

**PURPOSE  
AND APPLICATION  
OF THE  
DECISION SUPPORT TOOL**

# Why is a decision-support tool needed to help with fecal coliform BMAPs ?

- Florida currently has water quality criteria for fecal coliforms (e.g., no more than 10% of samples should exceed 400 CFU/100 mL), and is developing TMDLs and BMAPs for WBIDs that aren't meeting those criteria
- Coliform bacteria have been used as indicators of potential fecal contamination of water since the late 1800s. (Originally for detecting contamination of water by untreated or minimally-treated sewage.)
- In tropical and sub-tropical areas, fecal coliforms and other bacterial indicators can grow in soils and on vegetation, producing "false-positive" criterion exceedances that aren't correlated with elevated human health risk
- They can also produce "false-negative" results – e.g., pathogenic viruses and protozoa (*Giardia* or *Cryptosporidium*) can survive longer than fecal coliforms in surface waters, producing human health risk without elevated indicator levels

# How does this tool help ?

- Provides a framework for interpreting and responding to concentrations of fecal coliforms and other indicators observed in ambient monitoring programs
- Helps managers prioritize WBIDs and areas within WBIDs for investigation and follow-up action
- Based on conceptual approaches currently used by the World Health Organization (WHO 2003) and recommended by the National Research Council (NRC 2004)
- For communicating results to elected officials and the public, allows the use of a simple "stoplight" (green/yellow/red) graphical approach, similar to one used in Tampa Bay area to provide annual tracking of water quality conditions and attainment of water quality goals

# How should managers address technical limitations of fecal coliforms and other bacterial indicators, and protect public health?

- WHO (2000, 2003) recommends use of "Annapolis protocol", which combines bacterial indicator counts with on-site assessments of potential pathogen sources
- NRC (2004) recommends Annapolis protocol and a "phased monitoring approach" to help identify and address pathogen sources



# "Annapolis protocol" (WHO 2003) conceptual overview

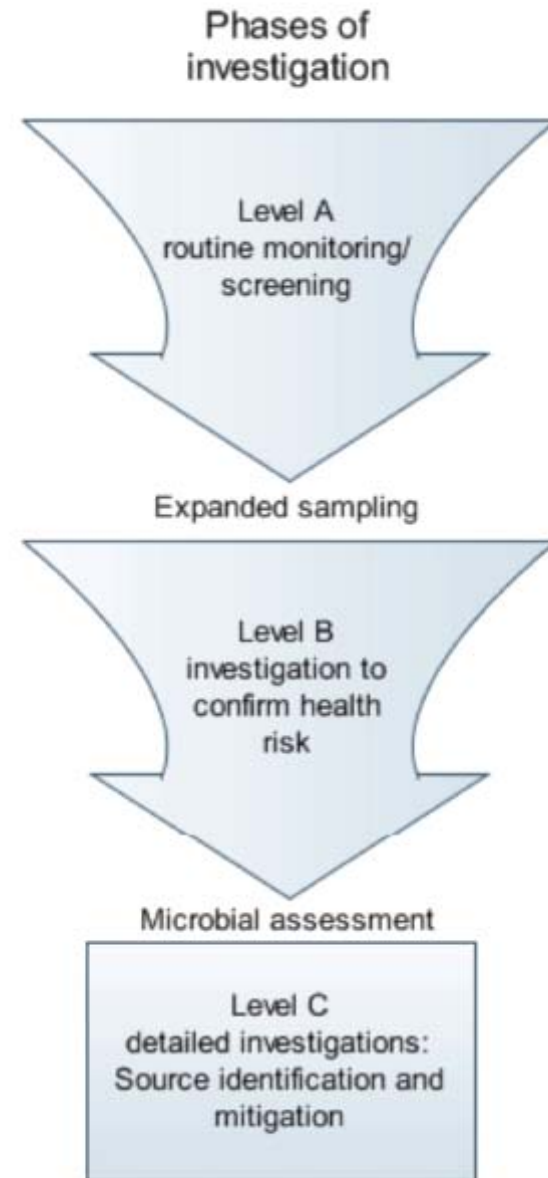
Bacterial Indicator  
Concentrations  
(based on monitoring data)



Estimated Health Risk

# Phased monitoring approach

Recommended by NRC (2004) to help maximize cost-effectiveness of microbial monitoring and MST



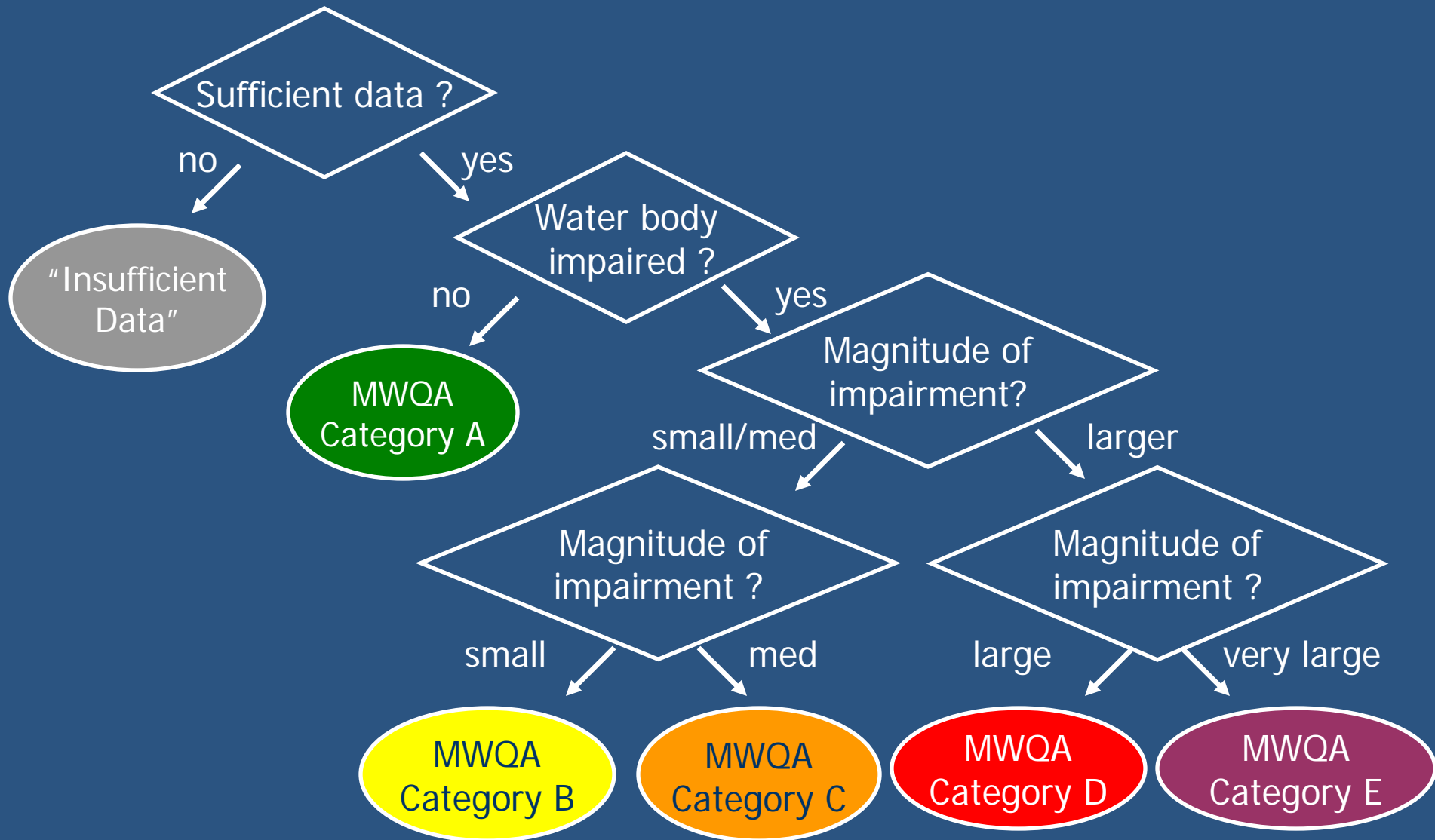
# How to incorporate these ideas into fecal coliform TMDLs and BMAPs ?

- Use monitoring data (e.g., fecal coliform concentrations) as a screening tool, to help identify and prioritize sites for management attention
- Use information from "contaminant source surveys" (CSS) to supplement the bacterial data
- Use "Annapolis protocol" approach to combine the information and guide management responses

# Step 1

- Characterize microbial water quality conditions within each WBID, based on fecal coliform concentrations observed in the available monitoring data

# Decision Tree to Define Microbial Water Quality Assessment (MWQA) Categories Using Fecal Coliform Data

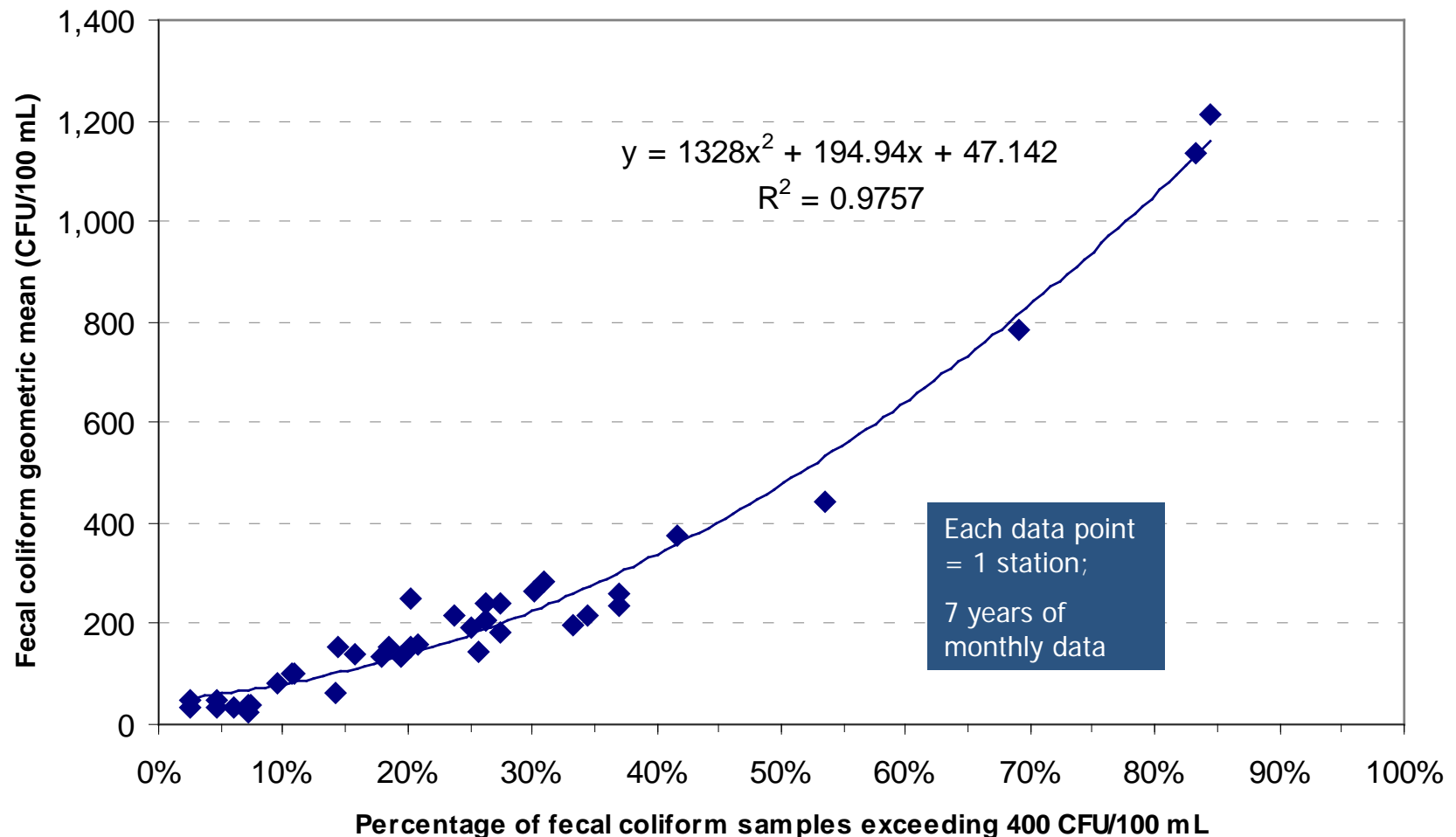


# How to Make the Decision Tree Quantitative ?

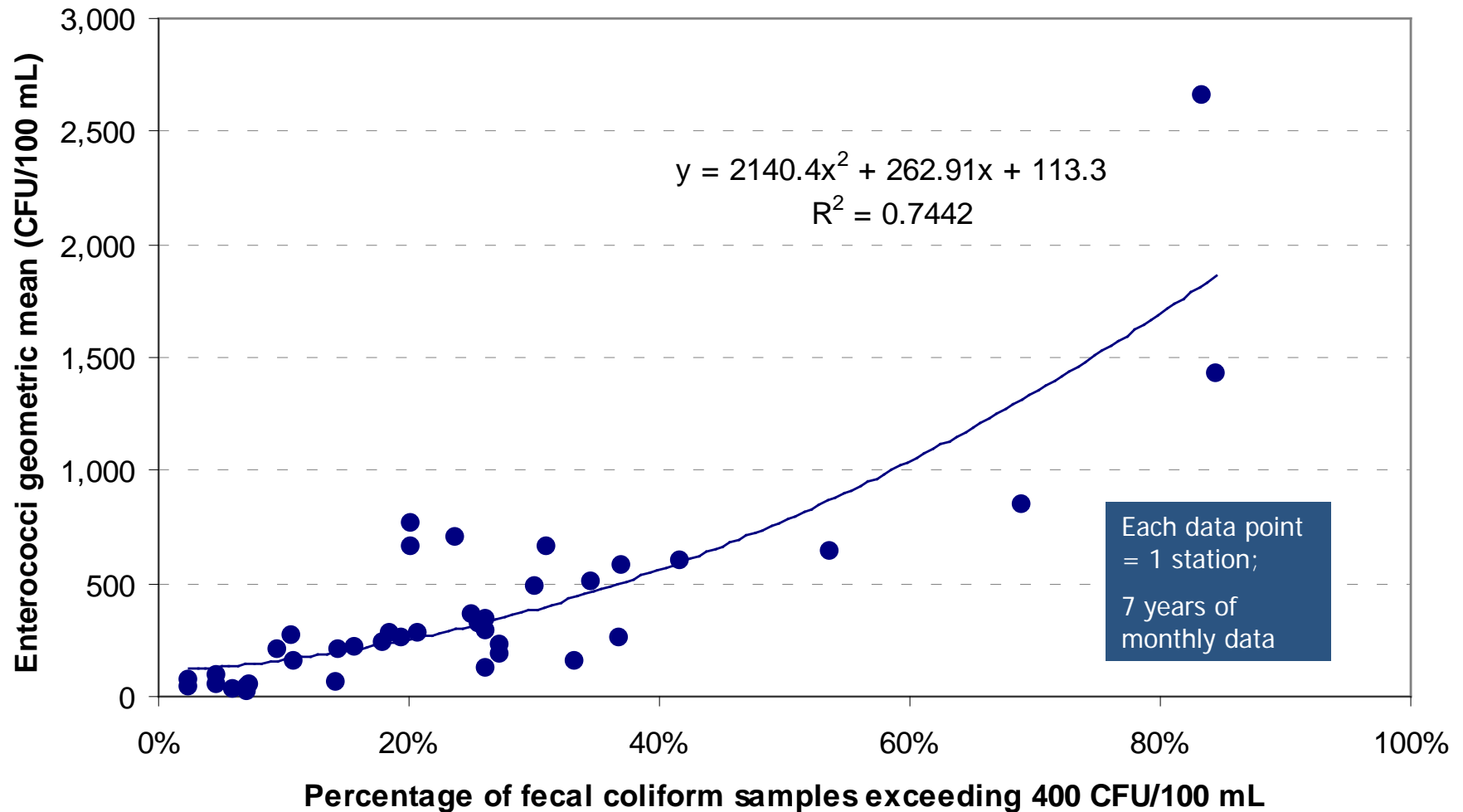
- Incorporate potential human health risk
- Incorporate information on frequency of exceedances of State water quality criterion (400 CFU/100 mL)
- Incorporate binomial test used by Florida DEP to determine statistical significance of criterion exceedances



# Geometric mean fecal coliform counts vs. exceedances of 400 CFU criterion (EPC data)



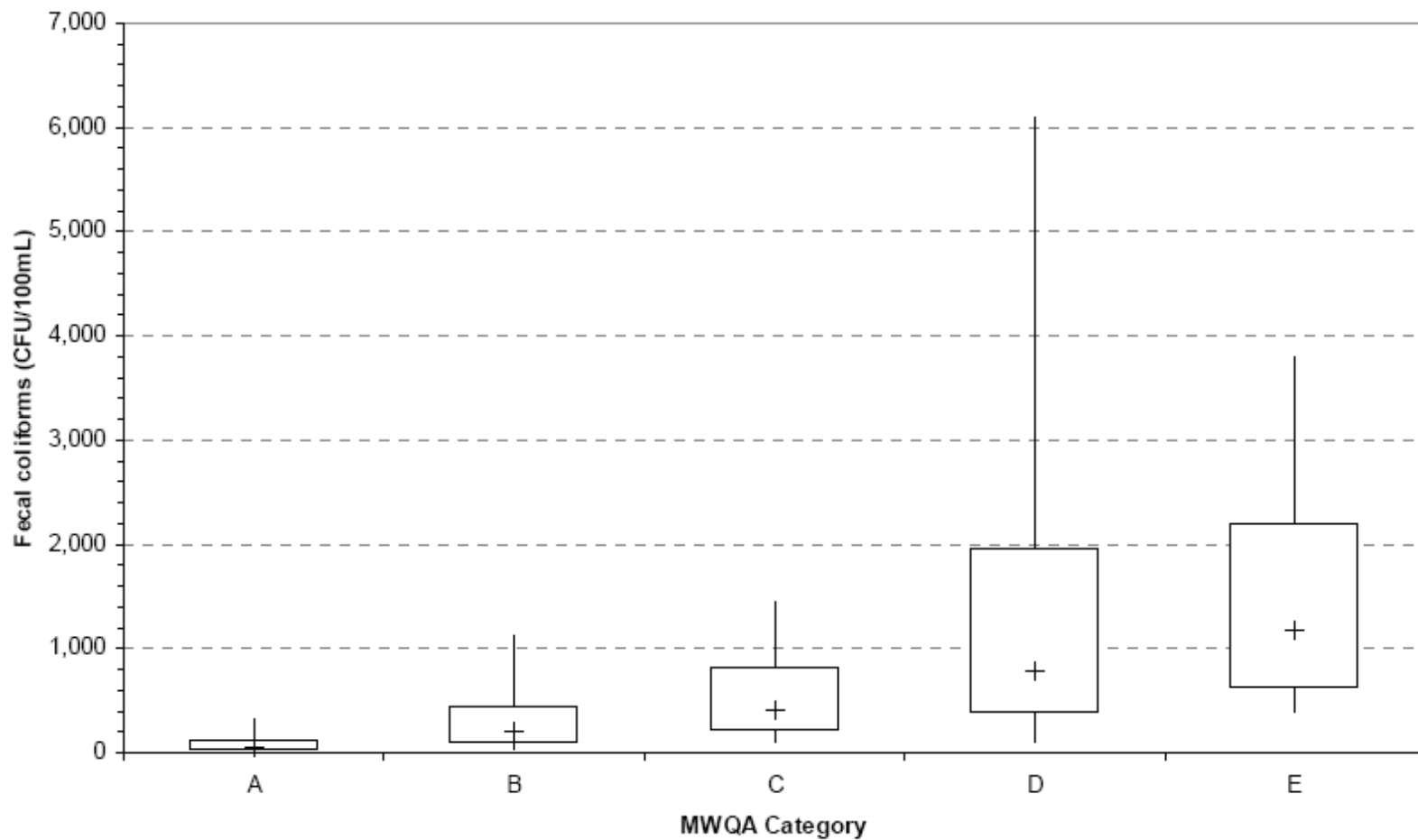
# Geometric mean enterococci counts vs. exceedances of 400 CFU criterion (EPC data)



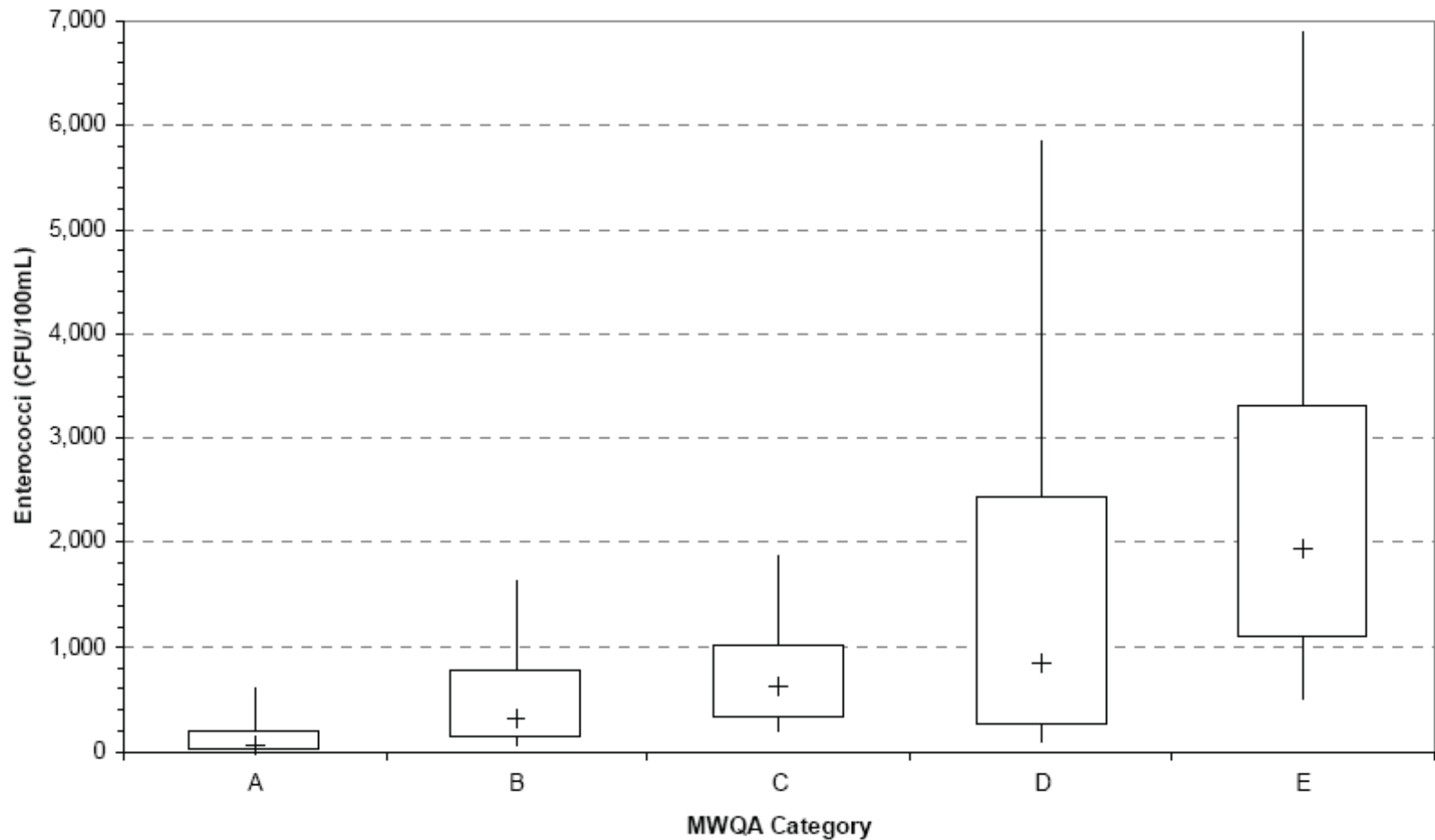
**Table 6. Proposed microbial water quality assessment (MWQA) categories, based on the percentage of samples exceeding the State’s 400 CFU/100 mL fecal coliform criterion. “Break points” separating the MWQA categories are at exceedance frequencies of 10%, 30%, 50% and 75%.**

MWQA Category	Break Point (percentage of samples exceeding the 400 CFU/100 mL fecal coliform criterion)	Range of exceedance frequencies (percentage of samples exceeding the 400 CFU/100 mL fecal coliform criterion) included in category
A	$\leq 10\%$	0% to 10%
B	$> 10\%$	$>10\%$ to 30%
C	$> 30\%$	$>30\%$ to 50%
D	$> 50\%$	$>50\%$ to 75%
E	$> 75\%$	$>75\%$ to 100%

