was sampled at the inflow and outflow of a single depressional wetland located just inside the property boundary where flow enters the site.

Monthly monitoring of 31 depressional wetlands at Ichuaway (Figure 3) and occurred for one year after an episodic flow event (March 2014-March 2015). Wetlands were classified among three connectivity types: Isolated (n=19), Agricultural Runoff (n=6), and Forest Runoff (n=6).

Water Quality Data

A common suite of biological (coliforms), chemical (Cl⁻, F⁻, SO₄²⁻, PO₄³⁻, inorganic nitrogen, dissolved carbon), and physical (conductivity, turbidity, dissolved solids) water quality parameters were measured during each sampling regime (see Deemy and Rasmussen 2017 for detailed methods). Coliform levels were measured using IDEXX Colilert defined substrate methods (Desi and Rifai 2013).

Analysis

Coliform ratios were determined by dividing the *E. coli* level (MPN) by the total coliform level (mpn). Longitudinal patterns in episodic flows were determined using ANOVA for the two wetland / five sampling location flow path (Ichuaway Drain) and t-test for the one wetland / two sampling location flow path (Parmalee Drain). ANOVA was used to compare coliform ratios among wetland connectivity types for the monthly monitoring data. Coliform ratios were correlated with other parameters to identify potential monitoring targets (Zeng and Rasmussen 2005). All statistical analyses were completed in the R programming environment (R Core Team 2019).

RESULTS

Coliforms decreased along both flow paths (Figure 4). In the larger flow path, a significant decrease in coliform ratio occurred between multiple sampling locations and the final two sampling locations (Table 1). Changes tended to be more likely between sampling locations, separated by at least one other sampling location in Ichuaway Drain. However, in the Parmalee drain, differences were observed between the upstream inflow to the wetland and the downstream outflow from the wetland ($p = 0.146$).

No differences occurred among wetland connectivity types (Figure 5). However, Big Cypress Creek did exhibit greater ratios of *E. coli* to total coliforms than either the Flint River or Ichawaynochaway Creek (Table 2).

In the smaller episodic flow path (Parmalee Drain) turbidity and total suspended solids emerged as potential monitoring parameters ($r > 0.8$, $p < 0.001$, Figure 6). No significant relationships between coliform ratios and other parameters were observed in the other episodic flow path (Ichuaway Drain).

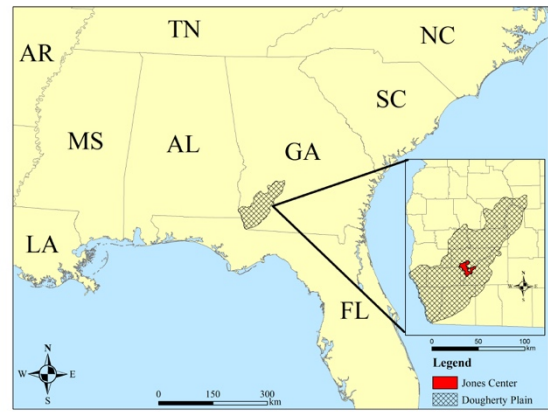


Figure 1. Data collection occurred at the Jones Center at Ichuaway which is centrally located within the Dougherty Plain of southwest Georgia.