# fecal coliform concentrations in MWQA categories A through E (EPC data, 2001 - 2007)



# enterococci concentrations in MWQA categories A through E (EPC data, 2001 - 2007)



# Bacteria levels and potential human health risk

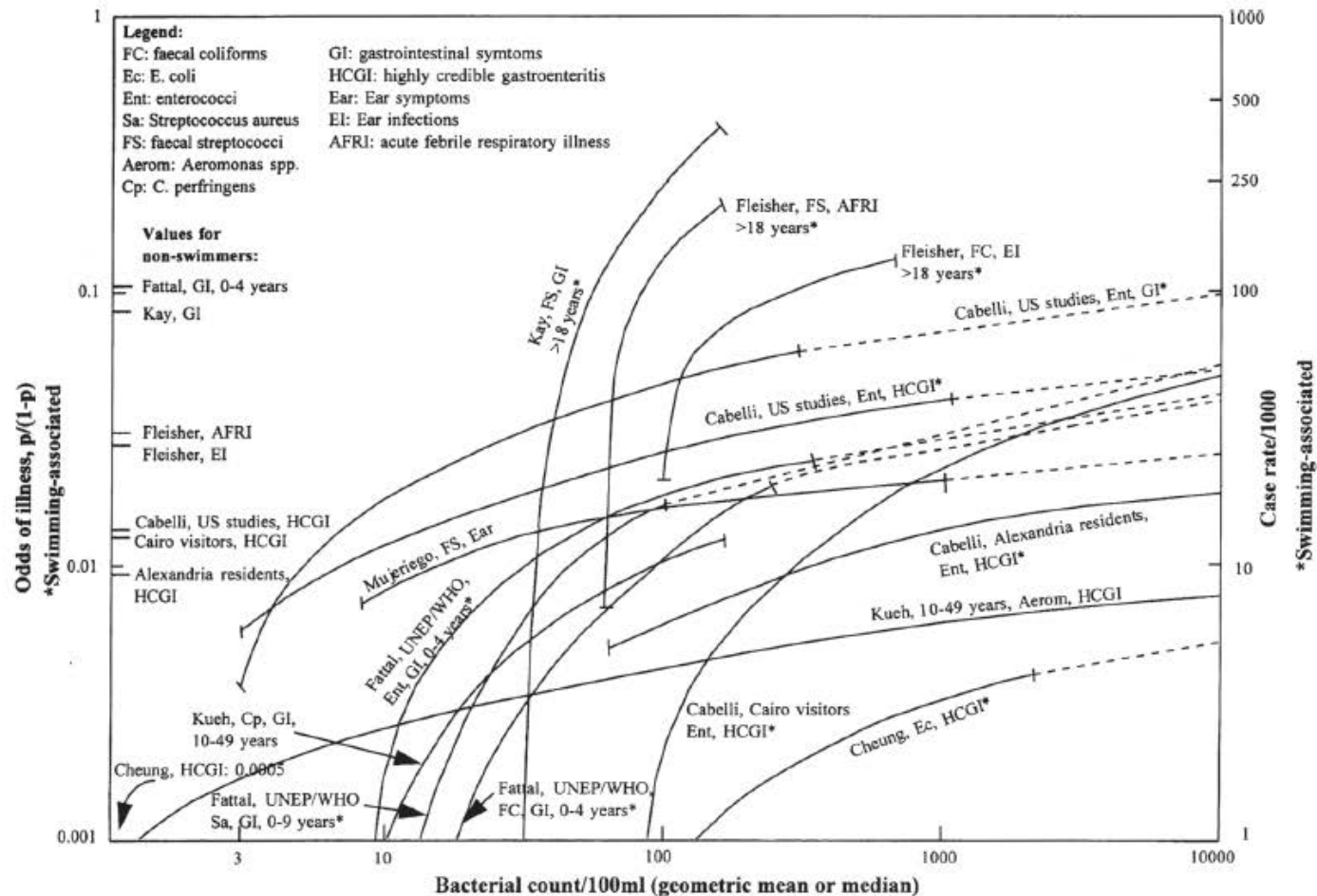
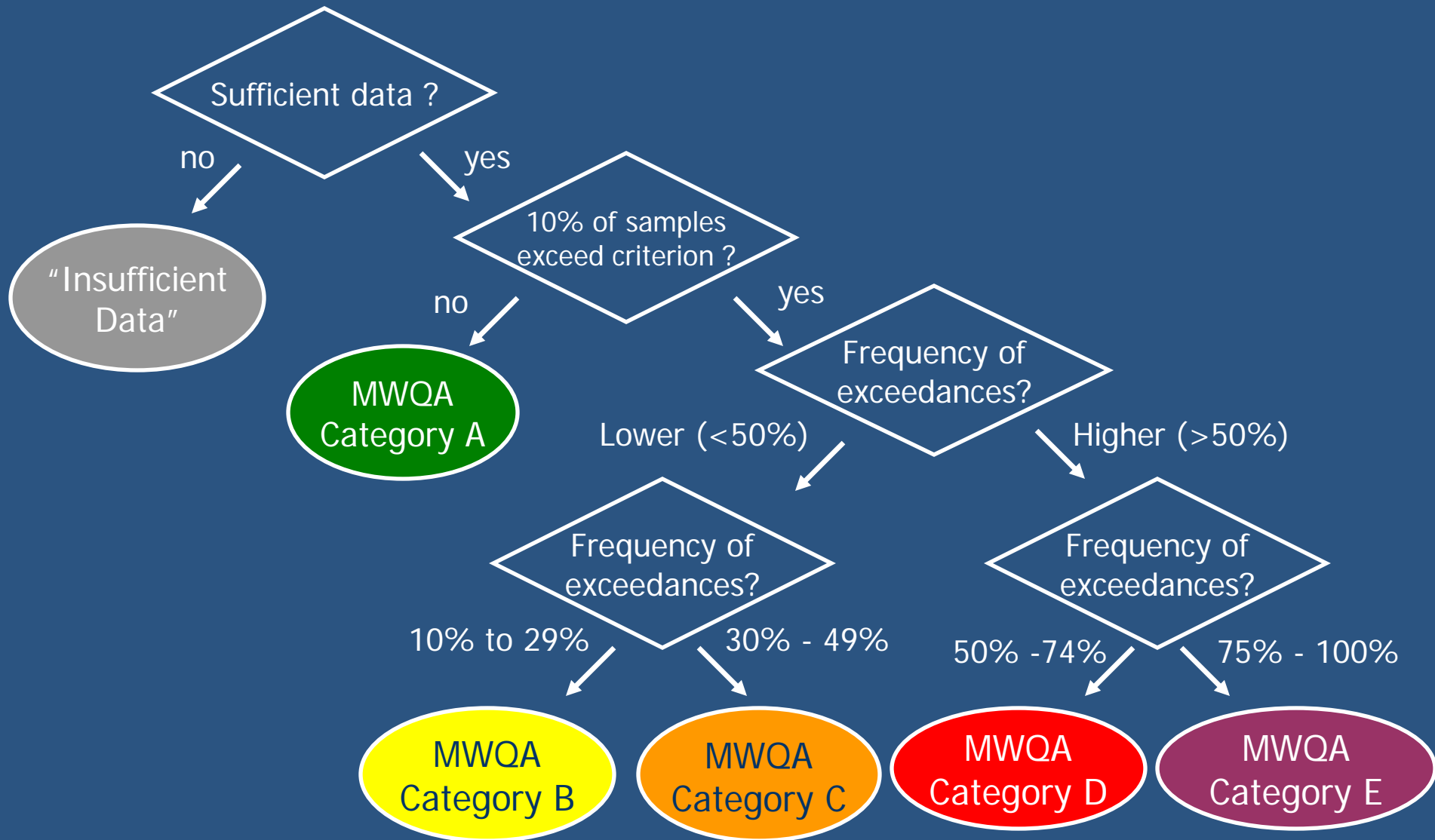


FIGURE 2-2 Relationships of risks of illness in swimmers to the microbial quality of water—fresh recreational waters. Note: Solid lines indicate actual data from original studies whereas dashed lines are extrapolations of data by Prüss (1998) or Pike (1991). SOURCE: Reprinted, with permission, from Prüss, 1998 © Oxford University Press.



# Decision Tree to Define Microbial Water Quality Assessment (MWQA) Categories Based on Exceedances of 400 CFU/100 mL Fecal Coliform Criterion



## Step 2

- Add information from contaminant source surveys (CSS) and, when available, MST
- Classify sites based on the likelihood of fecal contamination that would pose human health risks, using CSS assessment categories and MST data

# CSS assessment categories (likelihood of fecal contamination posing human health risks)

- **Very Low:** No visual evidence of potential sources of human pathogens; natural environment; no or minimal anthropogenic land uses; wildlife present (any density)
- **Low:** Low density agricultural and residential sources, including pets, livestock (without direct access to surface waters), or poultry operations; residences on septic systems
- **Moderate:** Urban stormwater sources (including pet waste) present; well-functioning wastewater infrastructure (both sewer and septic); episodic/low volume sanitary sewer overflows (SSOs) reaching surface waters; moderate-density livestock with little direct access to surface waters; Class A residual and/or septage spreading areas may be present
- **High:** Major stormwater outfalls present; history of failing wastewater infrastructure (central sewer or onsite systems); episodic or chronic/high volume SSOs reaching surface waters; concentrated livestock without direct access to surface waters; residual/septage spreading (Class B)
- **Very High:** Current failing wastewater infrastructure; chronic/high volume SSOs reaching surface waters; concentrated livestock with direct access to surface waters; evidence of direct sewage inputs (e.g., confirmed illicit discharges)

# Classification matrix ("Annapolis protocol") approach

		MWQA group (based on binomial assessment of frequency of 400 CFU/100 mL fecal coliform exceedances)					Exceptional Circumstances (e.g., sewer line break) <sup>c</sup>
		A (≤ 10%)	B (>10% - 30%)	C (>30% - 50%)	D (>50% - 75%)	E (>75%)	
Contaminant source survey (CSS) assessment category (likelihood of fecal contamination posing human health risks)	1. Very Low	A1	B1	C1 <sup>a</sup>	D1 <sup>a</sup>	E1 <sup>a</sup>	Immediate Action
	2. Low	A2 <sup>b</sup>	B2	C2	D2 <sup>a</sup>	E2 <sup>a</sup>	
	3. Moderate	A3 <sup>b</sup>	B3	C3	D3	E3	
	4. High	A4 <sup>b</sup>	B4 <sup>b</sup>	C4	D4	E4	
	5. Very High	A5 <sup>b</sup>	B5 <sup>b</sup>	C5 <sup>b</sup>	D5	E5	
Exceptional Circumstances (e.g., sewer line break) <sup>c</sup>		Immediate Action					

## Notes:

- These outcomes imply that the CSS may be providing an overly optimistic rating of water quality, or the fecal coliform sources in the area may be relatively low-risk or primarily environmental (e.g., wildlife, sediments, soils, vegetation), and the cause(s) of the discrepancy should be verified.
- These outcomes imply that the fecal coliform indicator may be providing an overly optimistic MWQA rating, or the CSS may be providing an overly negative assessment, and the cause(s) of the discrepancy should be verified.
- As explained by WHO (2003), exceptional circumstances involve acute situations known to be associated with higher public health risks, such as sewer line breaks and other SSOs that contaminate surface waters, which require immediate remedial action.

# Phased CSS Investigation Levels

- **Phase 1** – basic (screening-level) water quality and land use analyses, and boots-on-the-ground source evaluation. (Applied automatically to MWQA Group B, and to Group A if needed.)
- **Phase 2** – adds more intensive indicator monitoring and CSS. (Applied automatically to MWQA Group C, and to Group A or B if needed.)
- **Phase 3** – adds appropriate source tracking (MST) tools, as necessary. (Applied automatically to MWQA groups D and E, and to others if needed.)

Track and report outcomes annually to the  
public and elected officials  
(using the color-coded classification matrix)

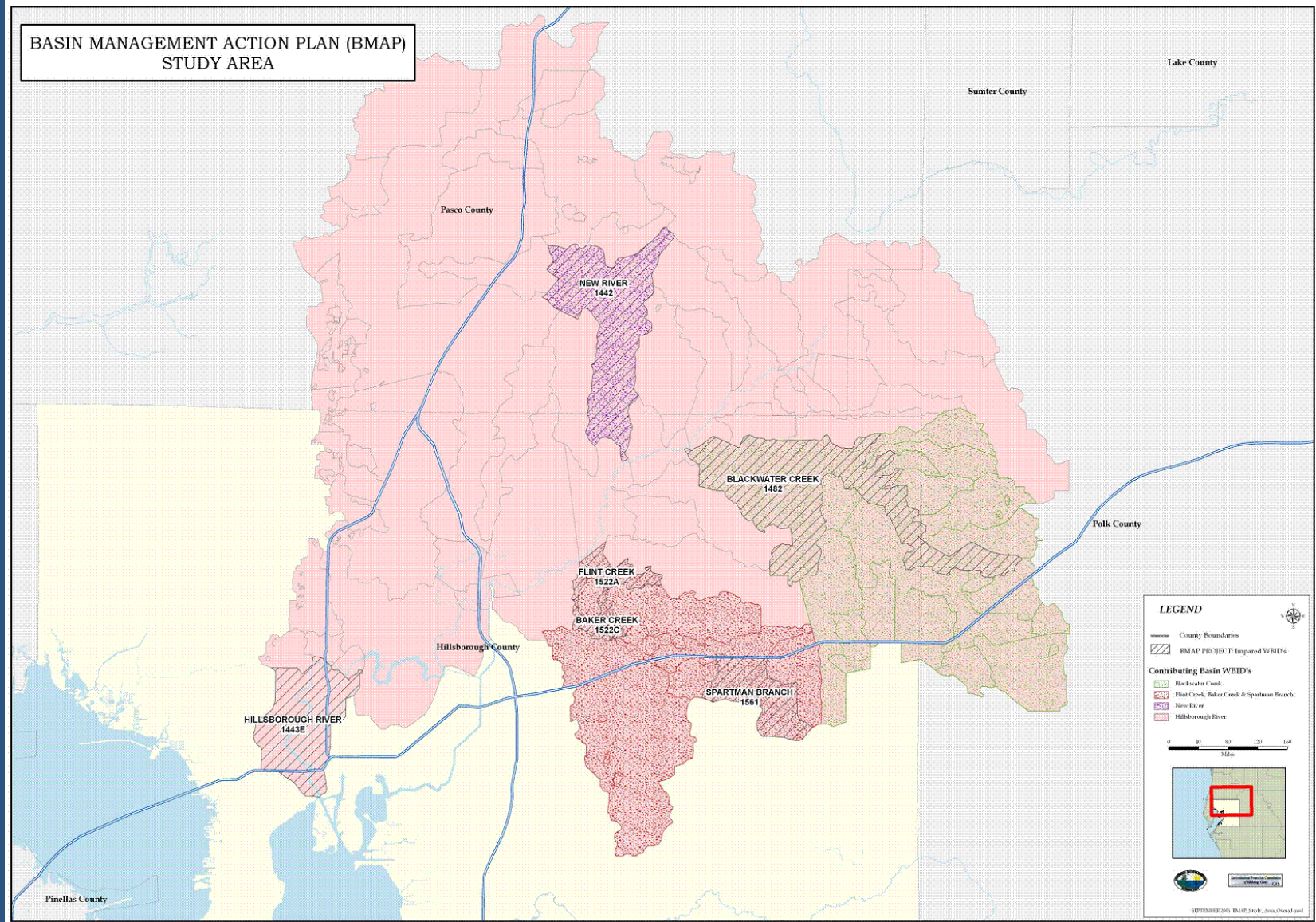
Allows water  
quality  
conditions to be  
tracked over  
time and across  
monitoring  
locations

	Monitoring Location			
Year	1	2	3	4
2004	D4	A1	B1	C3
2005	C4	B1	B1	C3
2006	D4	B1	C1	B3
2007	C3	A1	B1	B2
2008	C3	A1	A1	B2



# An application to 6 pilot WBIDs

(In the Hillsborough River portion of the Tampa Bay watershed)



Using the  
classification  
matrix to  
prioritize  
WBIDs  
(and sites  
within  
WBIDs)  
for  
management  
action

Sub-Basin	WBID	Monitoring Location	Classification Matrix Outcome	Human Fecal MST Markers		Ruminant Fecal MST Marker	
				No. Sampling Dates	% Dates Marker(s) Detected	No. Sampling Dates	% Dates Marker Detected
Lower Hillsborough River	1443E	DHR4A	D5	5	100 %	5	20 %
		HR3	D4	5	80 %	5	20 %
		DHR5	D4	2	50 %	2	0 %
		DHR8	D3	4	50 %	4	0 %
		HR2	C4	3	67 %	3	0 %
		HR4	C4	3	67 %	3	0 %
		DHR7	C3	5	60 %	5	60 %
		DHR6	C3	3	33 %	3	0 %
		HR1 REF	B3	3	100 %	8	0 %
		HR5	B3	5	60 %	5	0 %
		DHR10	A4	1	100 %	1	0 %
Blackwater Creek	1482	BW3	D4	4	75 %	1	0 %
		BW2	D4	3	67 %	3	100 %
		DBW4	A3	1	100 %	1	0 %
		BW5A	A3	2	50 %	2	0 %
		BW1 REF	A2	7	43 %	6	17 %
Baker Creek	1552C	BK2	D3	6	67 %	6	67 %
		BK1	C3	7	57 %	7	29 %
		BK3	C2	2	50 %	2	0 %
		BK4	A4	1	100 %	1	0 %
		BK5	A3	2	100 %	2	0 %
Flint Creek	1552A	FL2	C2	6	50 %	6	0 %
		FL1	C2	5	20 %	5	0 %
		FL3	A3	6	33 %	6	0 %
Spartman Branch	1561	SB1	D3	6	17 %	6	0 %
		SB2	D2	5	20 %	6	0 %
		DSB3	C2	2	0 %	2	50 %
		SB3	B2	4	25 %	4	0 %
New River	1442	NR2	C3	7	29 %	7	29 %
		NR1	C2	2	0 %	2	50 %
		NR3	C2	2	0 %	2	50 %

# Using the detailed CSS information to suggest management actions for specific sites within a WBID (LHR examples)

Sub-Basin	WBID	Monitoring Location	Classification Matrix Outcome	Fecal Source(s) of Concern	% of Sampling Dates Markers Detected	Most Probable Source Categories and Recommended Management Actions
Lower Hillsborough River	1443E	DHR4A	D5	Human	100 %	Septic and possibly sanitary sewer; confirm and address specific source locations and provide public outreach explaining potential presence of health risk at site
		HR3	D4	Human	80 %	Septic and sanitary sewer; confirm and address specific source locations and provide public outreach explaining potential presence of health risk at site
		DHR8	D4	Human	50 %	Multiple sources suspected; identify and address specific source locations; provide public outreach explaining potential presence of health risk at site
		DHR5	D3	Human	50 %	Multiple sources suspected; identify and address specific source locations; provide public outreach explaining potential presence of health risk at site
		HR2	C4	Human	67 %	Sanitary sewer and stormwater; identify and address specific source location(s)
		HR4	C4	Human	67 %	Sanitary sewer and stormwater; identify and address specific source location(s)
		DHR7	C3	Human	60 %	Multiple sources suspected; identify and address specific source locations
		DHR6	C3	Human	33 %	Unknown; identify and address specific source location(s)
		HR1 REF	B3	Human	100 %	Homeless camp, sanitary sewer and possibly stormwater; confirm and address specific source locations
		HR5	B3	Human	60 %	Sanitary sewer; confirm and address specific source locations
		DHR10	A4	Human	100 %	Multiple sources suspected; identify and address specific source locations

This conceptual approach can also be used to prioritize sites for more intensive investigation, at a county-wide (or larger) scale...



# Initial county-wide site prioritization

(using MWQA  
scores based on  
fecal coliform data  
from EPC of  
Hillsborough  
County monitoring  
stations; 2001-  
2007 data)

Location	Sta. No.	No. Samples (N)	% >400 CFU/ 100 mL ( $\hat{P}$ )	MWQA category	Geometric mean fecal coli. (CFU/ 100 mL)	Geometric mean enterococci (CFU/ 100 mL)
Turkey Creek at SR 60	111	77	84%	E	1,212.0	1,426.0
Bullfrog Creek at Symmes Rd	132	78	83%	E	1,134.8	2,658.9
Sweetwater Creek at Hillsborough Ave	104	84	69%	D	786.4	849.9
Delaney Creek at 36th Ave	138	84	54%	C	443.3	642.2
Delaney Creek at US 41	133	84	42%	C	376.7	596.8
Little Manatee River at CR 579	140	84	31%	B	283.6	664.9
English Creek at SR 60	154	83	30%	B	266.6	487.7
Mill Creek at I-4	149	81	37%	B	258.0	578.7
Little Manatee River at US 301	113	84	20%	B	250.8	664.0
Rocky Creek at Hillsborough Ave	103	84	26%	B	242.6	285.2
Hillsborough River at Sligh Ave	152	84	27%	B	239.8	182.8
Bullfrog Creek at US 41	144	84	37%	B	233.7	254.7
Little Manatee River at CR 674	129	84	24%	B	218.5	704.0
Baker Creek at Thonotosassa Rd	107	84	35%	B	216.5	508.7
Hillsborough River at Rowlett Pk	105	84	26%	B	208.7	127.7
Rocky Creek at Waters Ave	141	84	26%	B	205.8	343.4
Hillsborough River at Columbus Ave	137	84	33%	B	195.5	153.7
Flint Creek at US 301	148	84	25%	B	191.2	359.8
Double Branch Creek at Hillsborough Ave	101	84	27%	B	185.1	222.6
Blackwater Creek at SR 39	143	82	21%	B	156.9	284.2
Trout Creek at CR 581	145	65	18%	B	152.9	281.8
Sweetwater Creek at Anderson Rd	142	84	20%	B	152.2	764.8
Turkey Creek at Durant Rd	151	82	26%	B	142.0	322.6
Alafia River South Prong at Bethlehem Rd	139	83	16%	B	138.8	215.7
Alafia River at Bell Shoals Rd	114	84	18%	B	136.4	240.7
Cypress Creek at CR 581	120	72	19%	B	132.9	261.9
Alafia River at US 301	153	83	14%	A	152.7	209.7
Hillsborough River at US 301	108	83	11%	A	101.7	159.4
Alafia River North Prong ab confluence	115	84	11%	A	100.4	267.3
Alafia River South Prong ab confluence	116	84	10%	A	80.6	207.3
Channel A at Hillsborough Ave	102	84	14%	A	63.2	63.1
L. Thonotosassa at Flint Creek	118	82	2%	A	46.8	68.5
Hillsborough River at Fowler Ave	106	84	5%	A	46.6	96.4
TBC at Fowler Ave	146	82	7%	A	40.5	51.0
Palm River at US 41	109	84	7%	A	39.1	36.4
Little Manatee River at US 41	112	84	5%	A	34.6	55.1
L. Thonotosassa middle	135	82	2%	A	32.9	41.7
TBC at MLK Blvd	147	84	6%	A	31.9	31.0
Palm River at SR 60	110	84	7%	A	25.4	21.8

# This is an initial application of the “Annapolis protocol” approach in Florida

Some issues not yet addressed are:

- How to deal with tidal / estuarine sites ?
  - *fecal coliform die-off in estuarine waters may affect site ratings in tidal streams and rivers*
  - *a different set of MWQA categories may be needed*
- How to deal with Class I and Class II waters ?
  - *different MWQA categories probably necessary*
  - *additional indicators (e.g., Crypto, Giardia) may also be needed*



# Summary of support-tool application

- Within WBIDs, assign each long-term monitoring site to an MWQA category (A through E), based on exceedance frequency of the 400 CFU/100 mL fecal coliform criterion
- Also assign each site to an initial CSS category (1 through 5)
- Use classification matrix to prioritize WBIDs (and multiple sites within WBIDs, if applicable) for follow-up work, based on initial MWQA and CSS categories.
- Apply phased monitoring approach to identify potential hot spots within WBIDs and investigate their fecal contamination sources. Revise the initial MWQA and CSS categories, if necessary, based on results of this step.
- Correct fecal contamination sources that pose human health risk, as they are identified
- Continue monitoring, and provide annual report documenting changes in MWQA and CSS scores that occur over time in response to management actions

# Benefits of the approach

- Can be used as a screening tool to prioritize bacterial impairments at the station and WBID levels, based on the level of impairment
  - Enables targeting of efforts to identify and address sources, resulting in a significant saving of time and money
- Uses a phased approach for identifying sources, based on the level of impairment and estimated likelihood of human health risk
  - Increases efficiency and effectiveness in identifying sources, resulting in a significant saving of time and money
- Encourages active stakeholder involvement in identifying potential sources
  - Helps to coordinate and leverage available resources in the identification of sources, and assist in building consensus on addressing sources
- Can be used to evaluate the “sufficiency of effort” of the projects identified in the BMAP to restore water quality
  - Helps ensure that projects identified in BMAP can be expected to address the impairment and in the most effective way
  - Helps reduce the possibility of 3rd party challenges to the BMAP

Thanks !

Questions ?

Gerold Morrison  
[TerraCeia@Tampabay.rr.com](mailto:TerraCeia@Tampabay.rr.com)  
941-723-8980

## Some additional reading...

- National Research Council (NRC). 2004. Indicators for waterborne pathogens. National Academy Press, Washington, DC
- Rose, J.B., and others. 2001. Healthy beaches Tampa Bay: Microbiological monitoring of water quality conditions and public health impacts. Tampa Bay Estuary Program Technical Report #03-01. TBEP. St. Petersburg, FL
- U.S. Environmental Protection Agency (U.S. EPA). 2007. Report of the experts scientific workshop on critical research needs for the development of new or revised recreational water criteria. EPA 823-R-07-006. Washington, DC
- World Health Organization (WHO). 2003. Guidelines for Safe Recreational Water Environments. Volume 1. Coastal and Fresh Waters. WHO, Geneva, Switzerland



# Microbial Source Tracking: The Science

**Valerie J. Harwood**  
**Department of Biology**



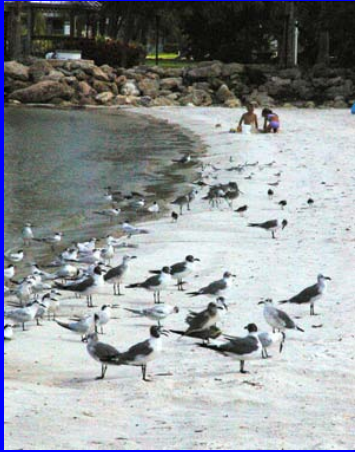
FDEP Tallahassee Meeting 06/10/08



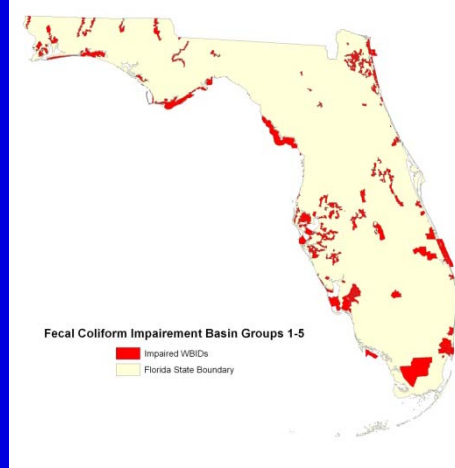
# Part I: Some Background



# The Central Question of Microbial Source Tracking (MST)



?



?



?



?





# Indicator Organisms (IOs) Are Generally Nonpathogenic!



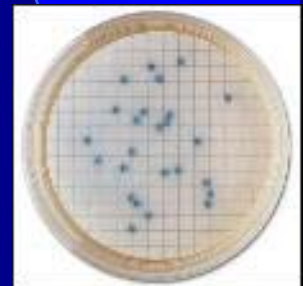
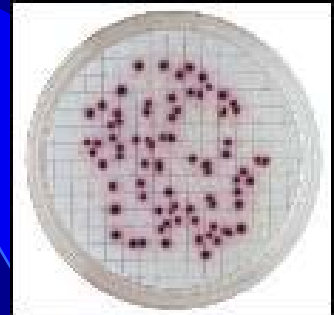
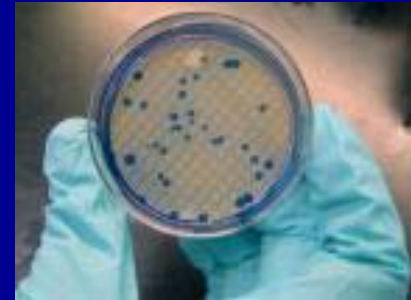
- Meant to act as a warning signal of fecal pollution and increased risk of associated pathogens in water





# Current Recognized IOs for Recreational & Shellfishing Water Quality

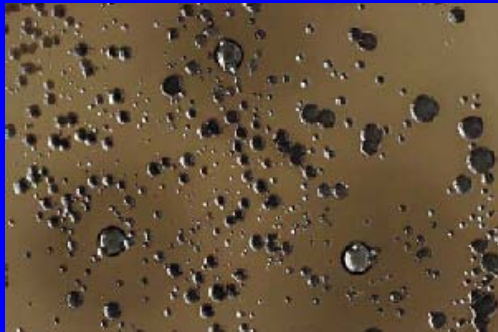
- Fecal coliforms – thermolerant coliforms of fecal origin (Florida & other states)
- *Escherichia coli* – correlated w/risk of gastroenteritis in freshwater (U.S. EPA)
- Enterococci –relatively salt-tolerant; correlated w/risk of gastroenteritis in freshwater & saltwater (U.S. EPA)



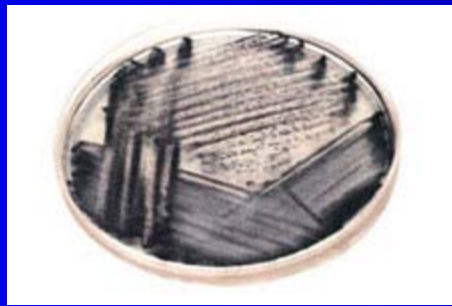


# Water Quality Standards Must Protect Against Many Types of Pathogens

**Bacterial pathogens**

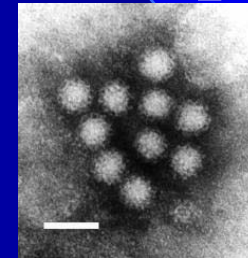


*Salmonella*



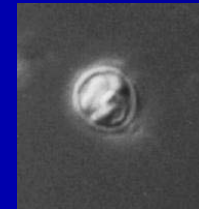
*E. coli* O157:H7

**Viral pathogens**



*Norovirus*

**Protozoan pathogens**



*Cryptosporidium*



*Giardia*

# Basis for Continued Use of Indicator Bacteria to Protect Human Health

- Correlation of *E. coli* concentration with risk of gastroenteritis

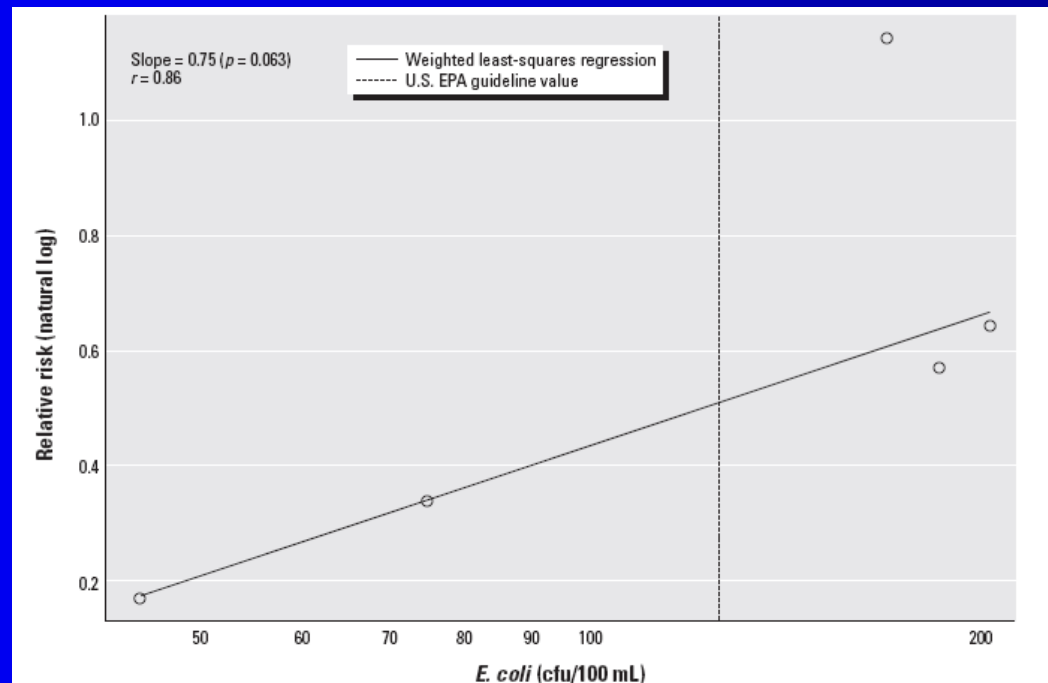


Figure 2. Scatterplot and weighted regression line (weighted by the inverse of the standard error of the natural log relative risk) of the natural log relative risk of GI illness from freshwater studies as a function of *E. coli* density.

# Risk

- Epidemiology studies show increased risk of gastroenteritis from recreational water use of ~ 8 individuals/1,000 at these levels.
- Enterococci 61 CFU/100 ml freshwater & 104 CFU/100 ml marine water
- *E. coli* 235 CFU/100 ml
- ~equivalent to 400 CFU/100 ml fecal coliform





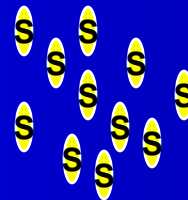
# Why Do We Need to Know the Source?

- Sources vary from highest risk (human sewage)...
- ...to definite risk (cattle, poultry)
- ...to unknown risk (wildlife, pets, sediments, soils, stormwater)
- Can't remediate or implement w/o knowledge of dominant source(s)

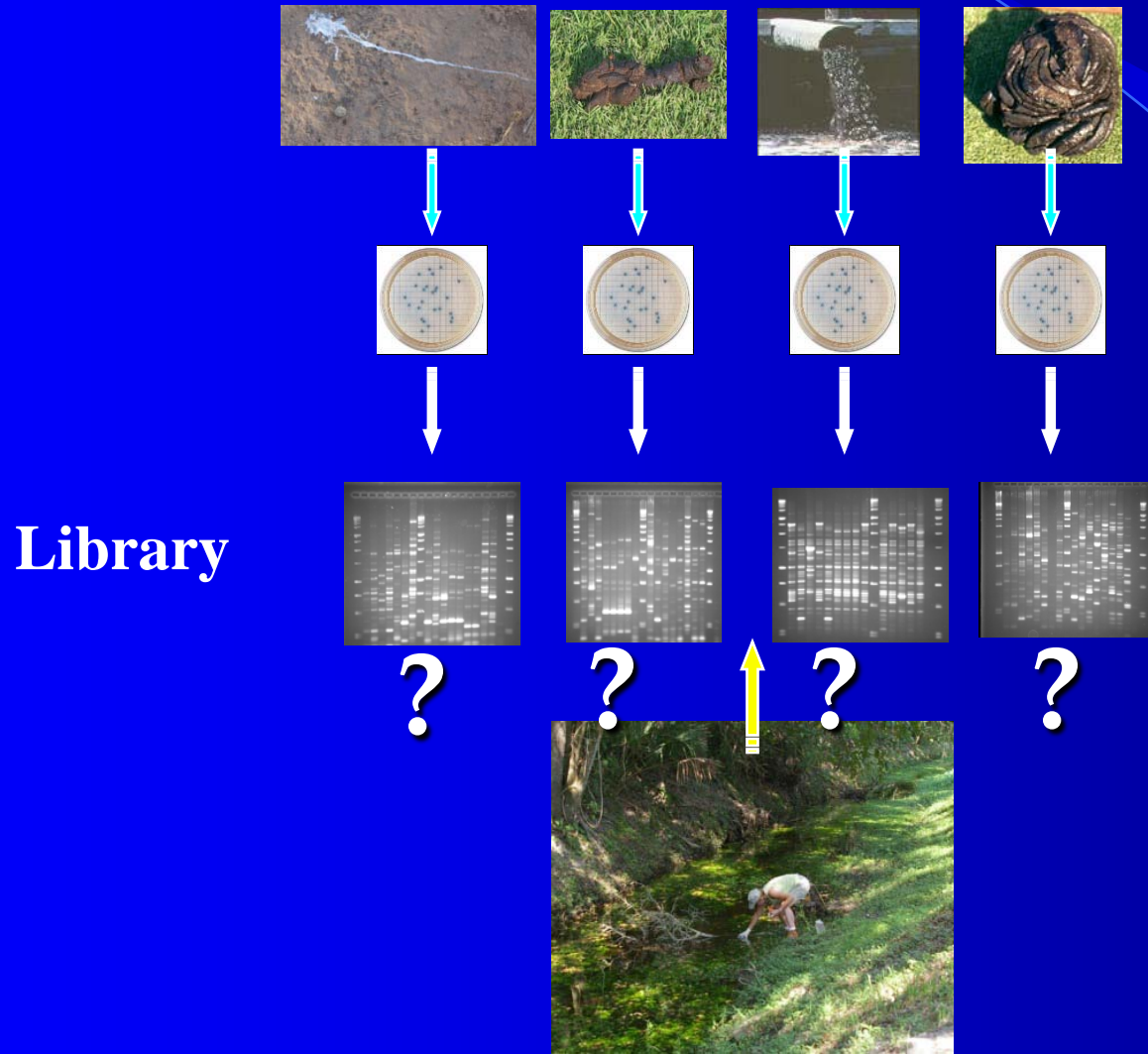


# The Central Hypothesis of MST

- Certain microbial species or types are associated with the gastrointestinal tract of specific animals hosts.
- This association can be used to “track” the fecal microorganism back to its host.



# Library-Dependent MST Approach



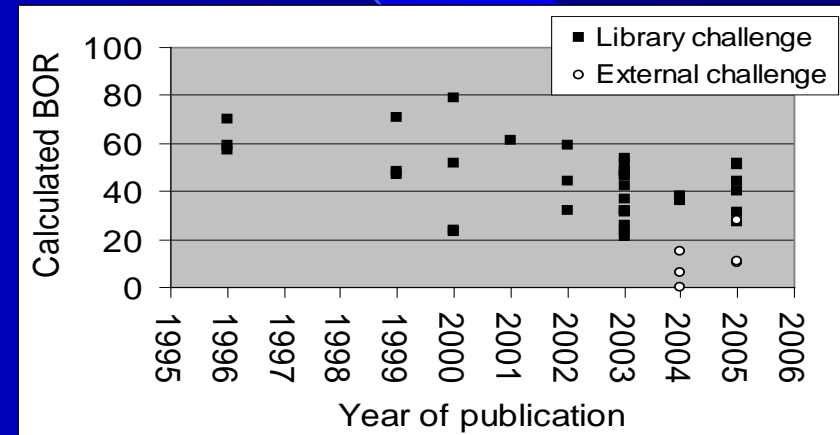
# Library-Dependent Methods

Determine percentage of isolates assigned to each source category (proportional contribution of sources?)

- **Representative sampling!**
  - Host species impacting watershed
  - Feces & sewage
  - Water
  - “Declone” library at sample level
- **Geographic range**
- **Temporal variability**
- **Sensitivity**
- **Specificity**

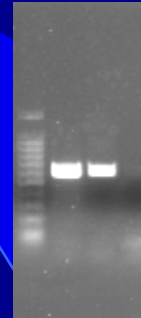
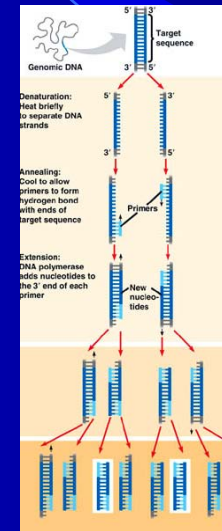
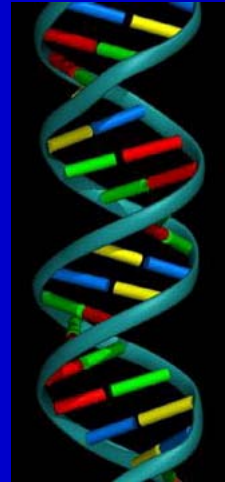


Use samples that are **NOT** represented in the library



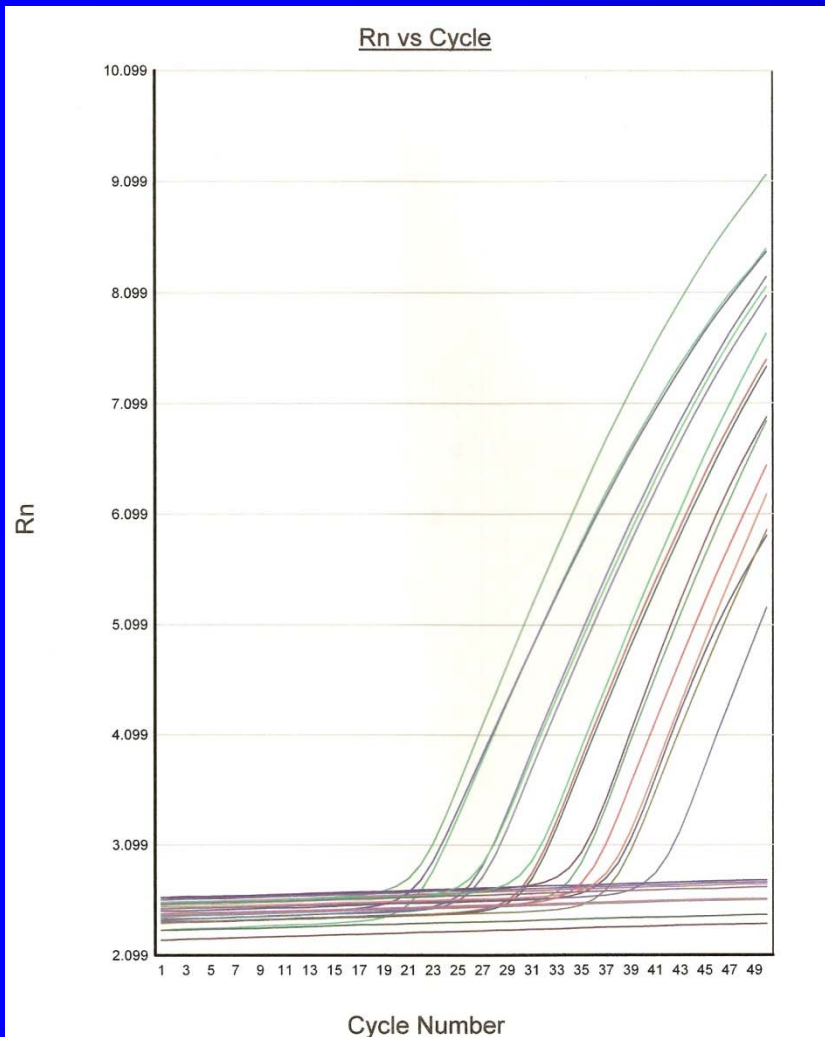


# The Library-Independent MST Approach – Based on DNA “Markers”

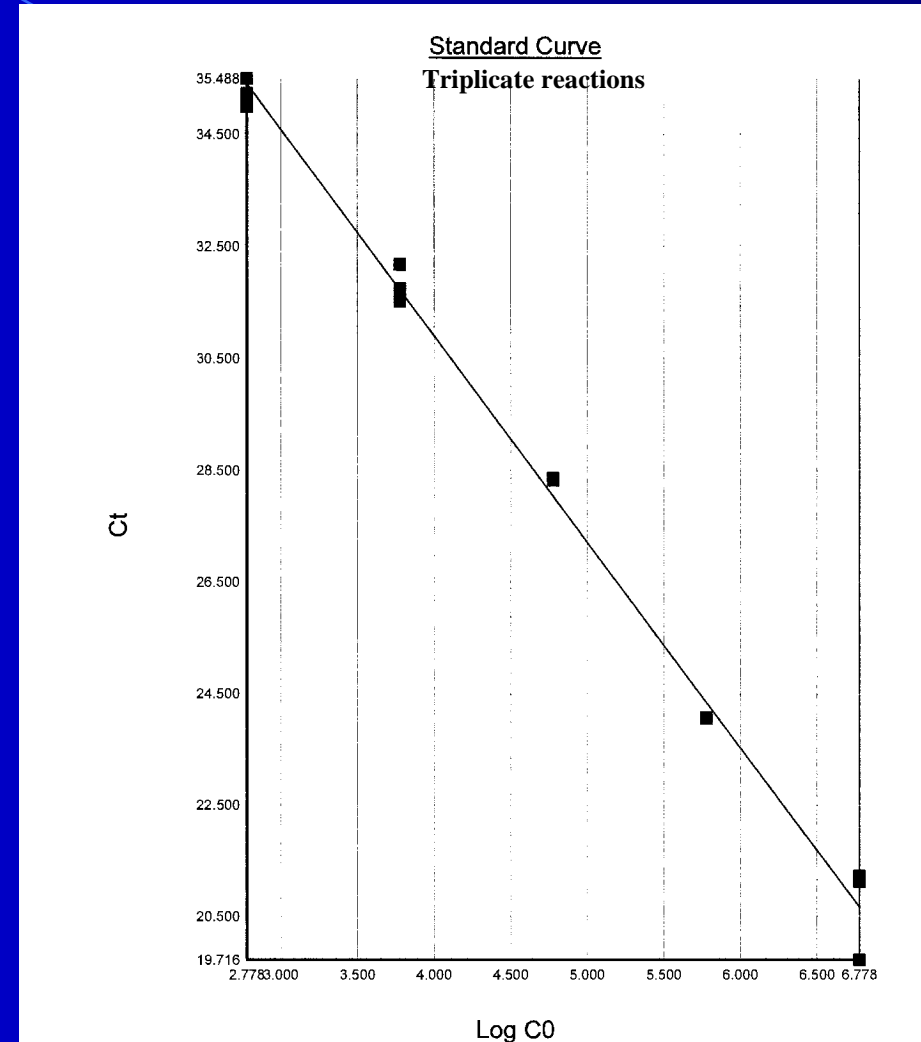


- Polymerase chain reaction targets a specific gene & copies it
- Forensic-type evidence for contamination source

# Quantitative (Taqman) PCR for Human Polyomaviruses

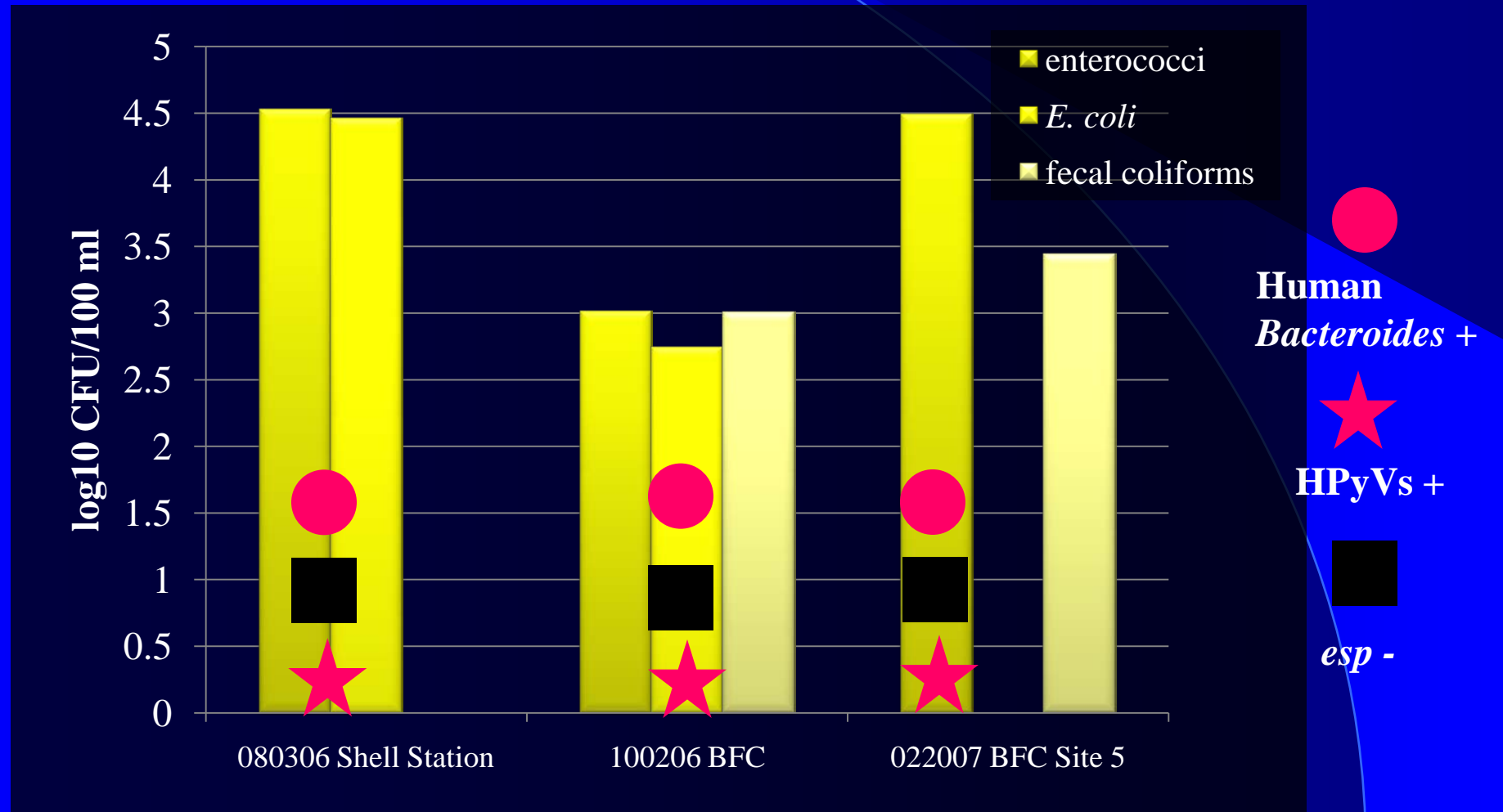


LOD ~10 viruses



Concentration in sewage  $10^3$ - $10^4$ /ml

# Field Validation of Human *Bacteroides* Marker & HPyVs at Septic System Overflows

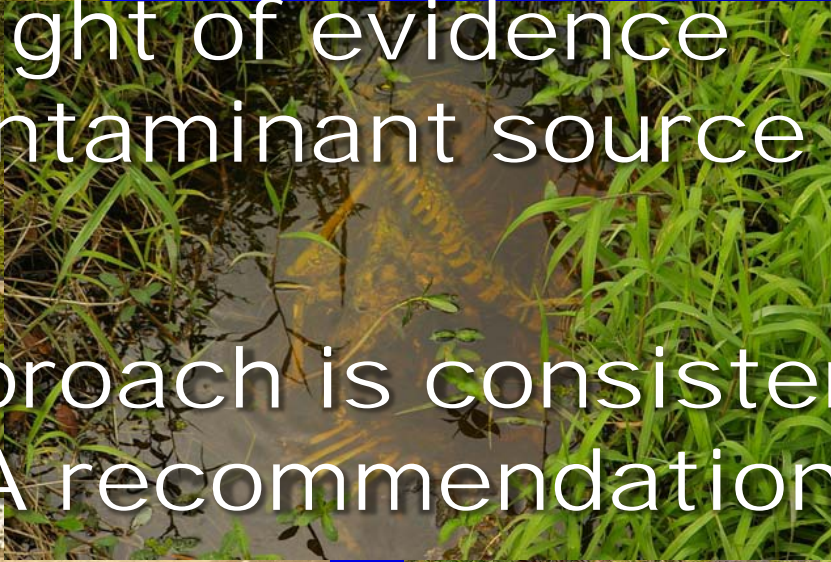
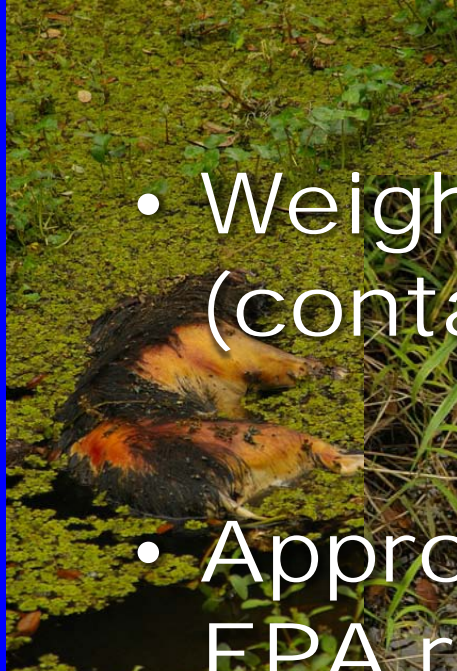




# Part II: Merging Science with Application

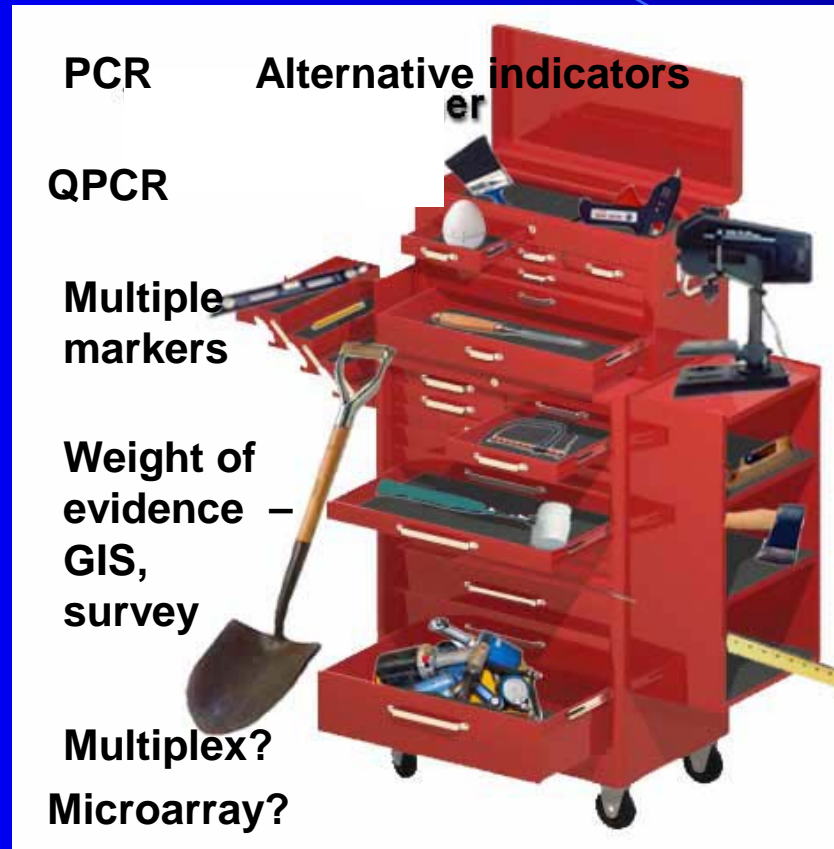
- Weight of evidence (contaminant source survey)
- Approach is consistent with EPA recommendations

- Collaboration with stakeholders and buy-in





# Weight of Evidence Approach

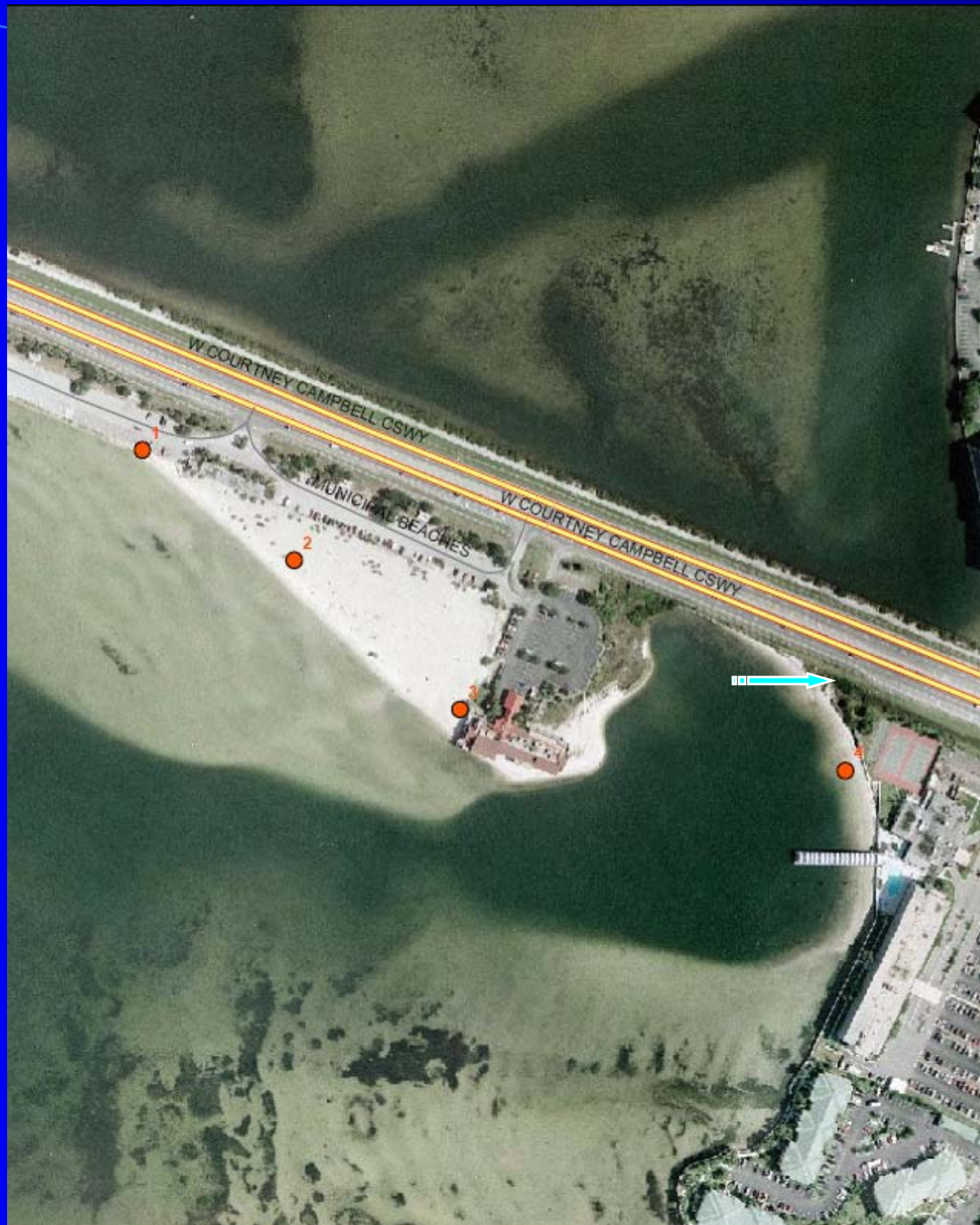


# Walking the Walk

**Knowledge of watershed and potential sources is crucial to successful MST application**

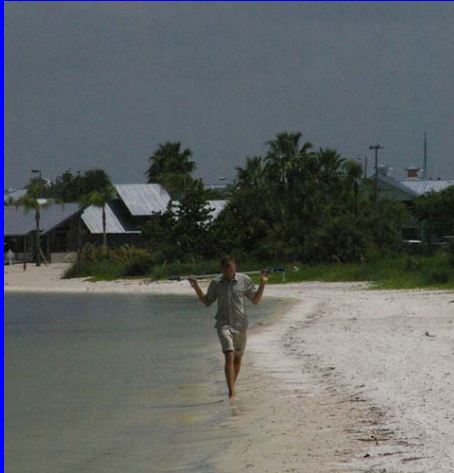
- **Maps – infrastructure, land use**
- **Partnerships – FDEP, DOH, DoACS; farmers; land-owners, residents**
- **Walk &/or boat the water body!**