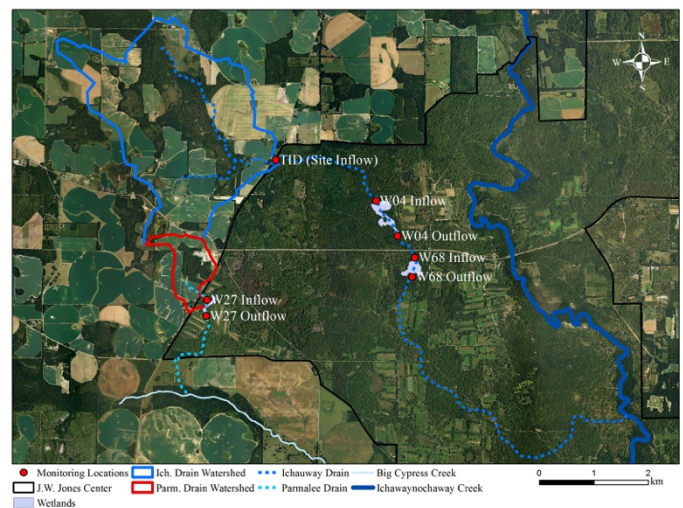


Figure 2. Sampling sites along episodic flow paths along Parmalee Drain (W27 Inflow & Outflow) and Ichauway Drain (W04 Inflow & Outflow, W68 Inflow & Outflow).

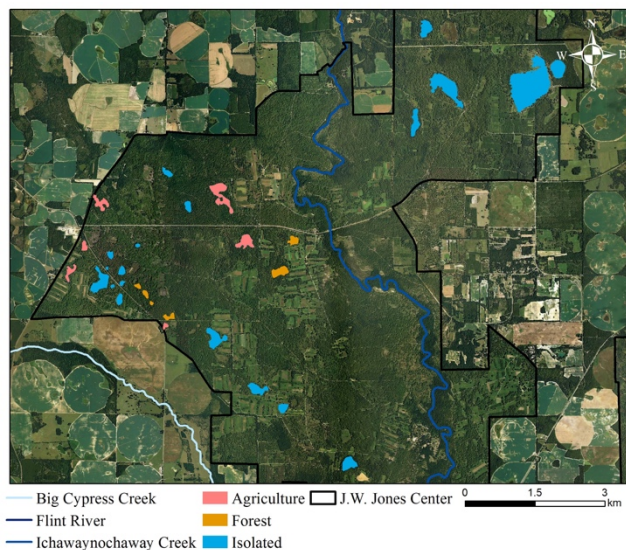


Figure 3. The 31 depressional wetlands monitored in the Dougherty Plain. Different colors indicate wetland connectivity type.

Table 1. Tukey HSD pairwise comparisons. Note: up designates the upstream or inflow sampling site at a wetland and dn designates downstream or outflow of a wetland.

Comparison	p-value
TID - W04up	--
TID - W04dn	--
TID- W68up	< 0.001
W04up - W04dn	--
W04up - W68up	--
W04up - W68dn	< 0.01
W04dn - W68up	--
W04dn - W68dn	< 0.01
W68up - w68dn	--

Table 2. Tukey HSD pairwise comparisons among streams.

Comparison	p-value
Flint River - Big Cypress	< 0.0001
Ichawaynochaway - Big Cypress	< 0.0001
Ichawaynochaway - Flint River	--

DISCUSSION

Coliform ratios decrease longitudinally along the episodic flow paths which may indicate that wetlands reduce *E. coli* at greater rates than total coliforms. These results are interesting because they indicate that human or livestock source pathogen indicating bacteria (*E. coli*) are reduced by flow through wetlands at a greater rate than total coliforms. This coupled with relationships between coliform ratio and turbidity / suspended solids may indicate that by sequestering sediments, wetlands are also removing pathogen indicators.

Wetlands connected to episodic flows did not differ in coliform ratios from depressional wetlands isolated from flows. This was unexpected but may indicate that wetlands remove pathogens at a rate sufficient to show no difference on an annual basis.

The greater coliform ratios in Big Cypress Creek were unsurprising as this stream flows directly through cattle pastures and receives direct agricultural runoff. This stream is ephemeral and generally flows seasonally whereas the

other two streams are perennial. Follow up analyses will focus on comparing the data in specific months rather than the aggregate dataset.

Results also indicate that turbidity and suspended solids could be potential monitoring parameters for coliform ratios in episodic flows. However, this requires further testing and focused inquiry.