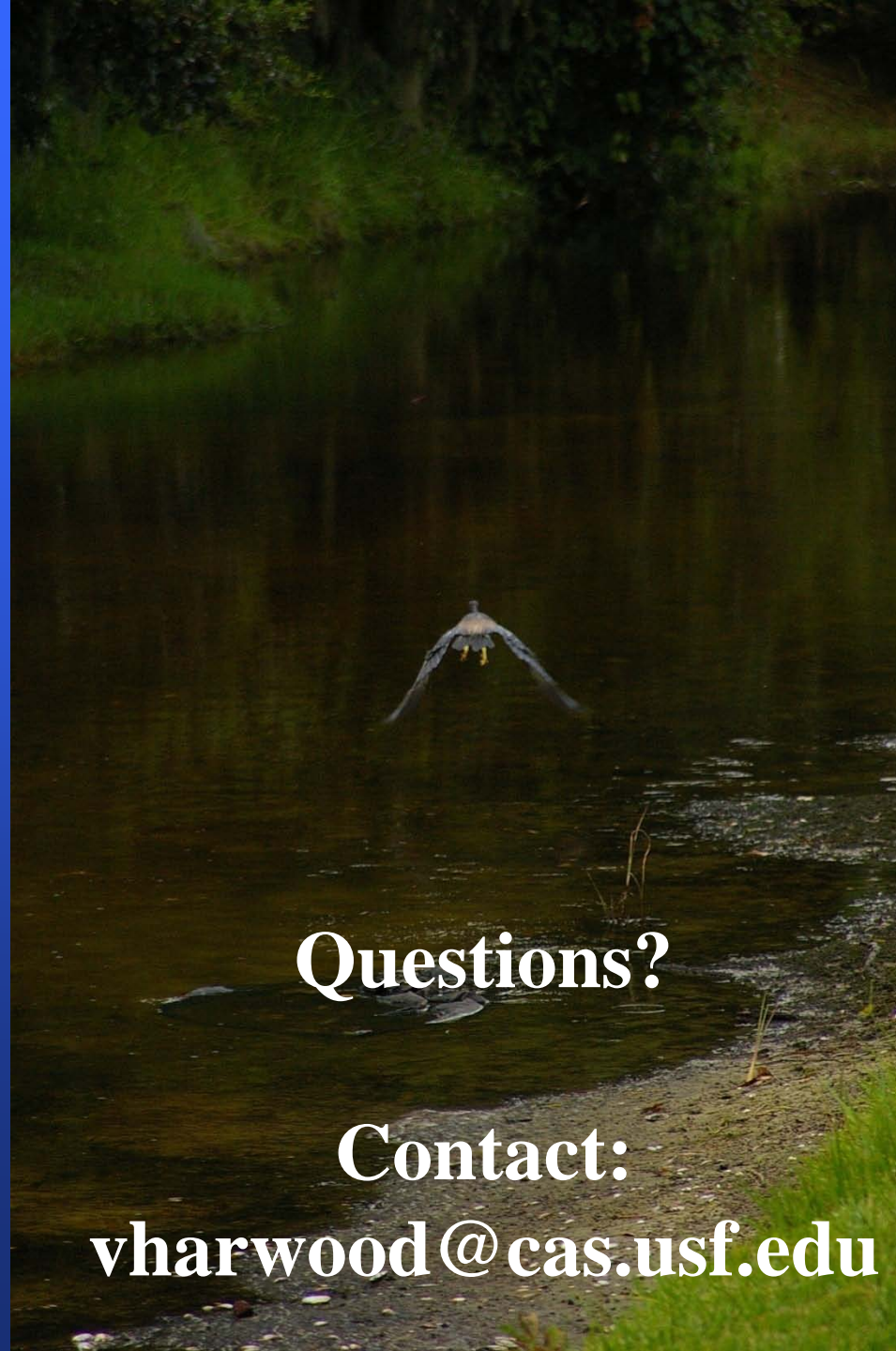




# Thanks to the Lab Crew!







Questions?

Contact:  
[vharwood@cas.usf.edu](mailto:vharwood@cas.usf.edu)



# **Recommendations for Future Work**

# Hillsborough River Watershed

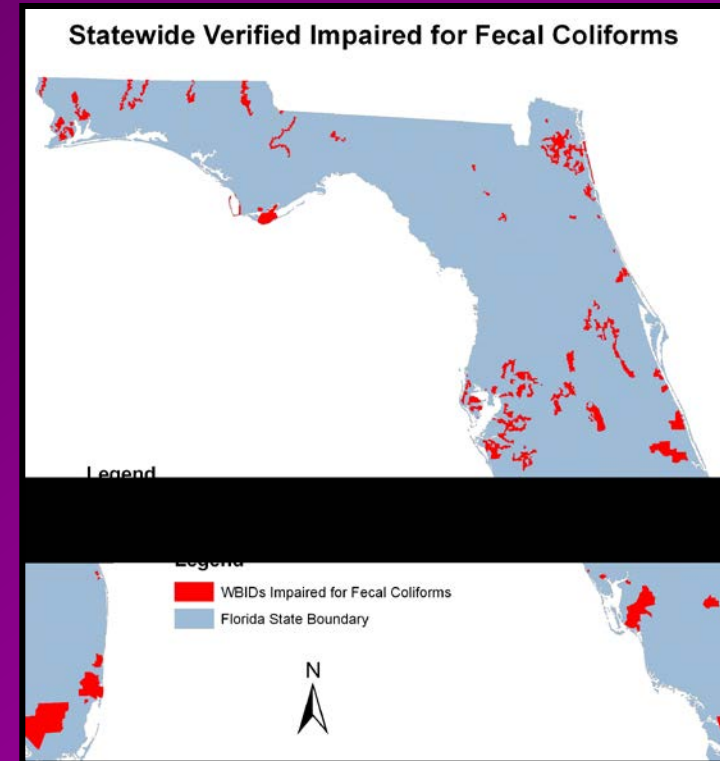
- Evaluate the sufficiency of the projects in the Hillsborough BMAP to address the probable sources identified in this project
- Use decision tool to focus additional field reconnaissance and sampling to further target the identification of sources
  - Targeted sampling using current and new MST assays
  - More targeted field reconnaissance in areas of interest
  - Wildlife surveys in areas of interest
- Continue to provide support for local site investigations to identify violations
  - Fountain Bridge Apartment Complex in the Lower Hillsborough River

Sub-Basin	WBID	Monitoring Location	Classification Matrix Outcome	Human Fecal MST Markers		Ruminant Fecal MST Marker	
				No. Sampling Dates	% Dates Marker(s) Detected	No. Sampling Dates	% Dates Marker Detected
Lower Hillsborough River	1443E	DHR4A	D5	5	100 %	5	20 %
		HR3	D4	5	80 %	5	20 %
		DHR5	D4	2	50 %	2	0 %
		DHR8	D3	4	50 %	4	0 %
		HR2	C4	3	67 %	3	0 %
		HR4	C4	3	67 %	3	0 %
		DHR7	C3	5	60 %	5	60 %
		DHR6	C3	3	33 %	3	0 %
		HR1 REF	B3	3	100 %	8	0 %
		HR5	B3	5	60 %	5	0 %
Blackwater Creek	1482		A4	1	100 %	1	0 %
		BW3	D4	4	75 %	1	0 %
		BW2	D4	3	67 %	3	100 %
		DBW4	A3	1	100 %	1	0 %
		BW5A	A3	2	50 %	2	0 %
Baker Creek	1552C	BW1 REF	A2	7	43 %	6	17 %
		BK2	D3	6	67 %	6	67 %
		BK1	C3	7	57 %	7	29 %
		BK3	C2	2	50 %	2	0 %
Flint Creek	1552A	BK4	A4	1	100 %	1	0 %
		BK5	A3	2	100 %	2	0 %
		FL2	C2	6	50 %	6	0 %
		FL1	C2	5	20 %	5	0 %
Spartman Branch	1561	FL3	A3	6	33 %	6	0 %
		SB1	D3	6	17 %	6	0 %
		SB2	D2	5	20 %	6	0 %
New River	1442	DSB3	C2	2	0 %	2	50 %
		SB3	B2	4	25 %	4	0 %
		NR2	C3	7	29 %	7	29 %
		NR1	C2	2	0 %	2	50 %
		NR3	C2	2	0 %	2	50 %



# Statewide

- Apply decision-support tool statewide to identify priority watersheds and stations for additional sampling and remediation
- Continue to validate methods and provide leadership to local governments to identify and remove sources
  - Provide cost-share funding to assist local governments
  - Track progress using decision-support tool through use of annual reports
- Conduct workshops throughout the state, and especially in priority communities, to encourage the identification of sources and the remediation of water quality
- Collaborate with FDACS to identify sources in priority watersheds dominated by agricultural uses







# Strategic Approach and Case Studies

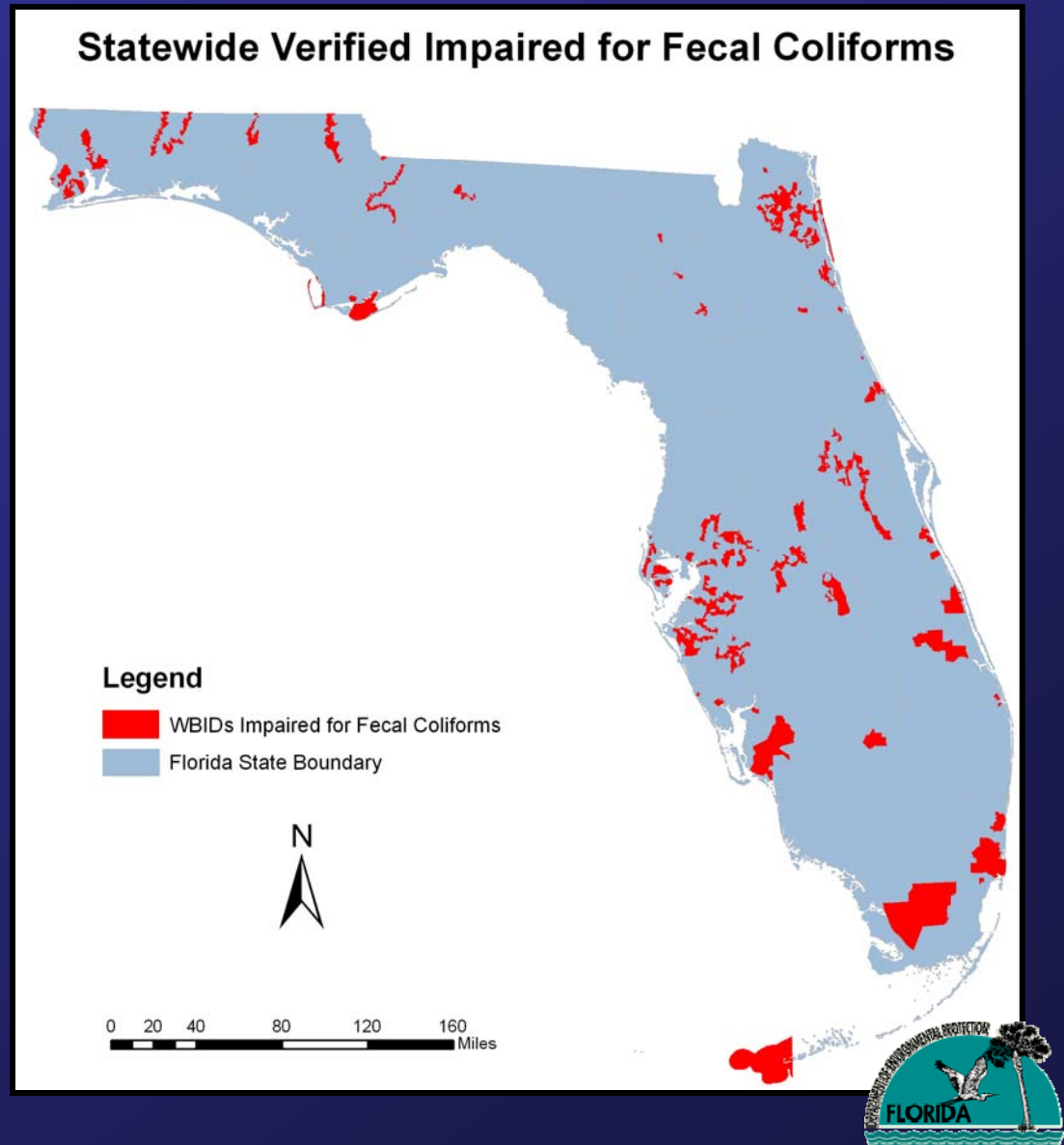


# Overview

- Issue in Florida
- Project Areas
- Development of Two Approaches / Tools
- Decision-Support Tool
- Identification of Sources using Weight-of-Evidence Approach
- Case Studies

# Issue in Florida

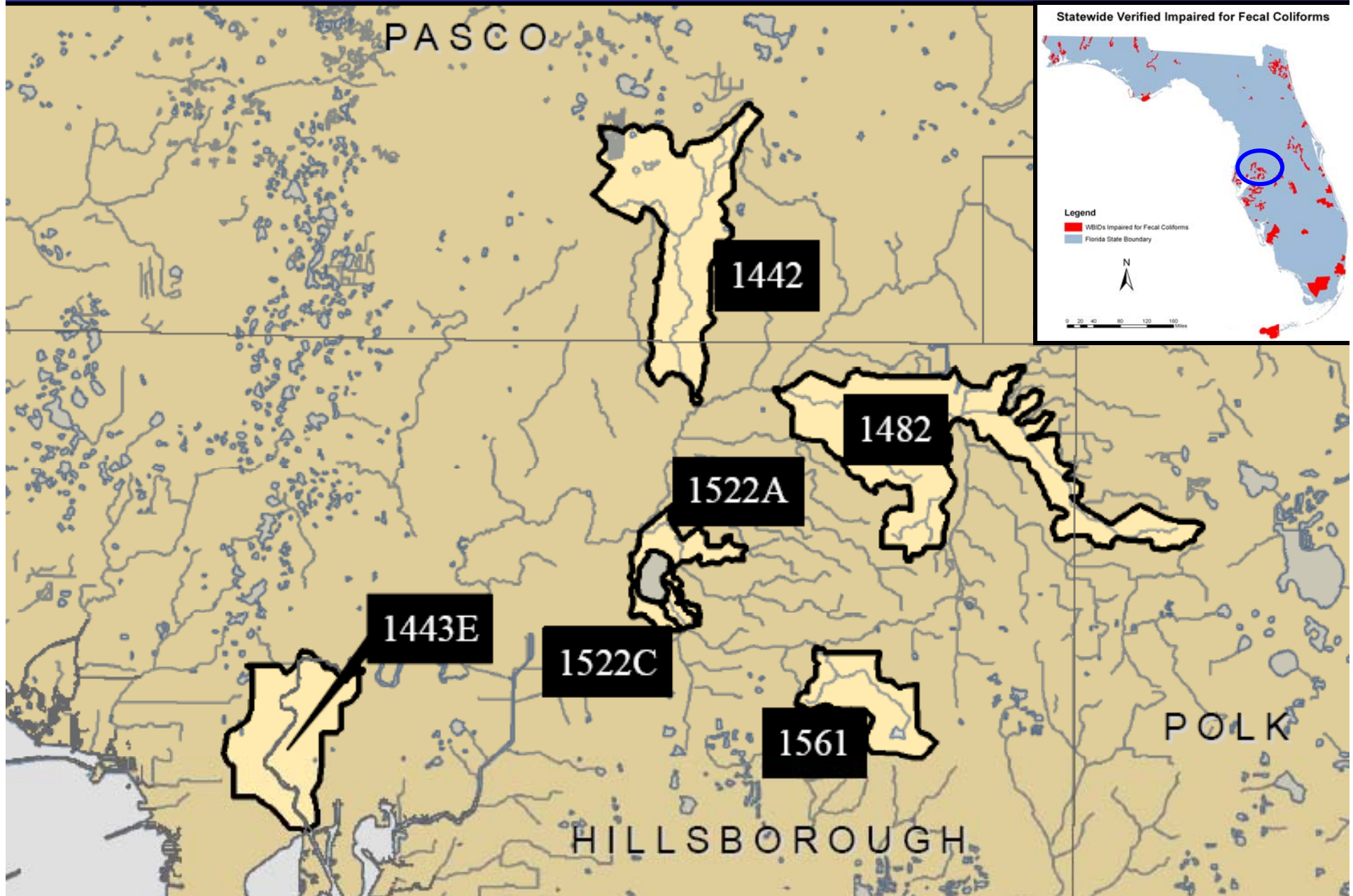
**226 WBIDs  
verified as  
impaired for  
fecal coliform  
contamination  
in Florida**



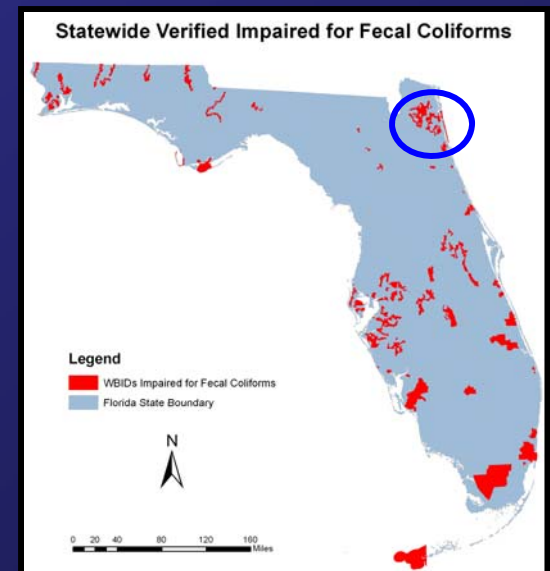
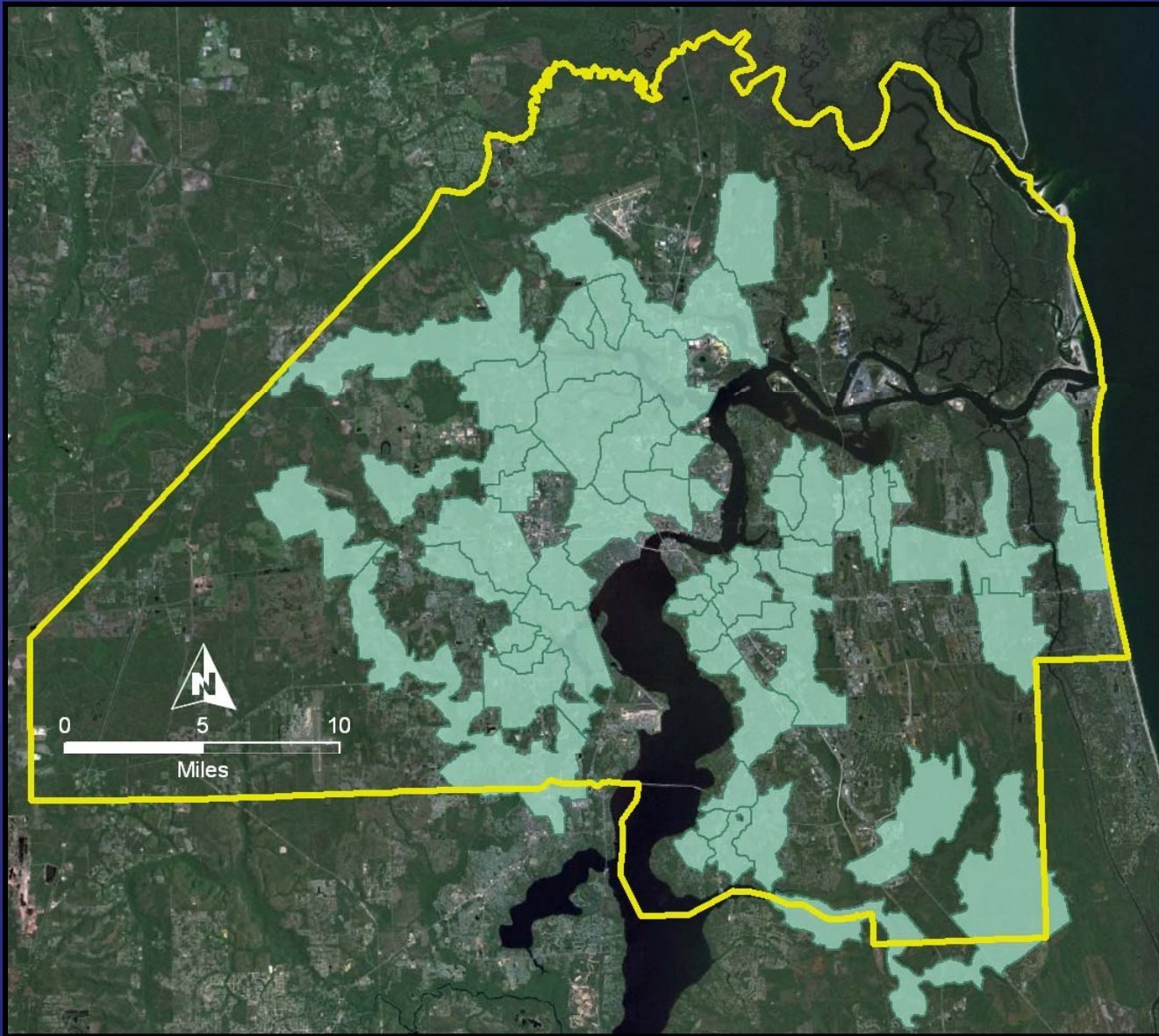


# Project Areas

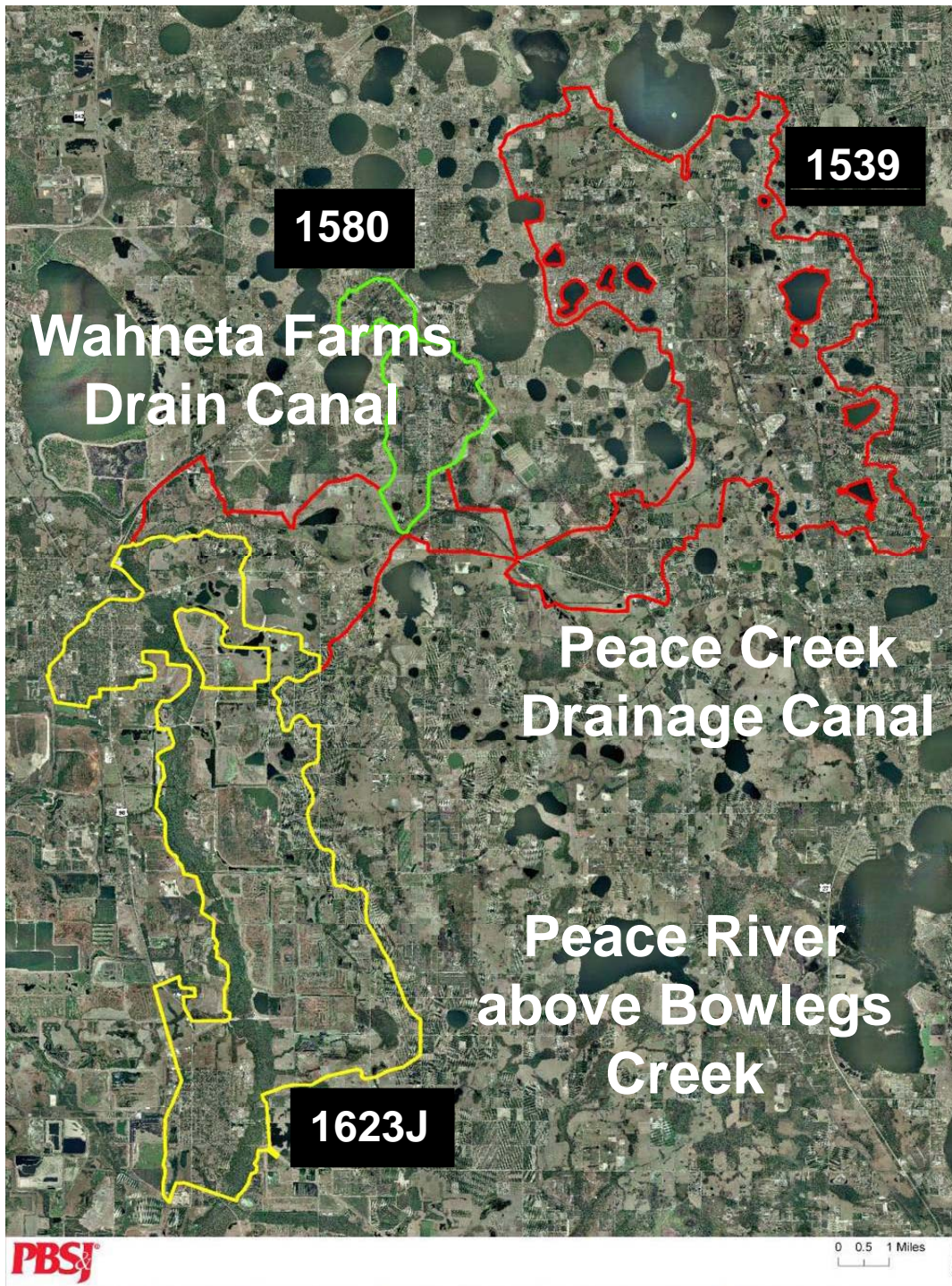
# Hillsborough River Watershed (6 WBIDs)



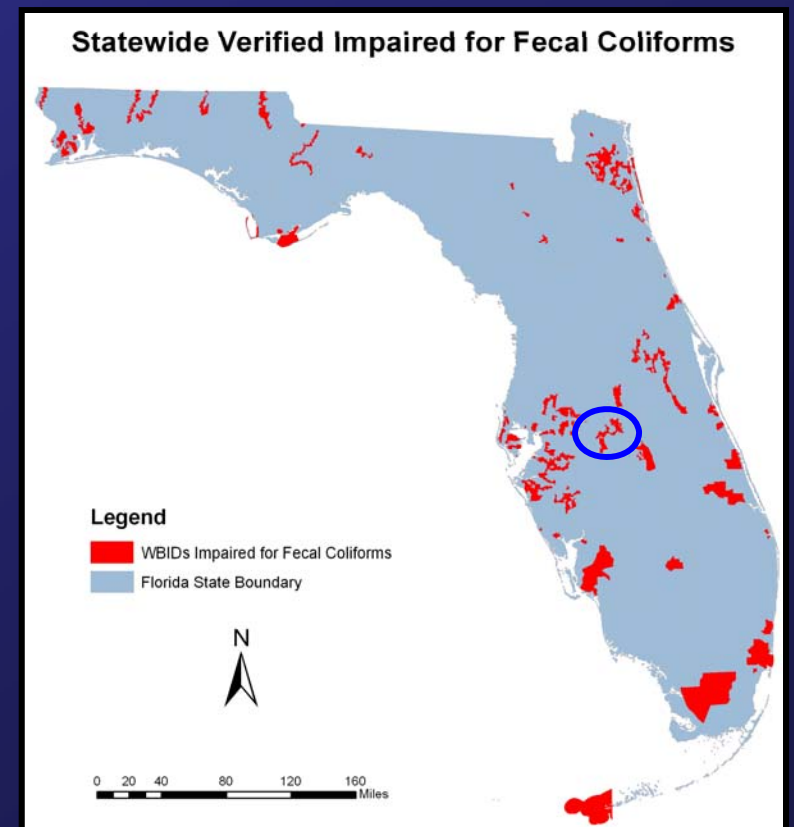
# Lower St. Johns River (55 WBIDs)







# Upper Peace Creek (3 WBIDs)





**Legend**

- WBIFCs Impaired for Fecal Coliforms
- Florida State Boundary

0 20 40 80 120 160 Miles

**Where is the pollution  
coming from?**

**What can we do with the  
information we have  
today?**



# **Developed Two Approaches / Tools**

**Identification of  
sources of fecal  
coliform  
contamination  
using weight-of-  
evidence**

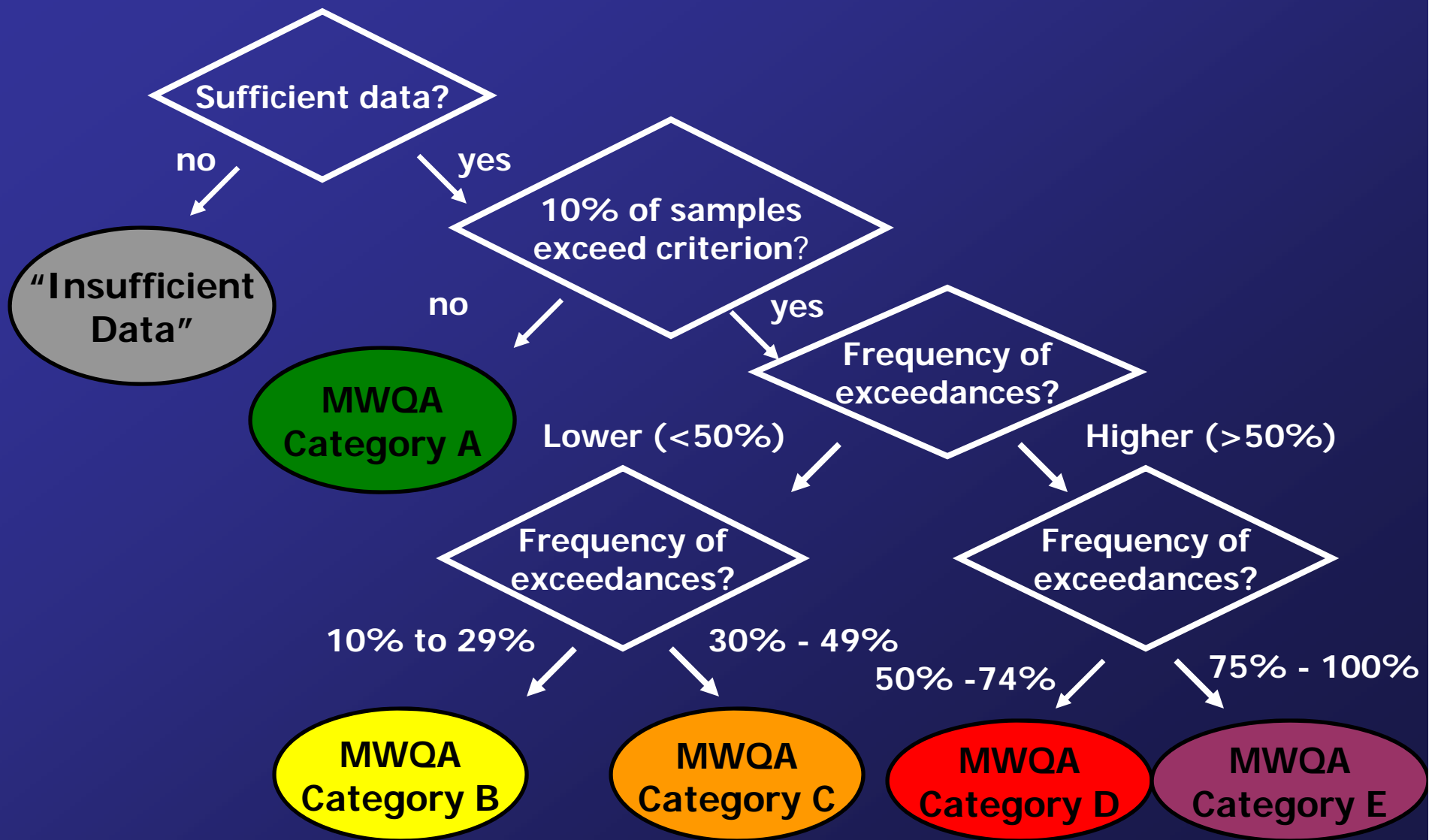
**Decision Support  
Tool developed to  
guide potential  
additional sampling  
and help prioritize  
basins for  
management action**

**Designed for state-wide application**

**Tested in the Hillsborough River watershed**

# **Decision-Support Tool for Guiding Additional Sampling and Prioritizing Management Actions**

# Decision Support Tool



# Prioritization of WBIDs (and sites within WBIDs) for management action

Sub-Basin	WBID	Monitoring Location	Classification Matrix Outcome	Human Fecal MST Markers		Ruminant Fecal MST Marker	
				No. Sampling Dates	% Dates Marker(s) Detected	No. Sampling Dates	% Dates Marker Detected
Lower Hillsborough River	1443E	DHR4A	D5	5	100 %	5	20 %
		HR3	D4	5	80 %	5	20 %
		DIIR5	D4	2	50 %	2	0 %
		DHR8	D3	4	50 %	4	0 %
		HR2	C4	3	67 %	3	0 %
		HR4	C4	3	67 %	3	0 %
		DHR7	C3	5	60 %	5	60 %
		DHR6	C3	3	33 %	3	0 %
		HR1 REF	B3	3	100 %	8	0 %
		HR5	B3	5	60 %	5	0 %
		DHR10	A4	1	100 %	1	0 %
Blackwater Creek	1482	BW3	D4	4	75 %	1	0 %
		BW2	D4	3	67 %	3	100 %
		DBW4	A3	1	100 %	1	0 %
		BW5A	A3	2	50 %	2	0 %
		BW1 REF	A2	7	43 %	6	17 %
Baker Creek	1552C	BK2	D3	6	67 %	6	67 %
		BK1	C3	7	57 %	7	29 %
		BK3	C2	2	50 %	2	0 %
		BK4	A4	1	100 %	1	0 %
		BK5	A3	2	100 %	2	0 %
Flint Creek	1552A	FL2	C2	6	50 %	6	0 %
		FL1	C2	5	20 %	5	0 %
		FL3	A3	6	33 %	6	0 %
Spartman Branch	1561	SB1	D3	6	17 %	6	0 %
		SB2	D2	5	20 %	6	0 %
		DSB3	C2	2	0 %	2	50 %
		SB3	B2	4	25 %	4	0 %
New River	1442	NR2	C3	7	29 %	7	29 %
		NR1	C2	2	0 %	2	50 %
		NR3	C2	2	0 %	2	50 %

# Identification of Sources using Weight- of-Evidence Approach



# Weight-of-Evidence Approach

- **Phase I**

- Target efforts
- Discriminate potential sources
- Effectively select the “right” tests
- Limit costs associated with sampling

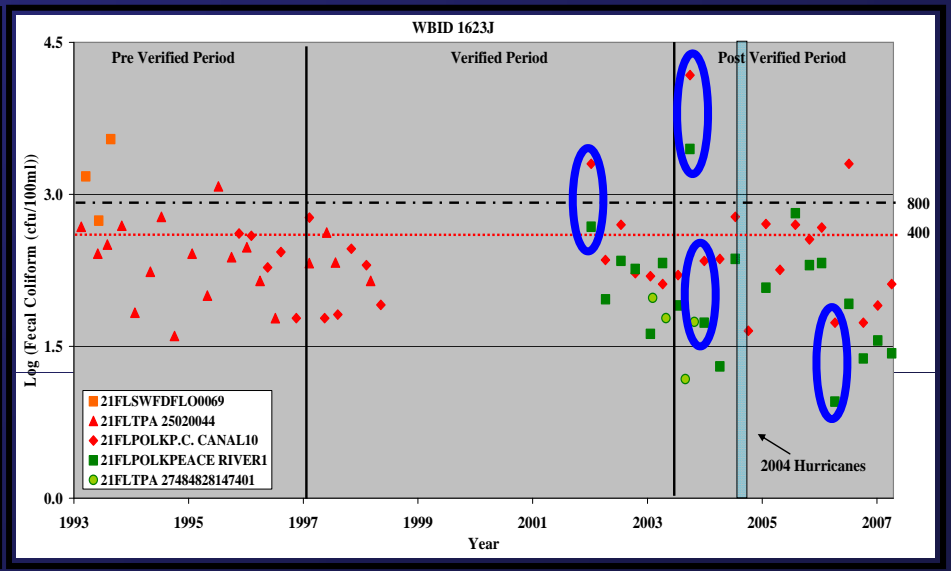
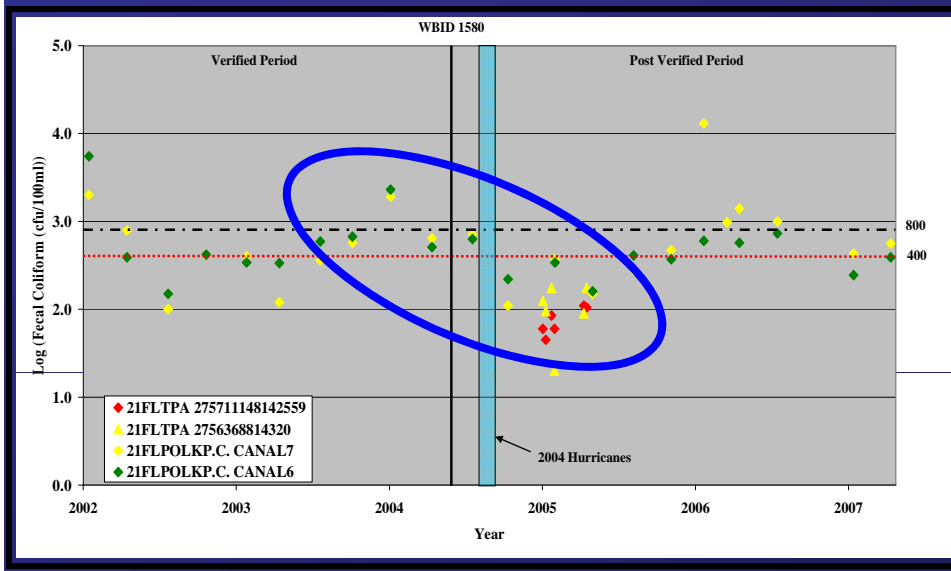
- **Phase II**

- First used in six LSJR tributaries
- Modified for Hillsborough River
- Further modified for additional LSJR basins

# Three Components of the Initial Screening Process

# Initial Screening

- Detailed review of long-term datasets
  - Chronic or episodic exceedances
  - Correlation with other physical parameters (e.g., turbidity)
  - Correlation with rainfall
  - Potential for transport from upstream stations/other tributaries



# Initial Screening

## 2. Compilation and evaluation of GIS data

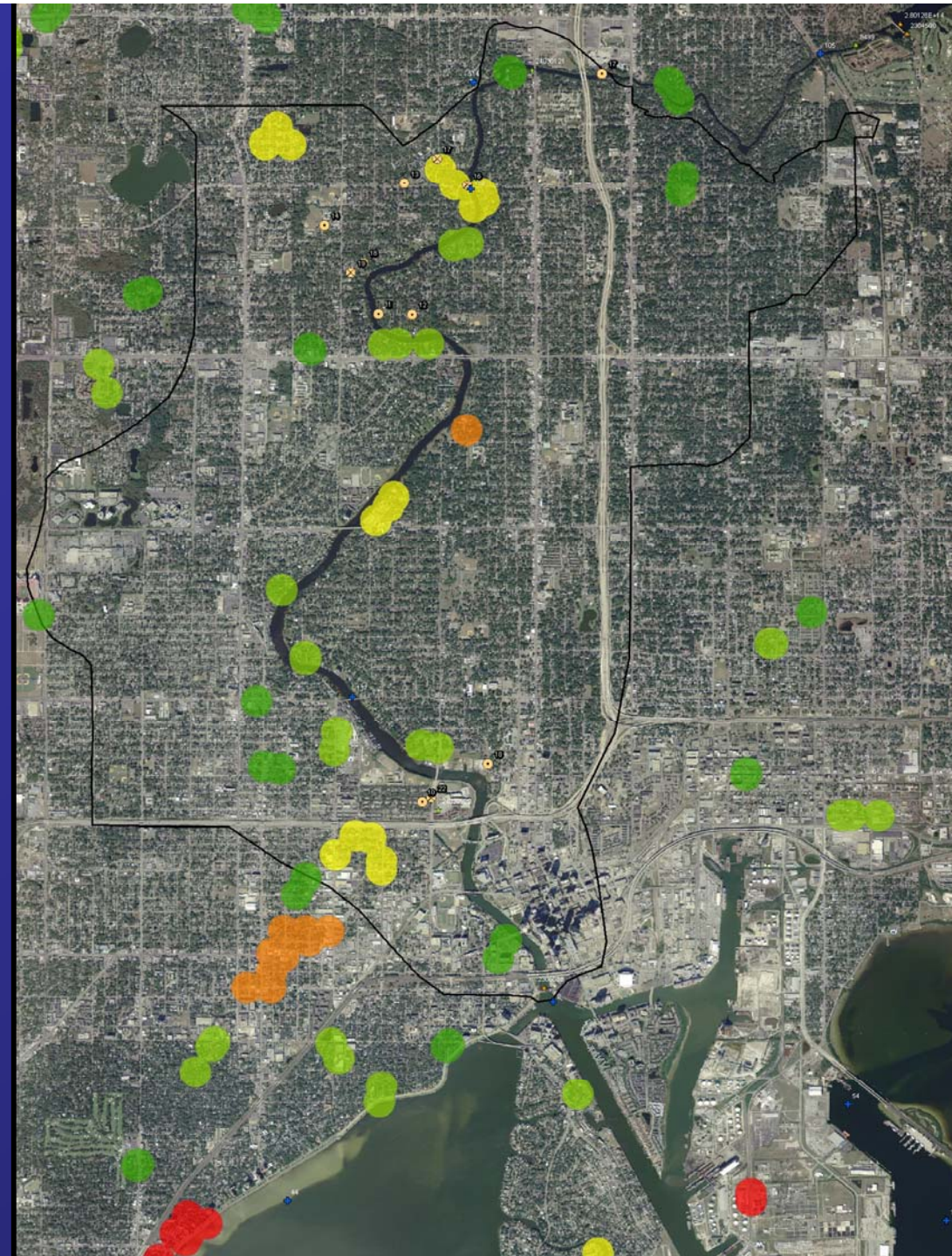
- Land use
- Soil type
- Long-term monitoring stations
- Red line properties (prone to flooding)
- Landfills
- Infrastructure
  - Stormwater
  - Septic Tanks
  - Sanitary Sewer
- Potential illicit discharges
- Septic tank repair permits
- Sanitary Sewer Overflows
- Permitted discharges





# City of Tampa

## Areas of Repetitive Reported SSOs 2001-2007



### Number of Reports

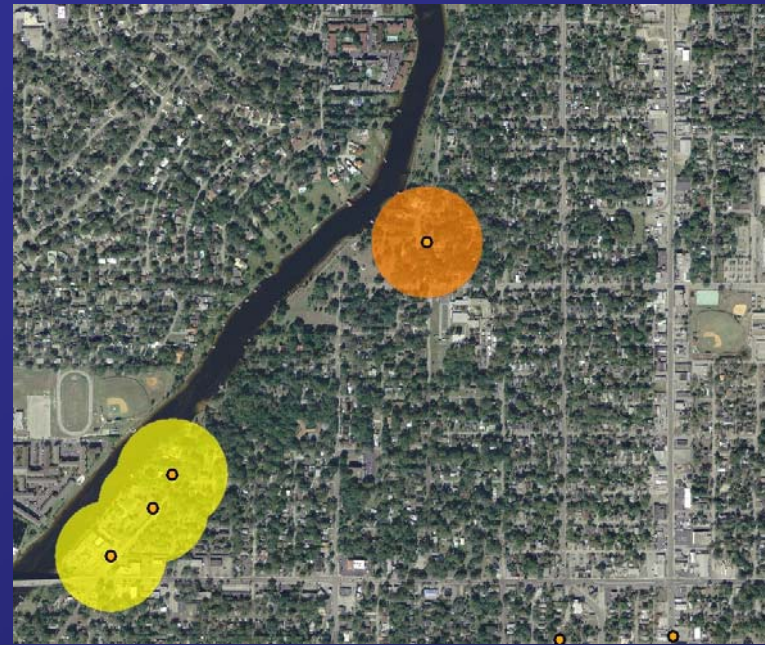
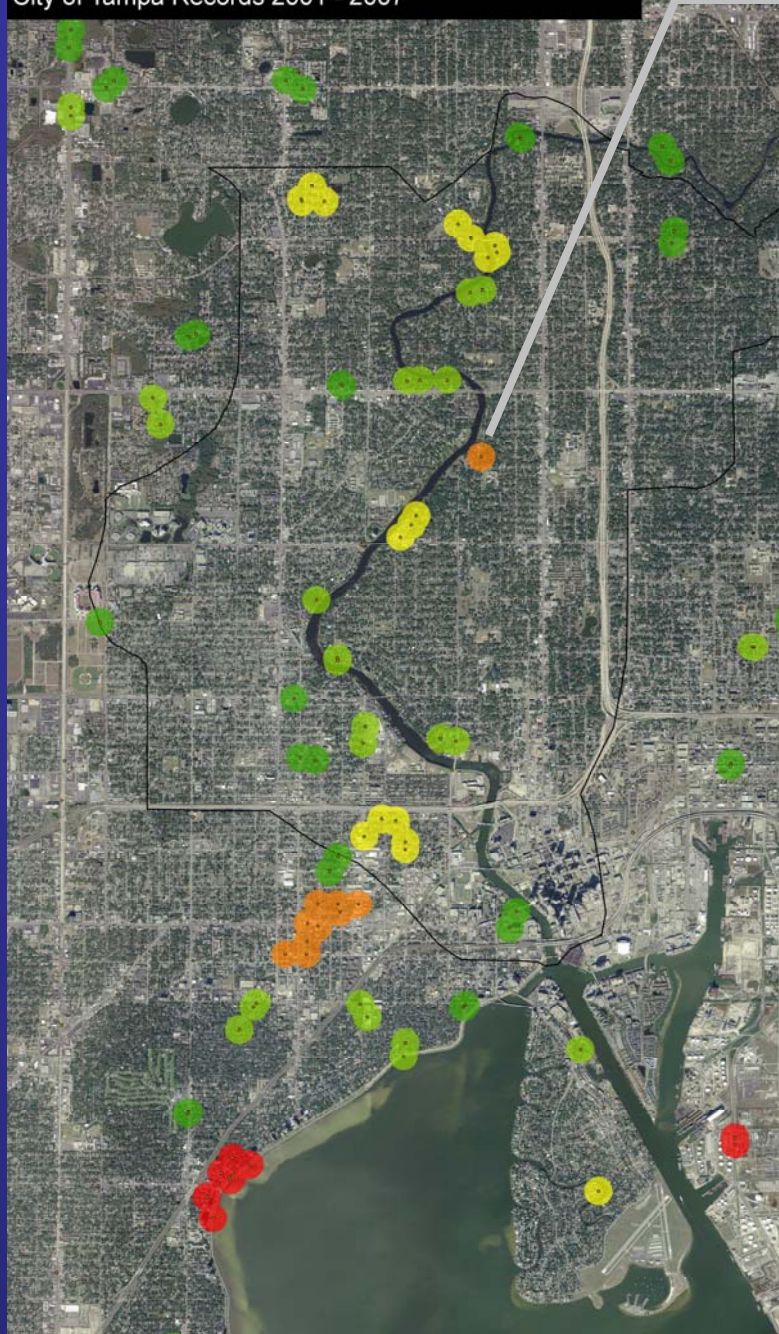


2 3-4 5-7 8-11 12-18

*Shaded regions represent multiple and/or repetitive overflow reports. A 500-ft buffer defines adjacency.*



## Areas of Repetitive SSO Reporting City of Tampa Records 2001 - 2007



- 10 Reported events (2001-2006) at same location
- Pump station and associated gravity lines
- Mechanical failure, infiltration/inflow due to extreme rainfall
- 200 – 94,800 gallons
- Most drained directly into river

(COT Data 2001 – 2007)

# Initial Screening

## 3. Collaboration with local stakeholders

- One-on-one interviews
- Project Workshop
  - Presentation of results to date
  - Maps-on-the Table exercise
  - Organize field effort
    - Brainstorm and document additional potential sources
    - Build trust and consensus among stakeholders
    - Increase accountability of stakeholders

# **Three Components of the Field Implementation Process**



# Field Implementation

## 1. Field Reconnaissance

- Internal field review
- Walk the WBIDs exercise
  - Confirmation of potential sources
  - “Boots on the ground” knowledge
  - Education and accountability of stakeholders
  - Selection and verification of sampling locations
- Build consensus





# Field Implementation

2. Use of decision tree based on “toolbox” of MST methods and weight-of-evidence
  - Improvement of existing methods
    - Indicator organisms in sediments
    - Identification of old vs. new sources
  - New available methods
    - Improved human markers (HPyV)
    - Addition of horse-specific *Bacteroides*
    - Addition of qPCR methods
    - Infrared Thermal Imagery
  - Methods that still need improvement
    - Fluorometry

# Field Implementation

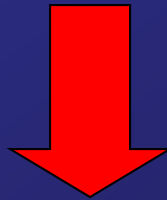
## 3. Collaboration with local stakeholders

- Bi-monthly meetings with BMAP Steering Committee
  - Dissemination of results
  - Ideas for upcoming sampling events
  - Build consensus and accountability
- Continual communication and field visits
  - Updates on local water quality issues
  - Field and infrastructure reconnaissance requests



# Analysis of Results

**Methods and regulatory aspects of MST are still being developed**



**Weight-of-Evidence and flexibility in the approach are requirements for this type of investigation**

# **Application of the Weight-of-Evidence Approach in the Hillsborough River Basin**



# Toolbox of MST Methods Used in Hillsborough River Watershed

## PCR Tests

*Bacteroides* – an anaerobic fecal bacterium  
(separate tests for **human**, **ruminant**, **horse**)

*Enterococcus faecium esp* gene

- Gene for virulence factor of bacterium found in **humans**

Human polyomavirus

- Nonpathogenic (harmless) virus shed in urine; high carrier rate in **human** population & ubiquitous in sewage

# Application of Methods

- **Month 1**
  - Use of indicator suite and advanced MST techniques simultaneously at all stations
- **Months 2-8**
  - ~50% of samples were re-tested using advanced MST techniques each month
  - Increased testing in some WBIDs and decreased testing in others
  - Sediments tested at 6-9 stations each month

# Basic Factors for Consideration

## 1. When to Sample

- Wet and dry seasons
- Monthly
- Within 3-7 days of significant rainfall events (when possible)
- During out-going tide (Hillsborough River)

## 2. Where to Sample

- Long-term monitoring stations
- Bracket potential contamination sources
- Land-use classification
- Tributary organization
- Safety and Accessibility



# Basic Factors for Consideration

## 3. What and How to Sample

- **Water column by grab sample method**  
(Standard Methods for the Examination of Water and Wastewater [9060A])
- **Sediment samples using Ponar®, if necessary**
- **Additional field data**
  - **Temperature, conductivity, DO, pH, turbidity**
- **Transport on ice to qualified laboratory for analysis**





# MST Sampling in the Hillsborough River Basin

- USF

- 20 “Fixed” Stations

- Sample using 2 Teams (USF / PBS&J)
    - Entire indicator suite (3 samples)
    - 6-9 sediment samples / month
    - Advanced MST analysis

USF ~ 68  
samples/month

- Additional Lab

- 17-28 “Flexible” Stations

- Sample using FDEP personnel
    - Fecal coliforms only
    - No sediments or advanced MST analysis

FDEP/Lab = 20  
samples/month

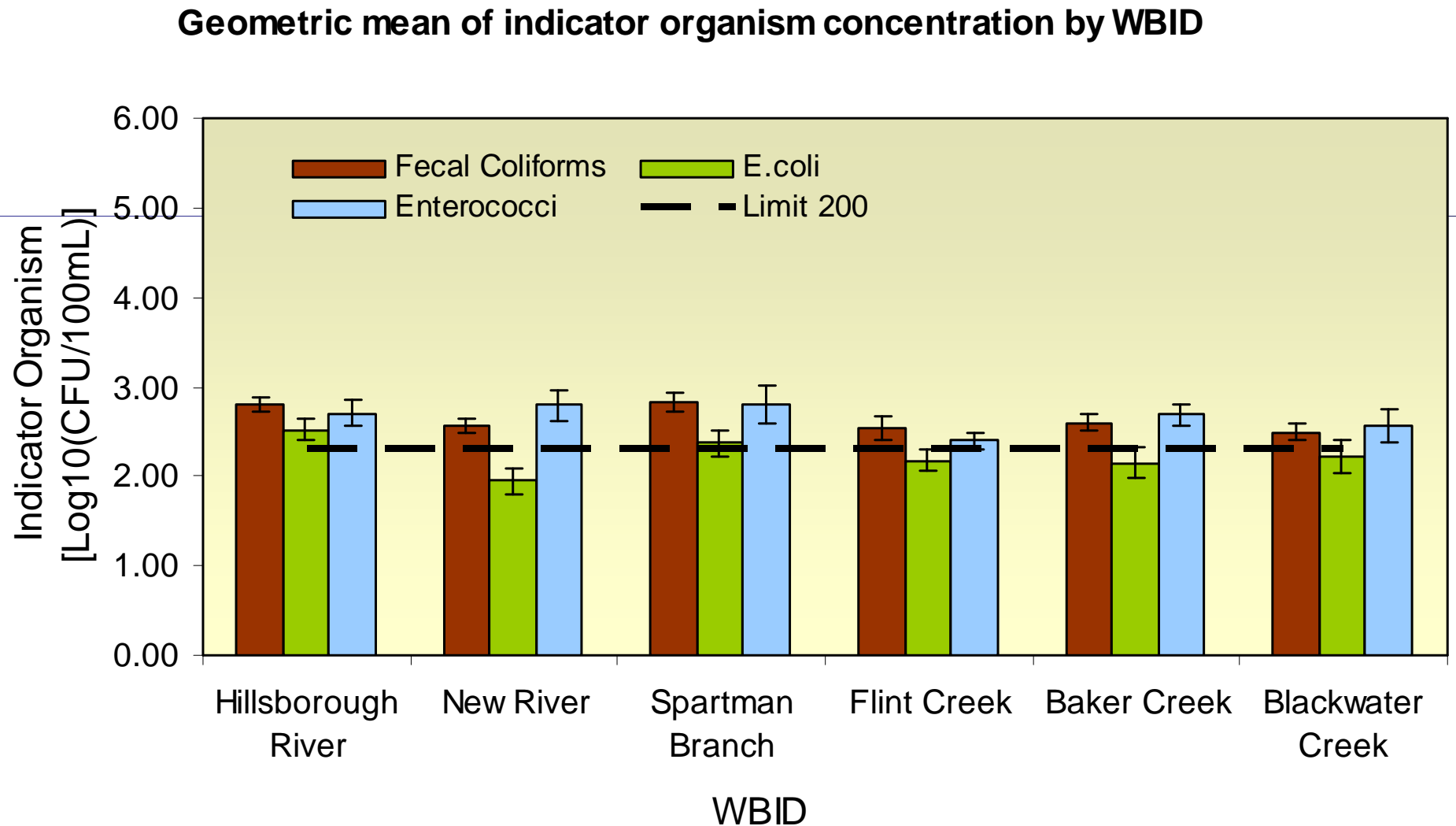
# **Case Studies**

**Impairment at Station Level**

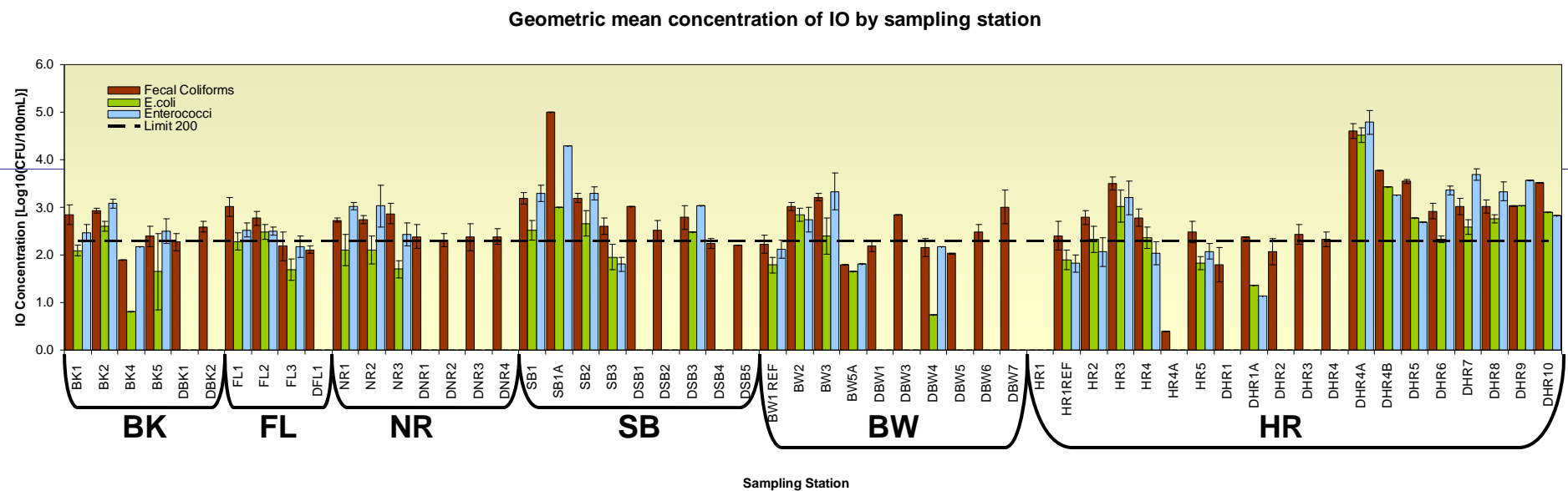
**Collaboration with Local  
Stakeholders**