CONCLUSION

Longitudinal changes in coliform ratios along episodic flow paths indicate that anthropogenic indicator bacteria levels decrease faster than the total coliform levels in episodic flows. Additionally, turbidity and suspended solids emerged as potentially viable monitoring parameters for coliform ratios. Monthly monitoring appears to indicate that no difference in depressional wetland coliform ratio persists after episodic flow events.

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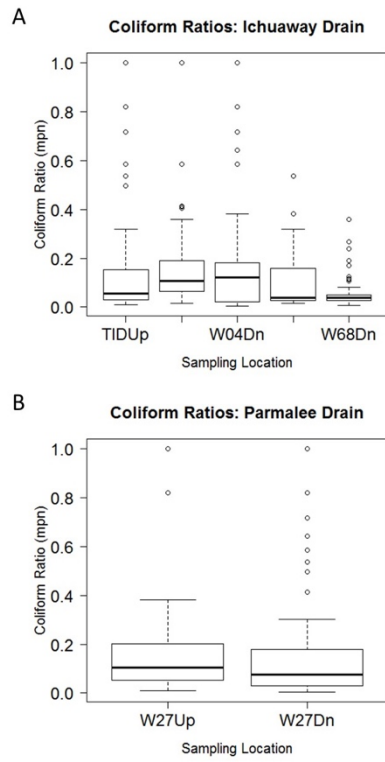


Figure 4. Coliform ratios decreased longitudinally along both Ichauway drain (A) and Parmalee (B) drain.

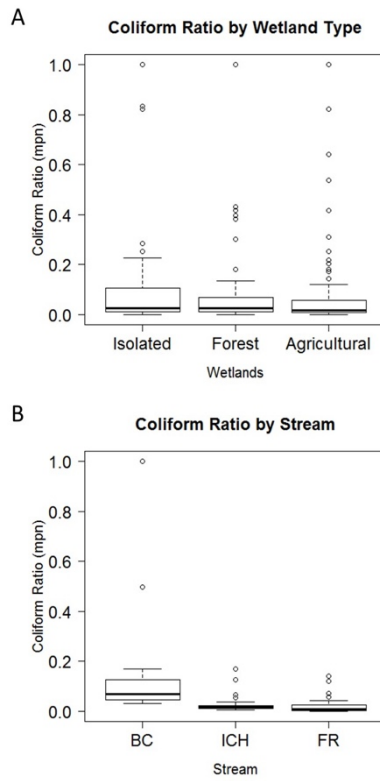


Figure 5. Coliform ratios did not differ among wetlands (A) but were greater in Big Cypress Creek than Ichauwaynochaway Creek and the Flint River (B).

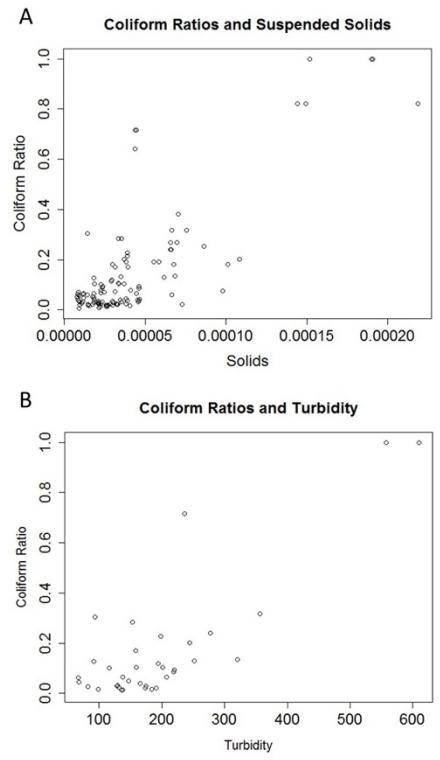


Figure 6. Suspended solids (A) and turbidity (B) were both strongly correlated with coliform ratios in the Parmalee Drain ($r > 0.8$, $p < 0.01$).