**Hillsborough River Watershed**

# Geometric Mean Concentration of Indicator Organism by WBID



# Geometric Mean Concentration of Indicator Organism by Sampling Station





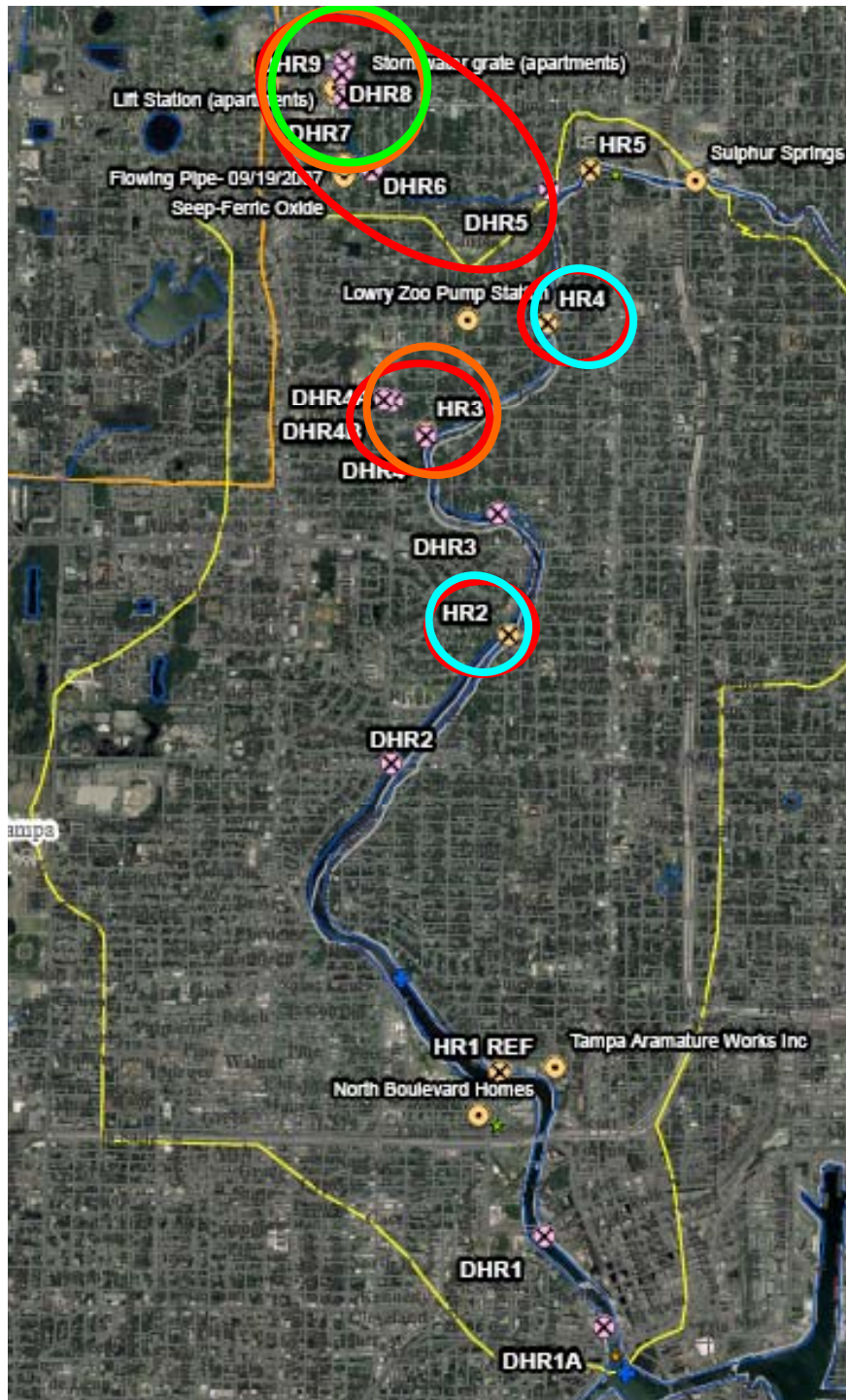


# Classification Matrix Outcome

## Lower Hillsborough River

				Human Fecal MST Markers		Ruminant Fecal MST Marker	
Sub-Basin	WBID	Monitoring Location	Classi- fication Matrix Outcome	No. Sampling Dates	% Dates Marker(s) Detected	No. Sampling Dates	% Dates Marker Detected
Lower Hillsborough River	1443E	<b>DHR4A</b>	D5	5	100 %	5	20 %
		<b>HR3</b>	D4	5	80 %	5	20 %
		<b>DHR5</b>	D4	2	50 %	2	0 %
		<b>DHR8</b>	D3	4	50 %	4	0 %
		<b>HR2</b>	C4	3	67 %	3	0 %
		<b>HR4</b>	C4	3	67 %	3	0 %
		<b>DHR7</b>	C3	5	60 %	5	60 %
		<b>DHR6</b>	C3	3	33 %	3	0 %
		<b>HR1 REF</b>	B3	3	100 %	8	0 %
		<b>HR5</b>	B3	5	60 %	5	0 %
		<b>DHR10</b>	A4	1	100 %	1	0 %

Station	H-Bac	esp	HPyV	R-Bac	Hr-Bac
HR1	2/3	1/3	0/3	0/6	0/6
HR2	1/3	2/3	0/3	0/3	0/3
HR3	2/5	2/5	0/5	1/5	0/5
DHR4A	4/5	0/5	1/5	1/5	1/5
HR4	1/3	2/3	0/3	0/3	0/3
HR5	1/5	2/5	0/3	0/3	0/3
DHR5	0/2	1/2	0/2	0/2	0/2
DHR6	0/3	0/3	1/3	0/3	1/3
DHR7	3/5	0/5	0/3	3/5	0/3
DHR8	1/4	0/4	1/4	0/4	0/4
DHR9	0/2	0/2	0/2	0/1	0/1
DHR10	1/1	0/1	0/1	0/1	0/1

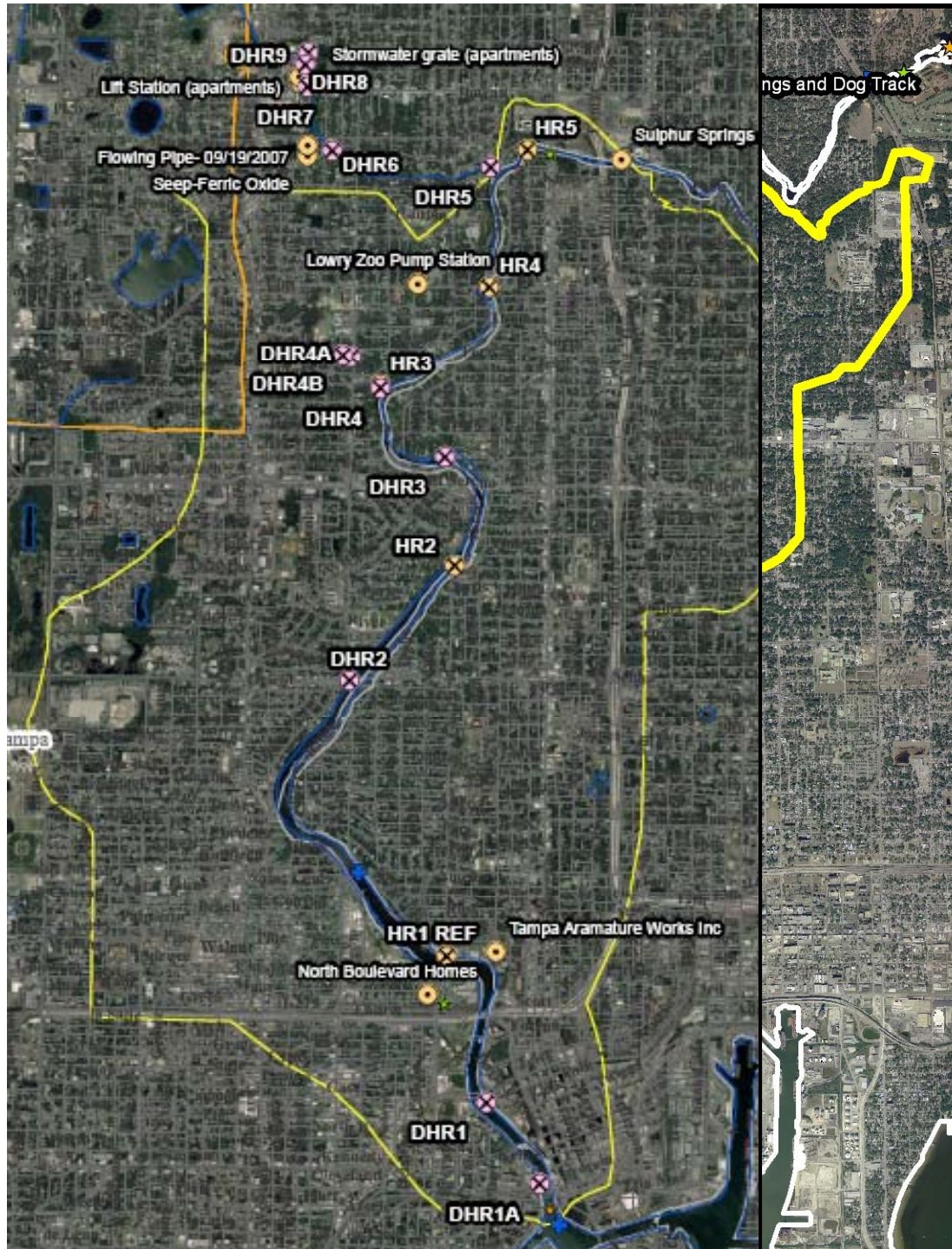


- Human-specific markers at every site tested (except DHR9)
  - Often coincided with IO exceedances
- Extremely elevated IO concentrations in sediments at several stations
- Ruminant sources apparent but relatively insignificant

### Most Likely Sources

1. Sanitary Sewer and SSOs
2. OSTDS
3. Illicit Discharges
4. Stormwater (as a conveyance system)





# Lower Hillsborough River WBID 1443E

North-to-South  
flow

One main channel

Potential human  
impacts likely  
throughout WBID

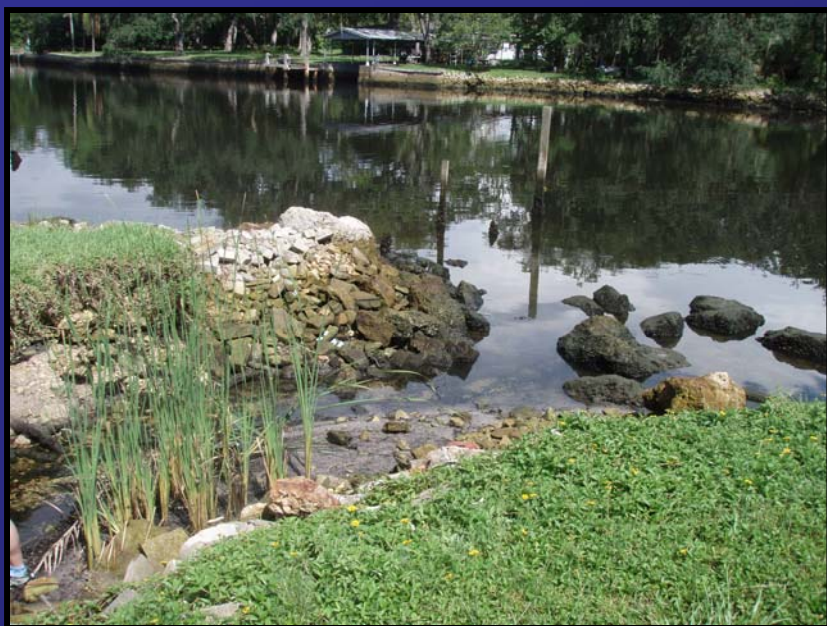


# Lower Hillsborough River

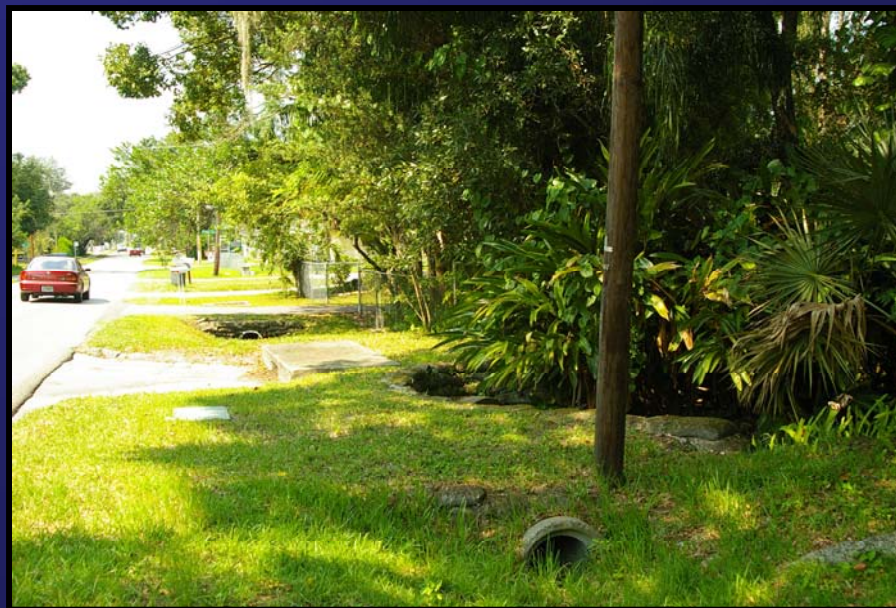
HR1 Ref



HR2



HR3



DHR4a





HR4



HR5





**DHR6**

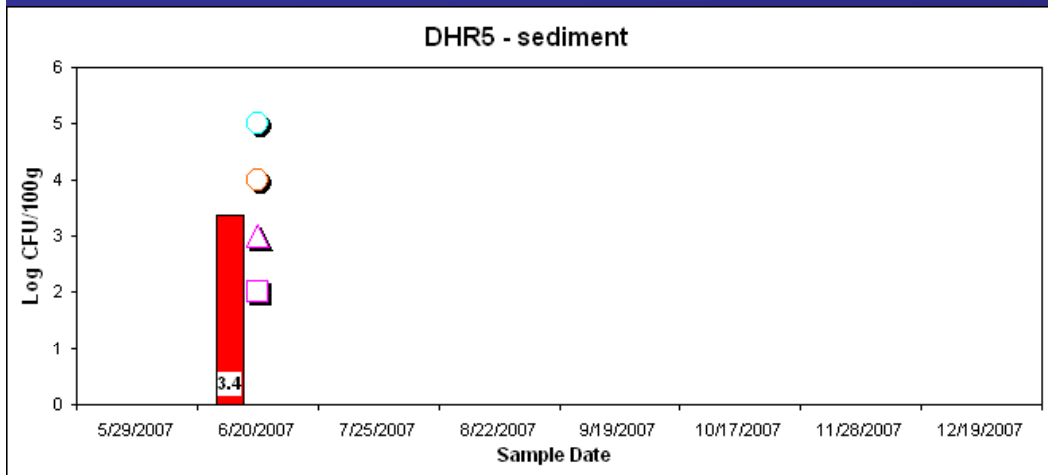
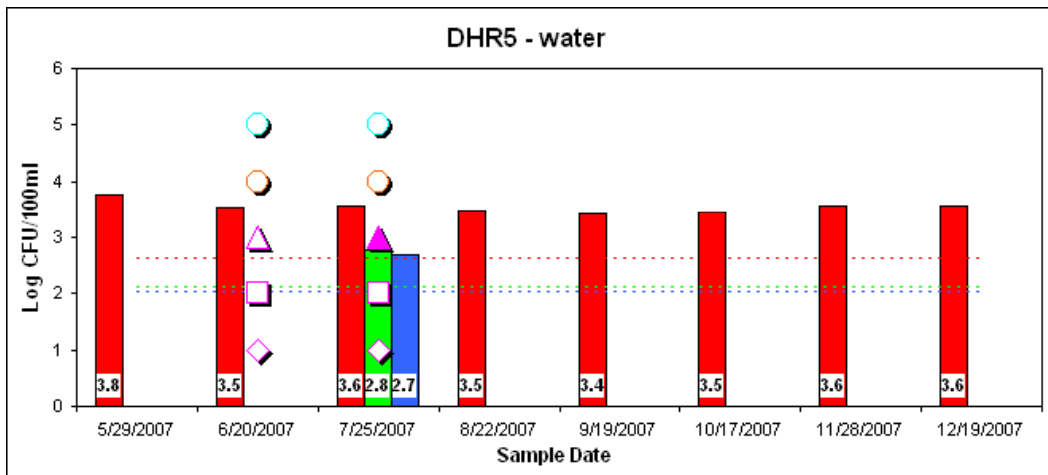


**DHR7**



**DHR8**

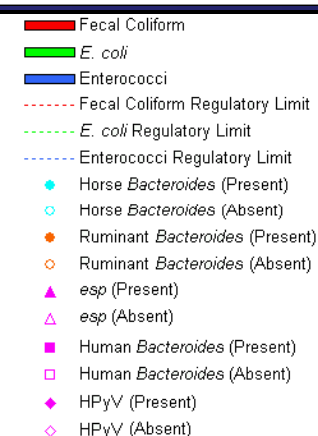
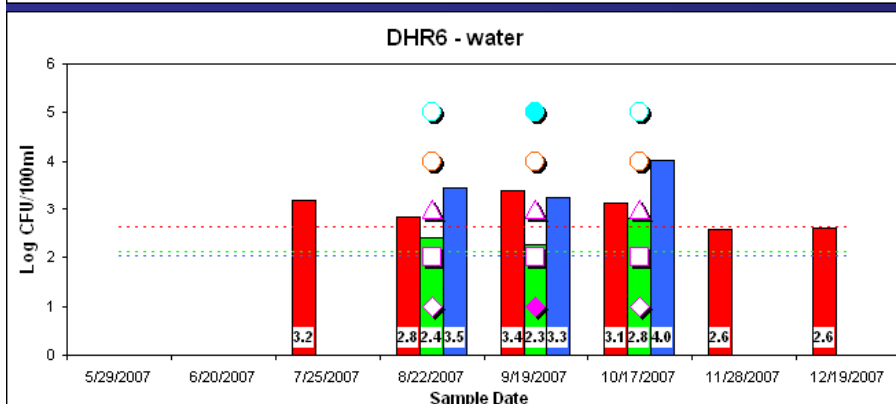
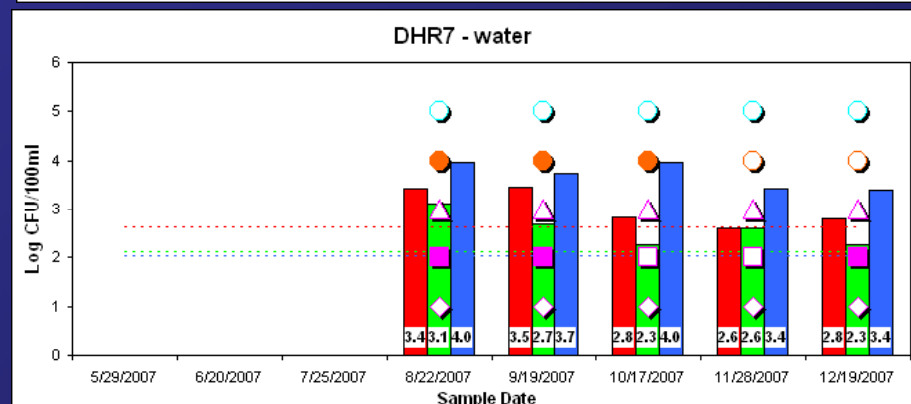
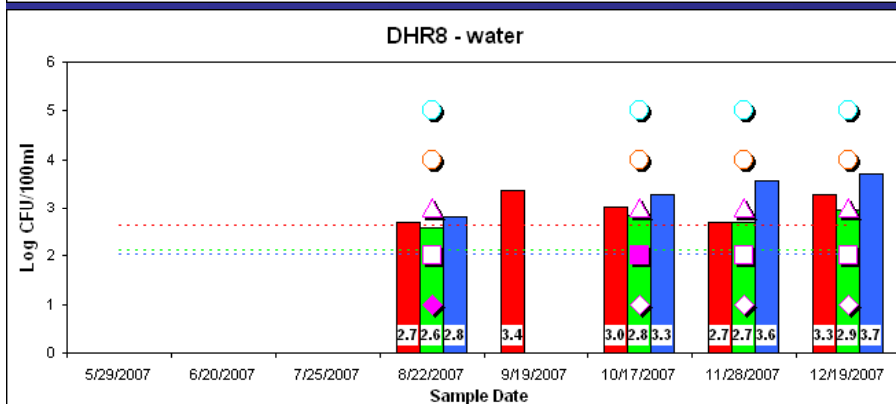
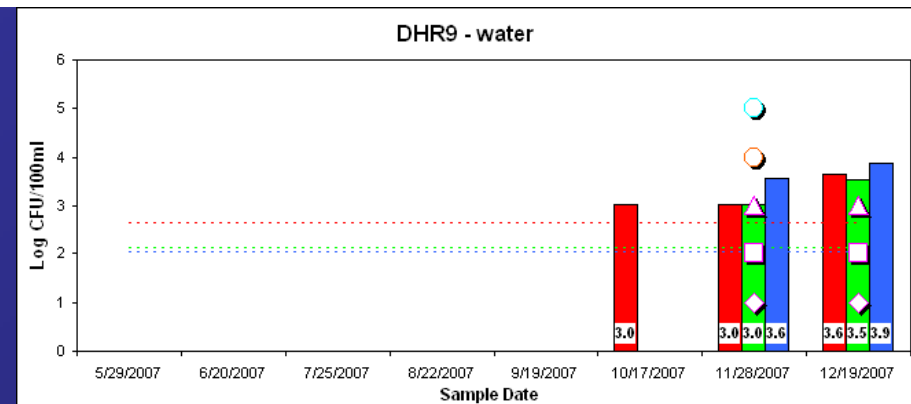
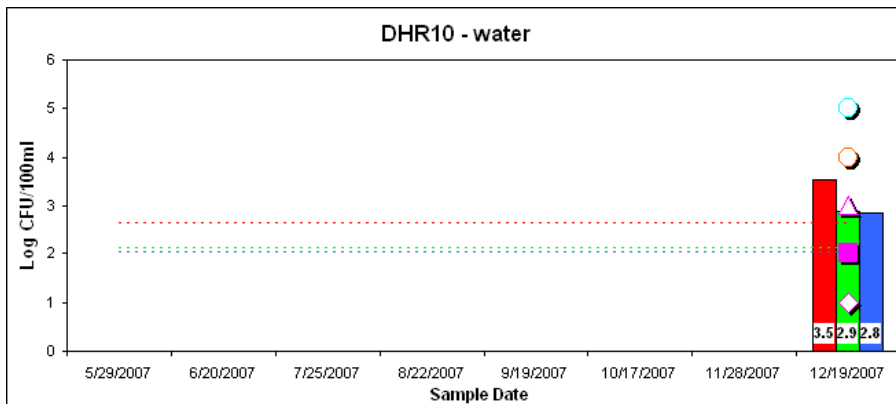




# Kirby Creek at Hillsborough River



- Fecal Coliform
- *E. coli*
- Enterococci
- Fecal Coliform Regulatory Limit
- *E. coli* Regulatory Limit
- Enterococci Regulatory Limit
- Horse *Bacteroides* (Present)
- Horse *Bacteroides* (Absent)
- Ruminant *Bacteroides* (Present)
- Ruminant *Bacteroides* (Absent)
- ▲ *esp* (Present)
- △ *esp* (Absent)
- Human *Bacteroides* (Present)
- Human *Bacteroides* (Absent)
- ◆ HPyV (Present)
- ◇ HPyV (Absent)



**Mixture of MST markers but land use information suggests human sources throughout Kirby Creek**

# COT Investigation of Upper Kirby Creek

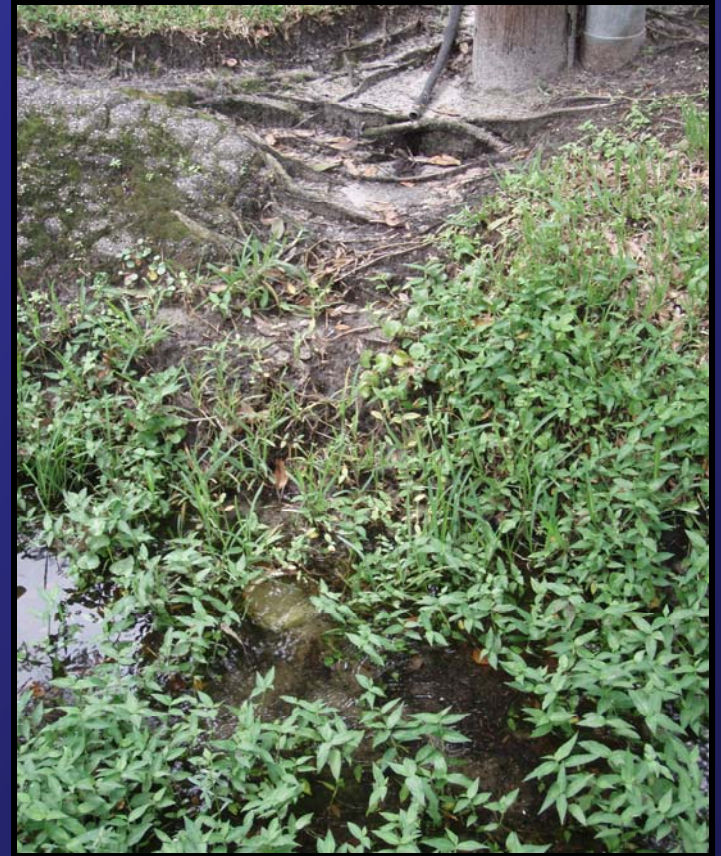
- Drainage system investigated upstream of DHR7 on Feb. 11, 2008
  - Flow in connecting ditch system after ~2 weeks with no appreciable rainfall
  - Stormwater grate (DHR10) full but no evidence of odor or sewage
  - Onsite retention pond appeared normal
- Redline properties identified
- Parcels within COT limits that do not have sewer accounts upstream of DHR8 (outlined in yellow)



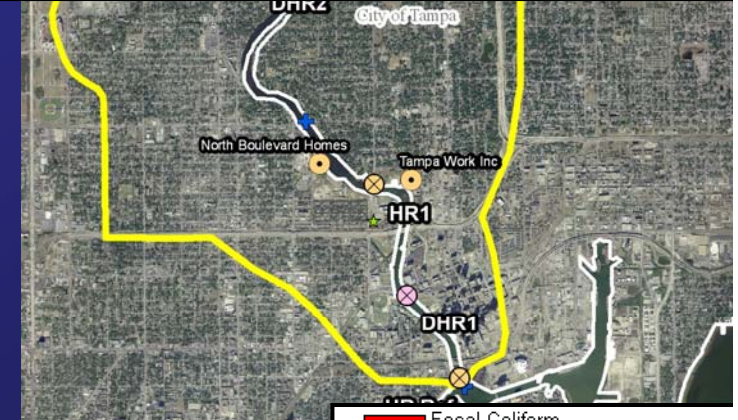
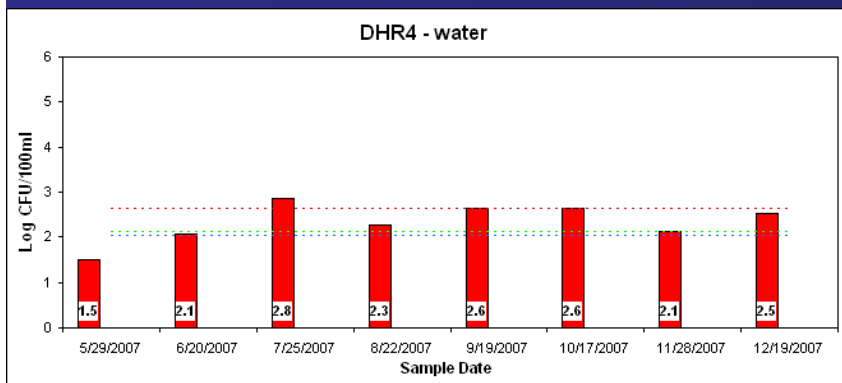
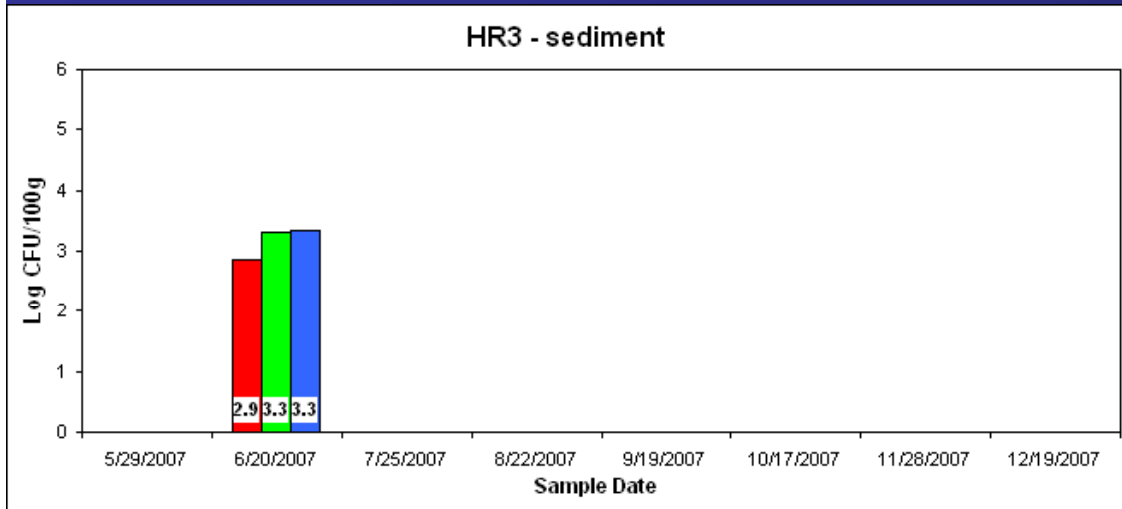
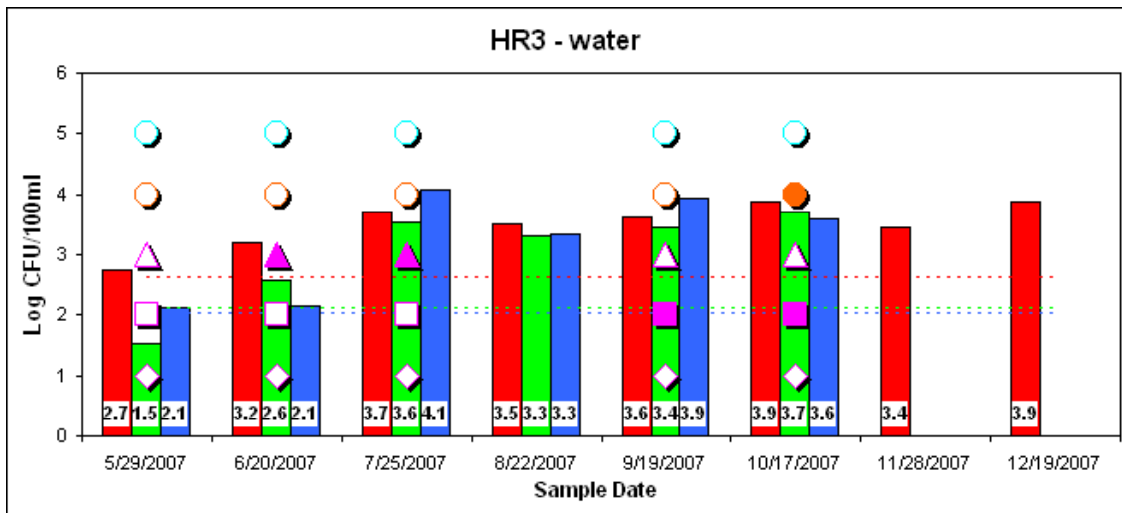


# Additional COT Investigations of Midstream Kirby Creek

- Flowing pipe connected to water softener-letter sent to resident
- Pond overflow pipe identified and left as is
- Groundwater seep identified in ditch
  - Ferric oxide where bottom of ditch interfaces with groundwater

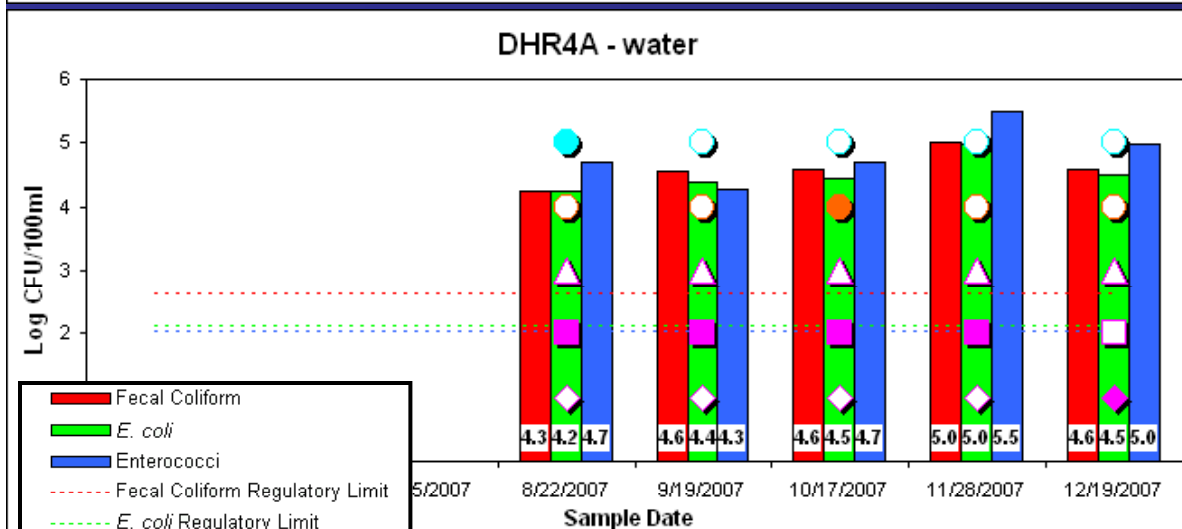
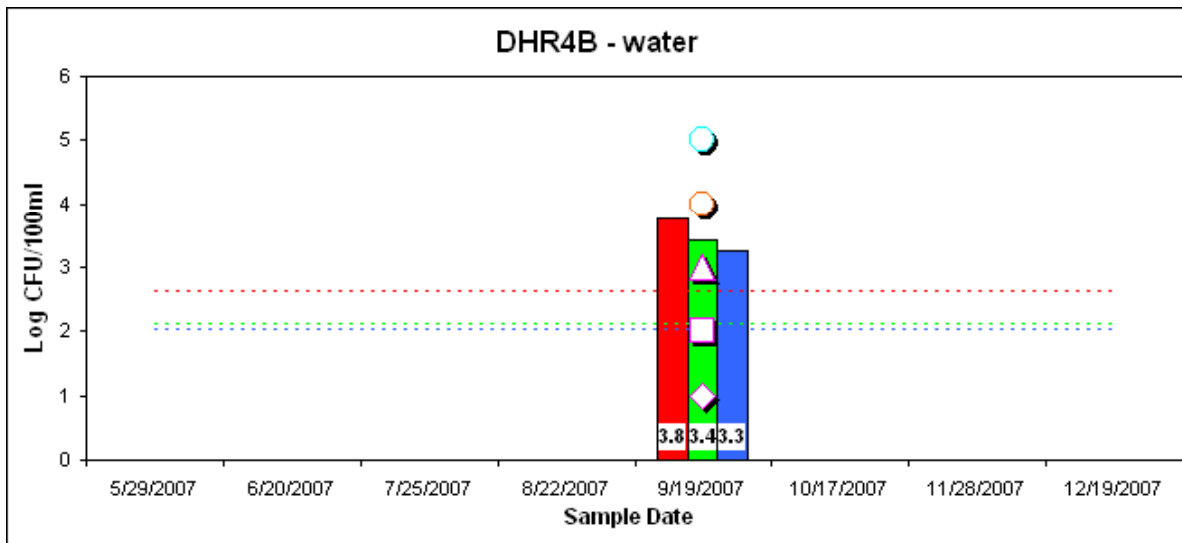






- Fecal Coliform
- *E. coli*
- Enterococci
- Fecal Coliform Regulatory Limit
- *E. coli* Regulatory Limit
- Enterococci Regulatory Limit
- Horse *Bacteroides* (Present)
- Horse *Bacteroides* (Absent)
- Ruminant *Bacteroides* (Present)
- Ruminant *Bacteroides* (Absent)
- ▲ *esp* (Present)
- △ *esp* (Absent)
- Human *Bacteroides* (Present)
- Human *Bacteroides* (Absent)
- ◆ HPyV (Present)
- ◇ HPyV (Absent)

## West Jean Street Tributary at Lower Hillsborough River



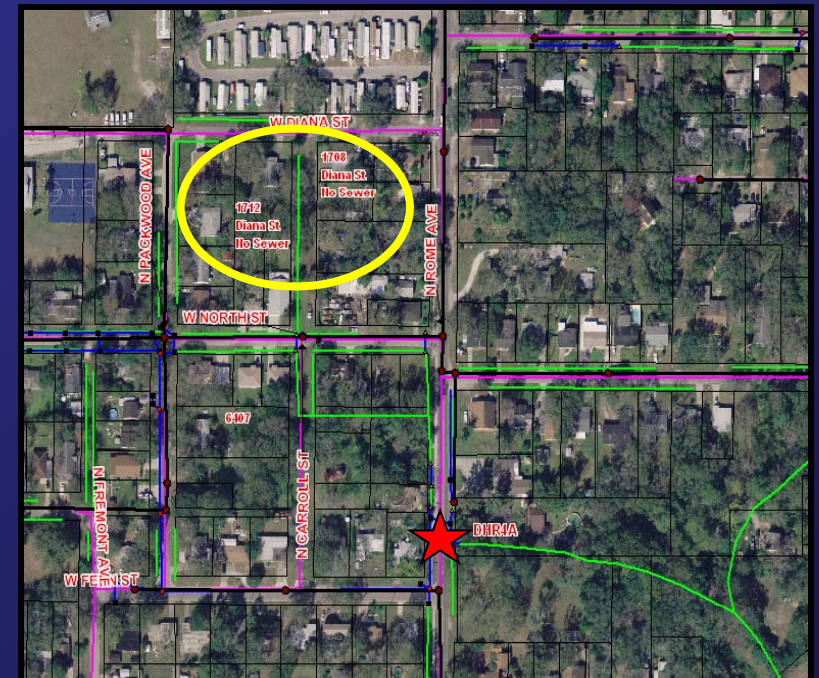
- Fecal Coliform
- *E. coli*
- Enterococci
- Fecal Coliform Regulatory Limit
- *E. coli* Regulatory Limit
- Enterococci Regulatory Limit
- Horse *Bacteroides* (Present)
- Horse *Bacteroides* (Absent)
- Ruminant *Bacteroides* (Present)
- Ruminant *Bacteroides* (Absent)
- ▲ *esp* (Present)
- △ *esp* (Absent)
- Human *Bacteroides* (Present)
- Human *Bacteroides* (Absent)
- ◆ HPyV (Present)
- ◇ HPyV (Absent)

**Consistent and extremely high counts isolated to DHR4A when no flow from upstream**



# COT/HCHD Investigation of West Jean Street Tributary

- Slipped joint identified ~225ft west of station DHR4A (likely only taking in water)
- Parcels within COT limits that do not have sewer accounts upstream of DHR4A (yellow circle)
- Standing water and brief sewage odor observed by COT at DHR4A in February 2008 despite lack of flow in contributing ditch, indicates local source
- Kennel of 4-5 dogs and 2 tents observed upstream by COT
- HCHD observed no evidence of OSTDS within at least 1 block north and south of DHR4A





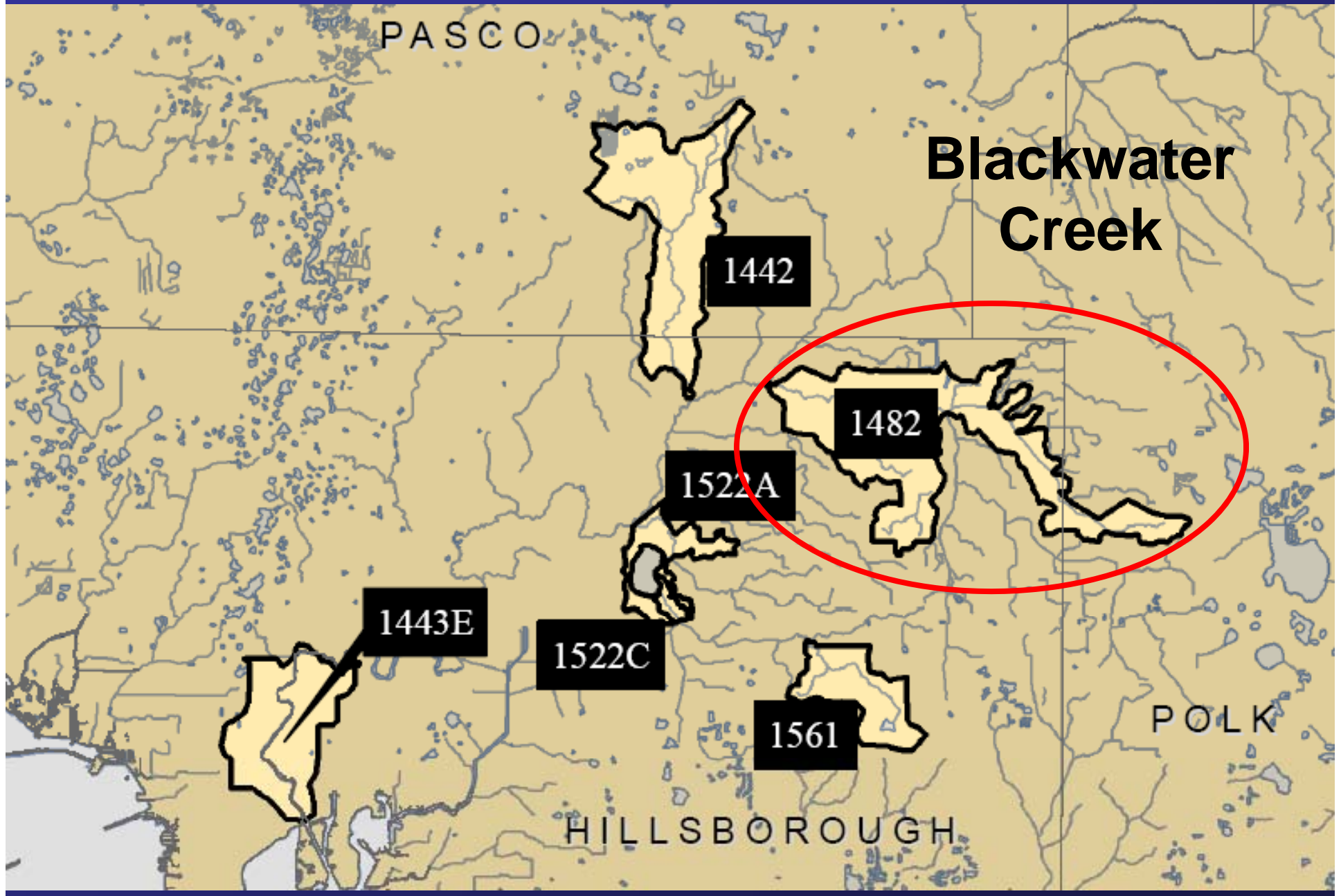
# HCHD and EPCHC Investigation

- Black corrugated pipe observed upstream between HR3 and DHR4A (March 25, 2008) by EPCHC
- Line of dark green grass led to residence and an area of the yard that was “soft and spongy”
- Samples downstream of pipe showed fecal coliform and enterococci exceedances
- HCHD found no evidence of sanitary nuisance (April 4, 2008)
  - Will attempt to contact owner about mandatory sewer connection due to proximity of available infrastructure





# Blackwater Creek (1482)

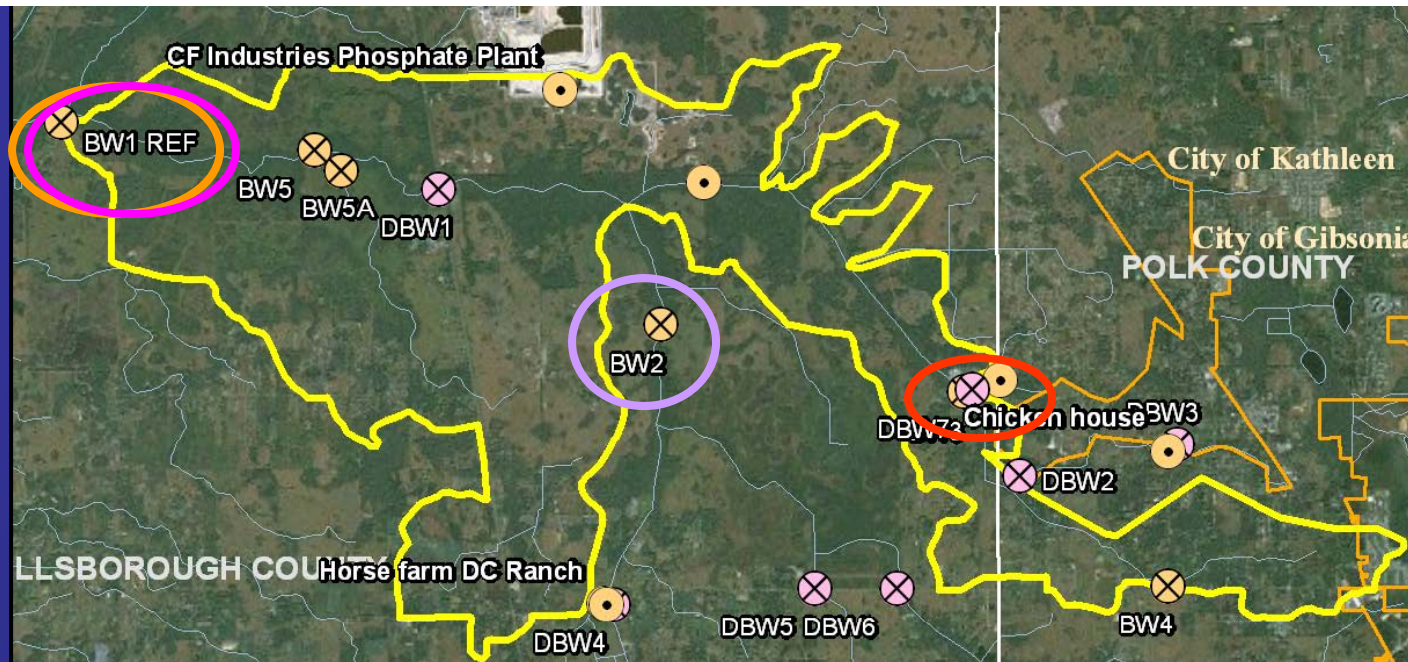


# Classification Matrix Outcome

## Blackwater Creek

Sub-Basin	WBID	Monitoring Location	Updated Classification Matrix Outcome	Sample Size (n) for Updated MWQA Score	Human Fecal Markers		Ruminant Fecal Marker	
					No. Sampling Dates	% Dates Marker(s) Detected	No. Sampling Dates	% Dates Marker Detected
Blackwater Creek	1482	<b>BW3</b>	D4	4	4	75 %	1	0 %
		<b>BW2</b>	D4	8	3	67 %	3	100 %
		<b>DBW4</b>	A3	7	1	100 %	1	0 %
		<b>BW5A</b>	A3	2	2	50 %	2	0 %
		<b>BW1 REF</b>	A2	8	7	43 %	6	17 %

Station	H-Bac	Esp	HPyV	R-Bac	Hr-Bac
BW1	2/5	0/5	1/5	0/4	0/4
BW5A	1/2	0/1	0/1	0/1	0/1
BW5	0/1	0/0	0/0	0/0	0/0
BW2	1/3	1/3	0/3	3/3	0/3
DBW4	0/1	1/1	0/1	0/1	1/1
BW3	1/4	1/4	1/4	0/1	0/1
BW4	0/1	0/1	0/1	0/1	0/1

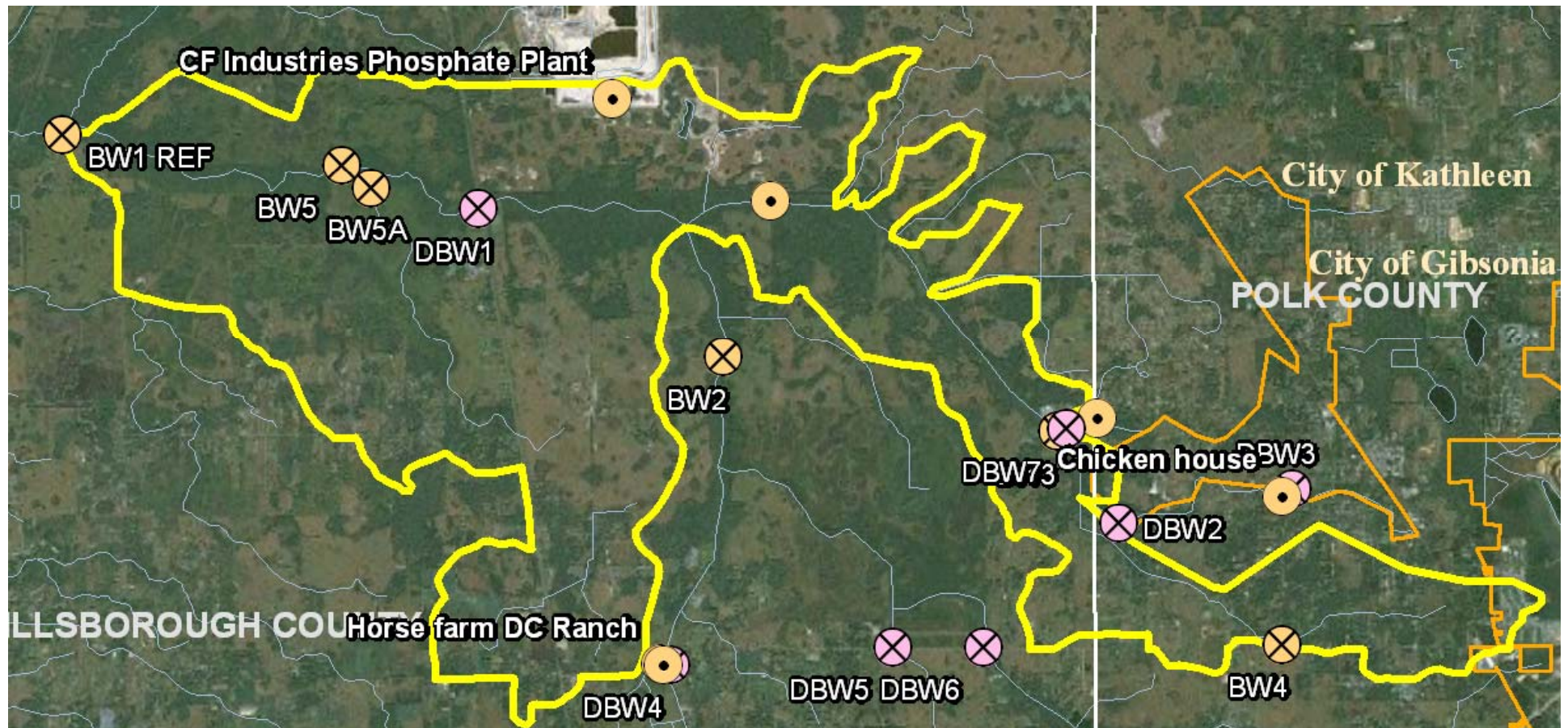


- Mixture of human-, ruminant- and horse-specific markers
- Extremely elevated IO concentrations in sediments at several stations

### Most Likely Sources

1. Sanitary Sewer / MHP Wastewater Facilities
2. Cattle
3. OSTDS
4. Wildlife





## Blackwater - WBID 1482

East-to-West flow

Several tributary streams intersect the main channel

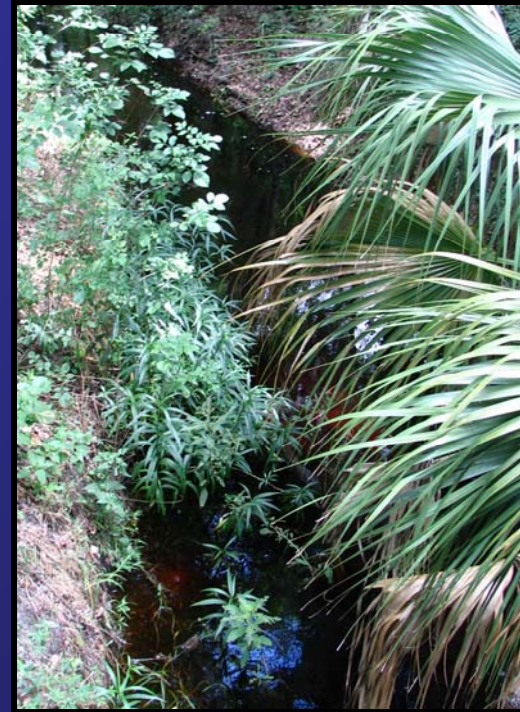
Potential source changes with area of WBID



# Blackwater Creek



**BW1**  
**Ref**



**BW3**



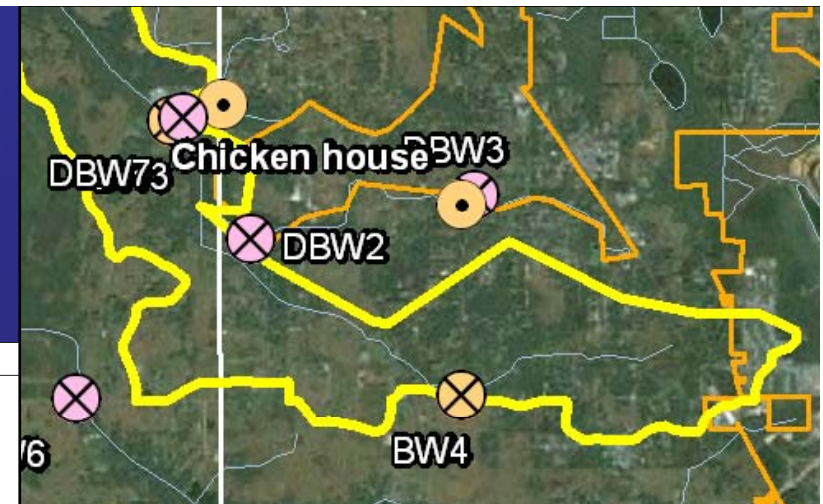
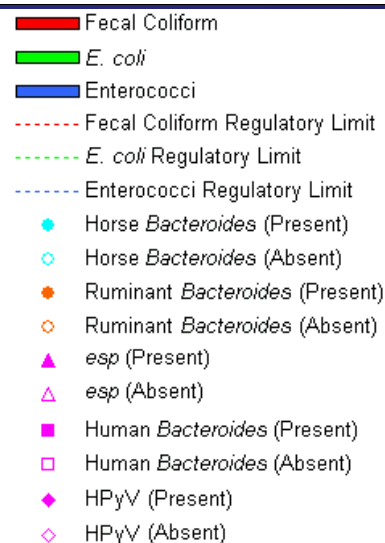
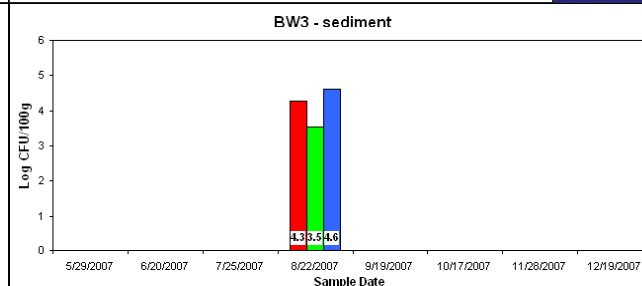
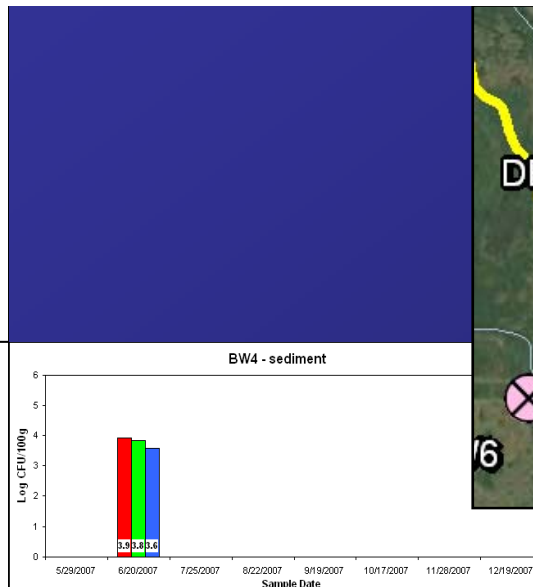
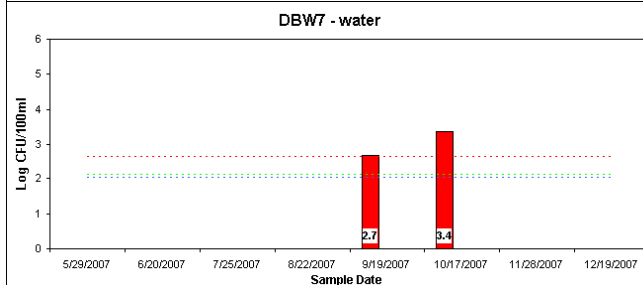
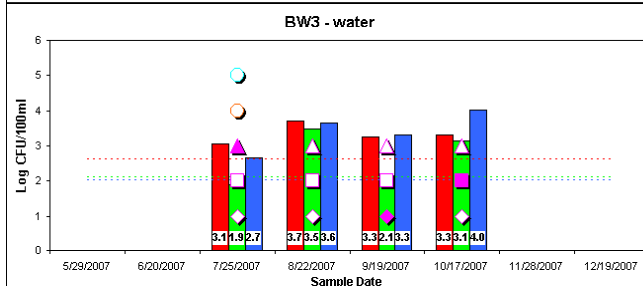
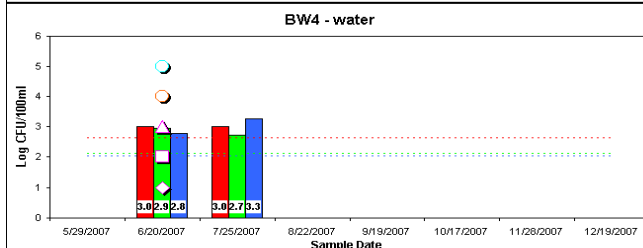
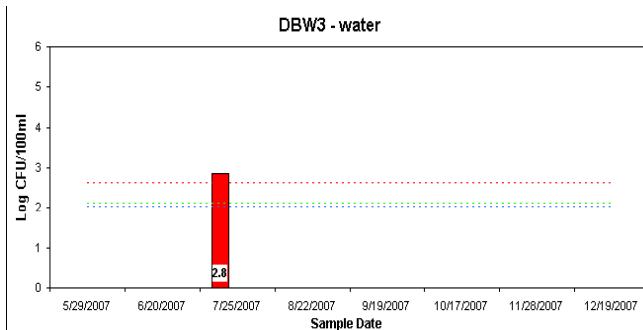
**BW2**



**BW4**

<b>Station</b>	<b>Month Added</b>	<b>Dry</b>	<b>No Flow</b>
DBW3	May	May, June, August, October	July, September
BW4	June	N/A	June, July
DBW2	May	Always	N/A
BW3	May	May, June, November, December	August
DBW7	September	November, December	N/A
DBW6	July	N/A	N/A
DBW5	July	July	N/A
DBW4	June	N/A	N/A
BW2	May	N/A	N/A
DBW1	May	May	N/A
BW5A	October	N/A	October
BW5	October	N/A	October
BW1 Ref	May	N/A	May, September, December





- Limited data due to lack of water and/or no flow
- All three human markers detected at BW3 (except Aug. when no flow but highest levels)
- All IOs show exceedances

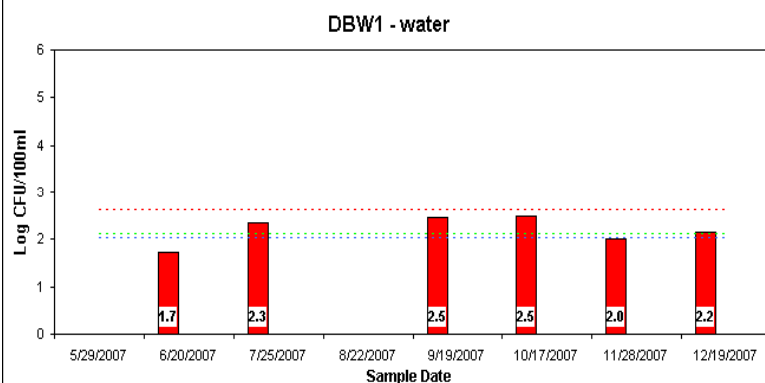
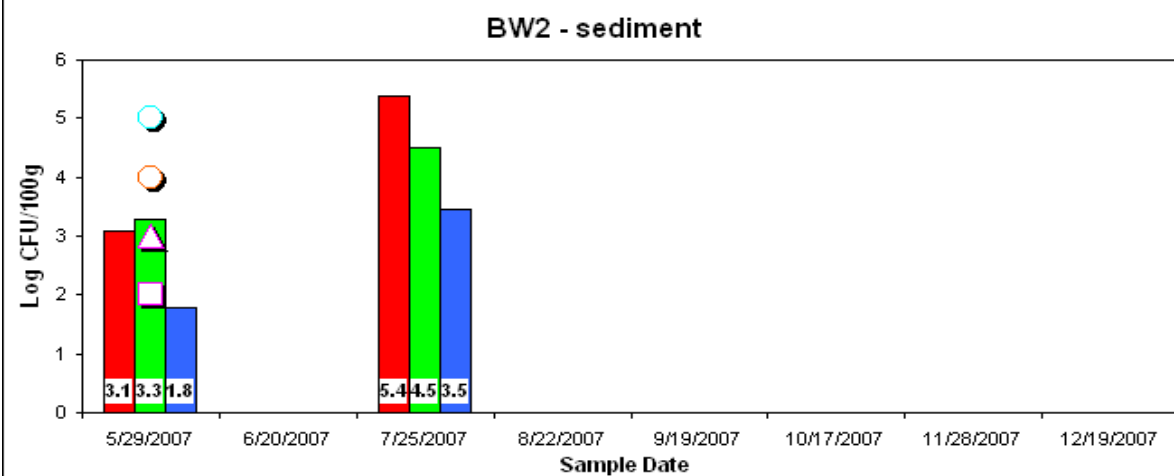
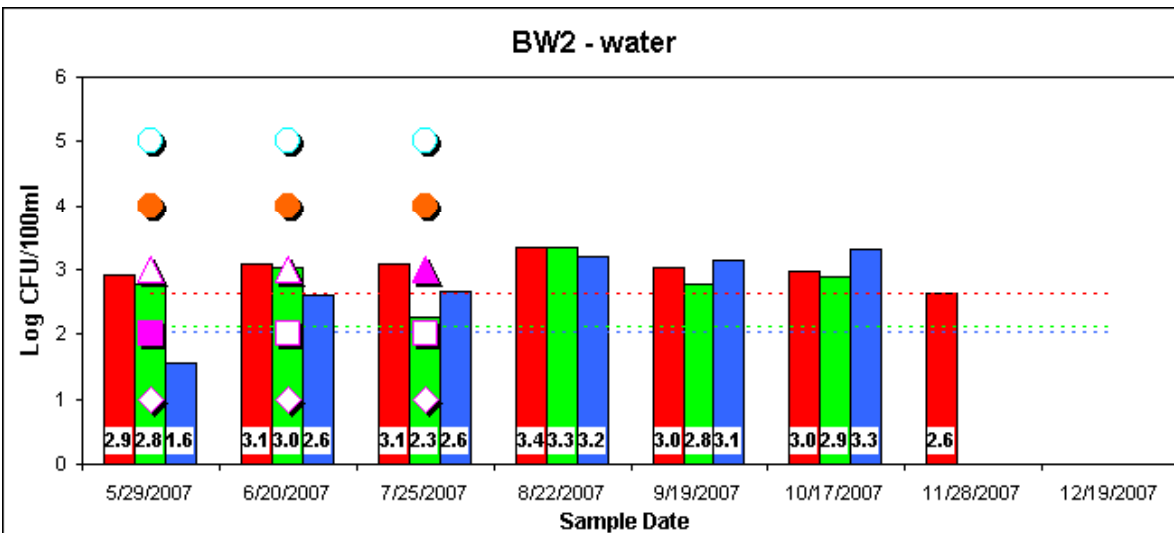
Likely multiple human sources



# DEP/EPCHC Investigation

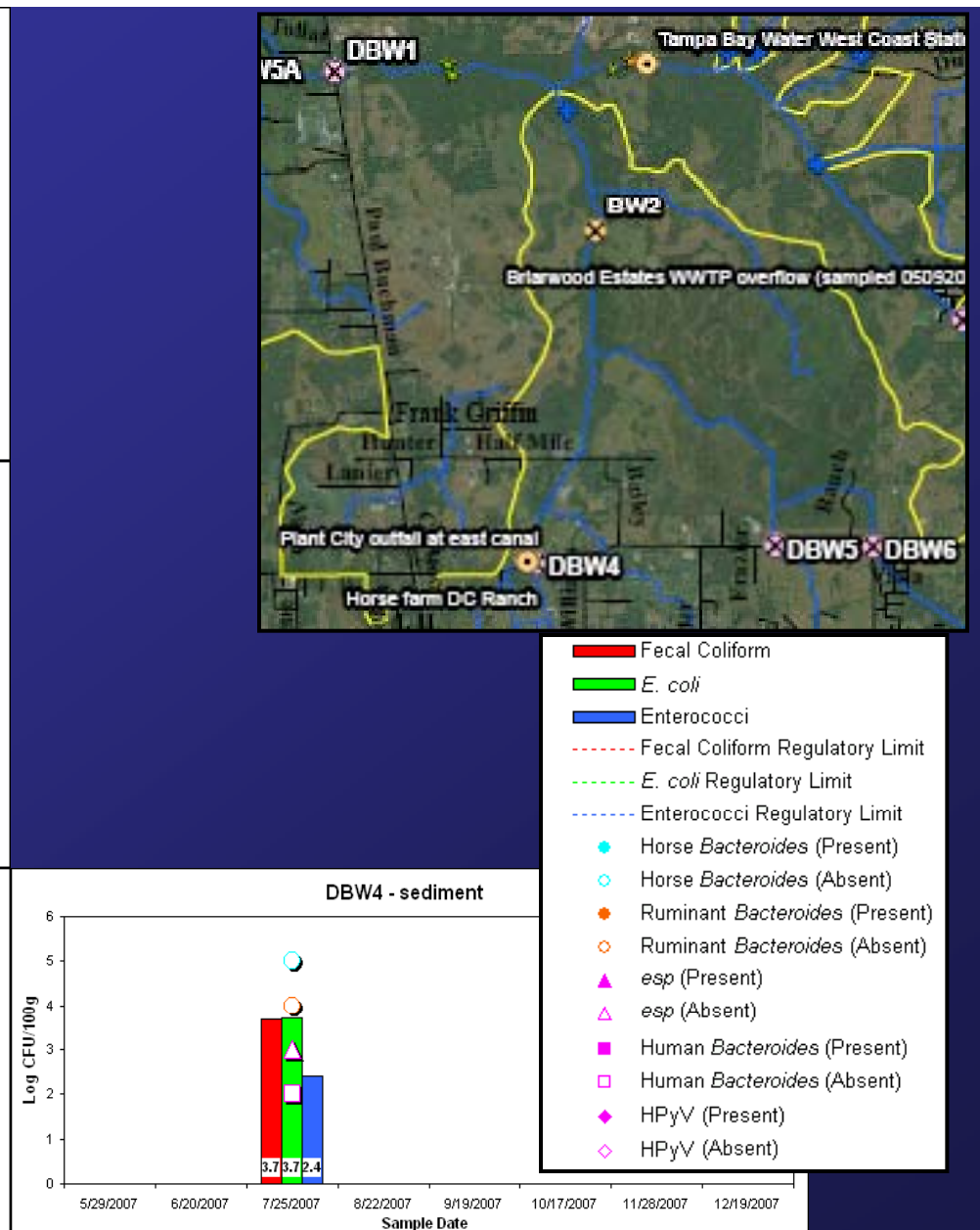
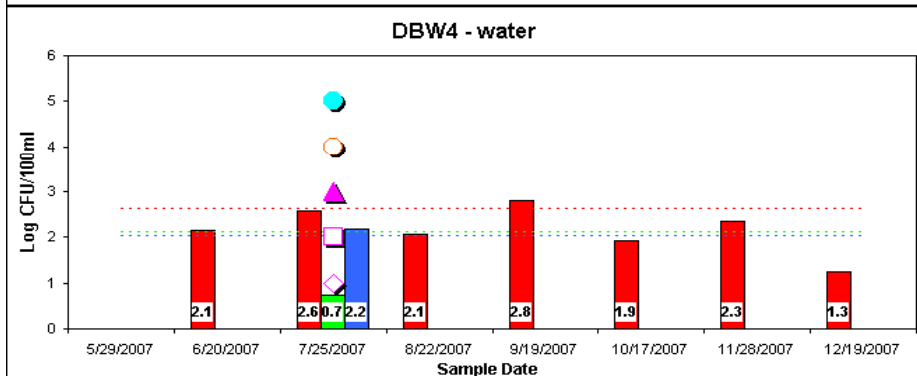
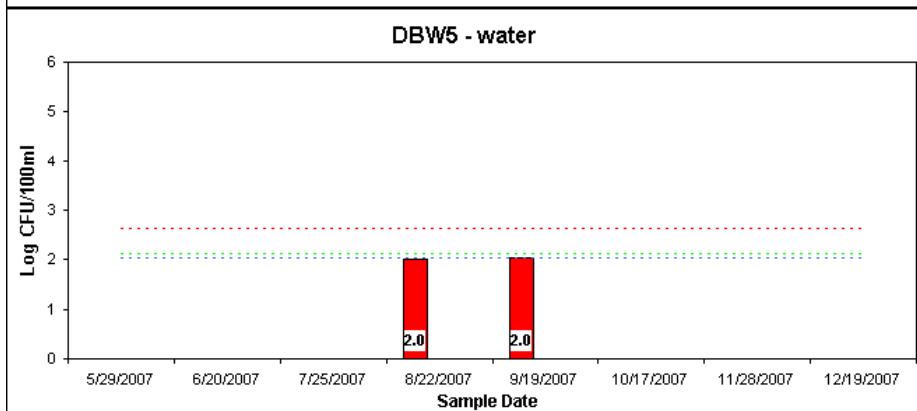
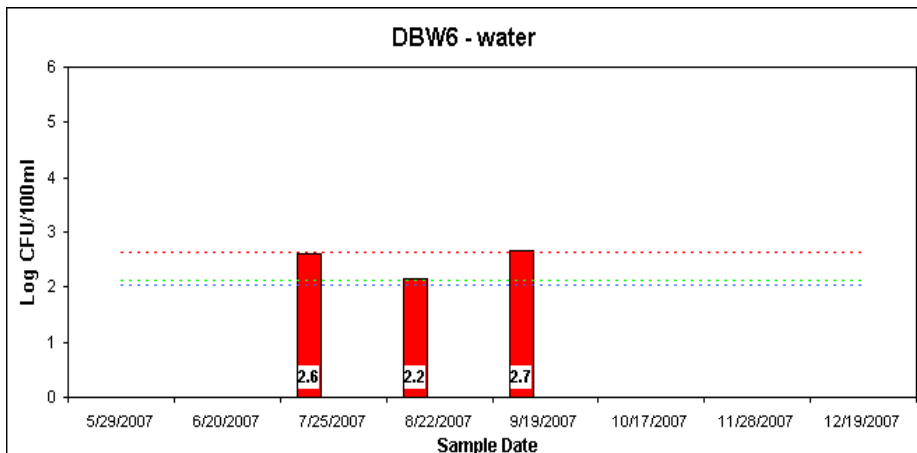
- Facility currently undergoing re-permitting for new 5-year cycle (including new temporary pond)
- Using temporary pond to gain freeboard; little freeboard observed still
  - May continue to be an issue in rainy season





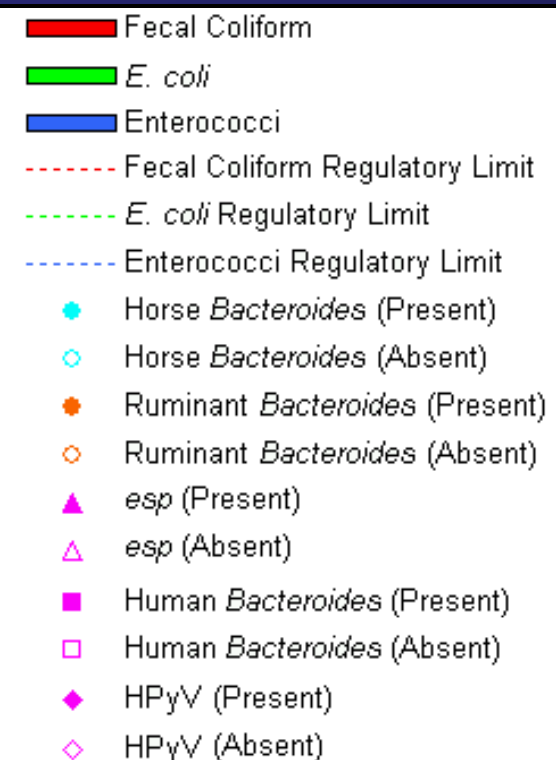
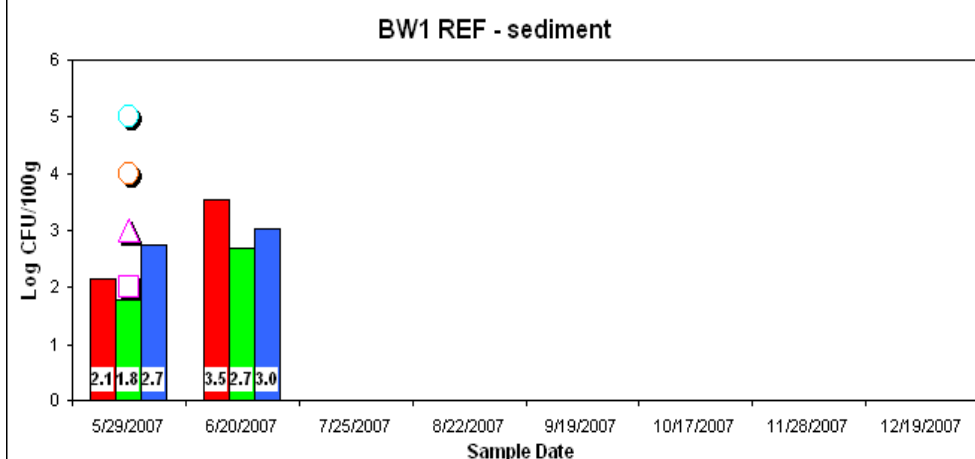
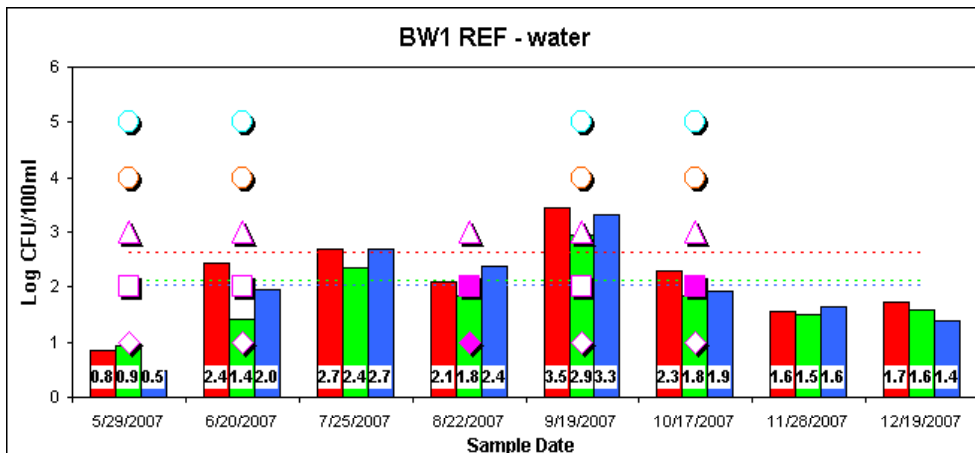
- Fecal Coliform
- *E. coli*
- Enterococci
- - - Fecal Coliform Regulatory Limit
- - - *E. coli* Regulatory Limit
- - - Enterococci Regulatory Limit
- Horse *Bacteroides* (Present)
- Horse *Bacteroides* (Absent)
- Ruminant *Bacteroides* (Present)
- Ruminant *Bacteroides* (Absent)
- ▲ *esp* (Present)
- △ *esp* (Absent)
- Human *Bacteroides* (Present)
- Human *Bacteroides* (Absent)
- ◆ HPyV (Present)
- ◇ HPyV (Absent)

**Unexpected  
human  
markers**



**Human markers explained by upstream station  
(no exceedances)**

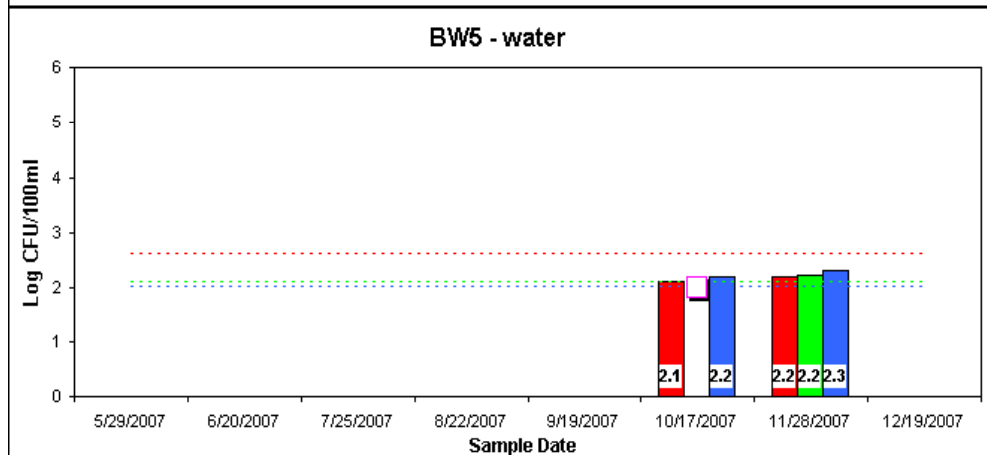
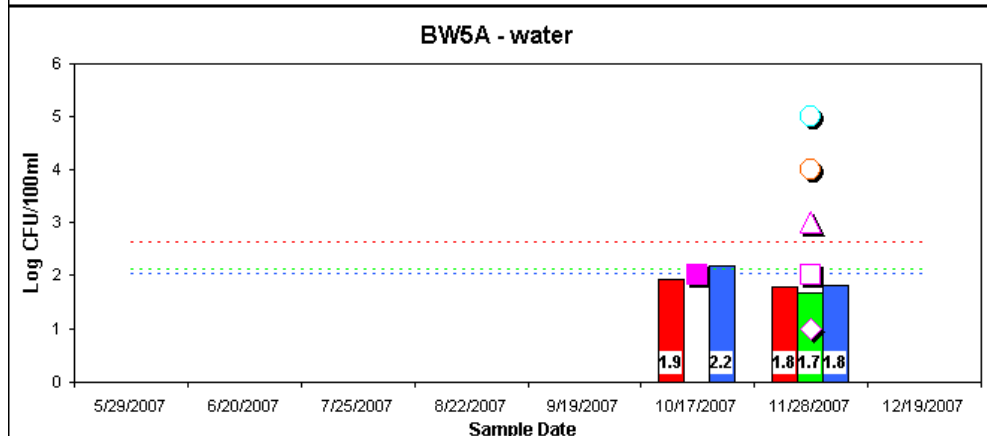
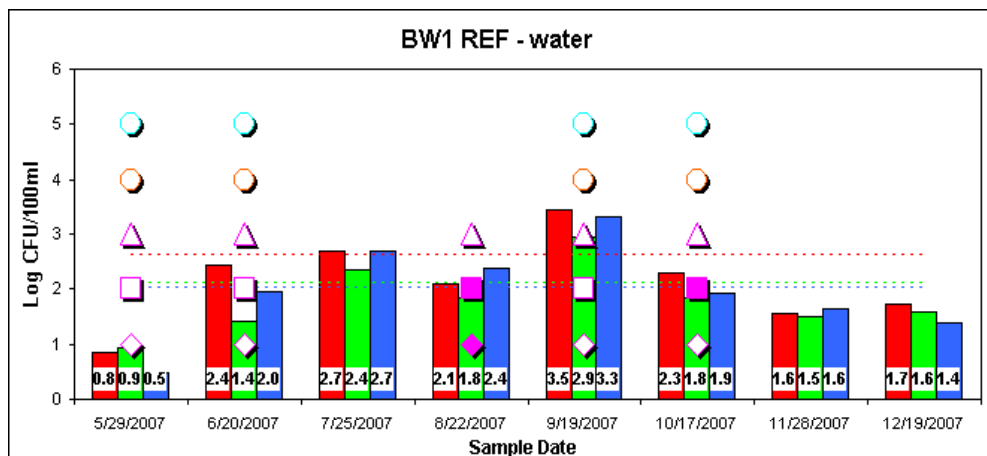




**Episodic exceedances at BW1**

**All 3 human markers at BW1; not associated with highest levels but during times of flow**





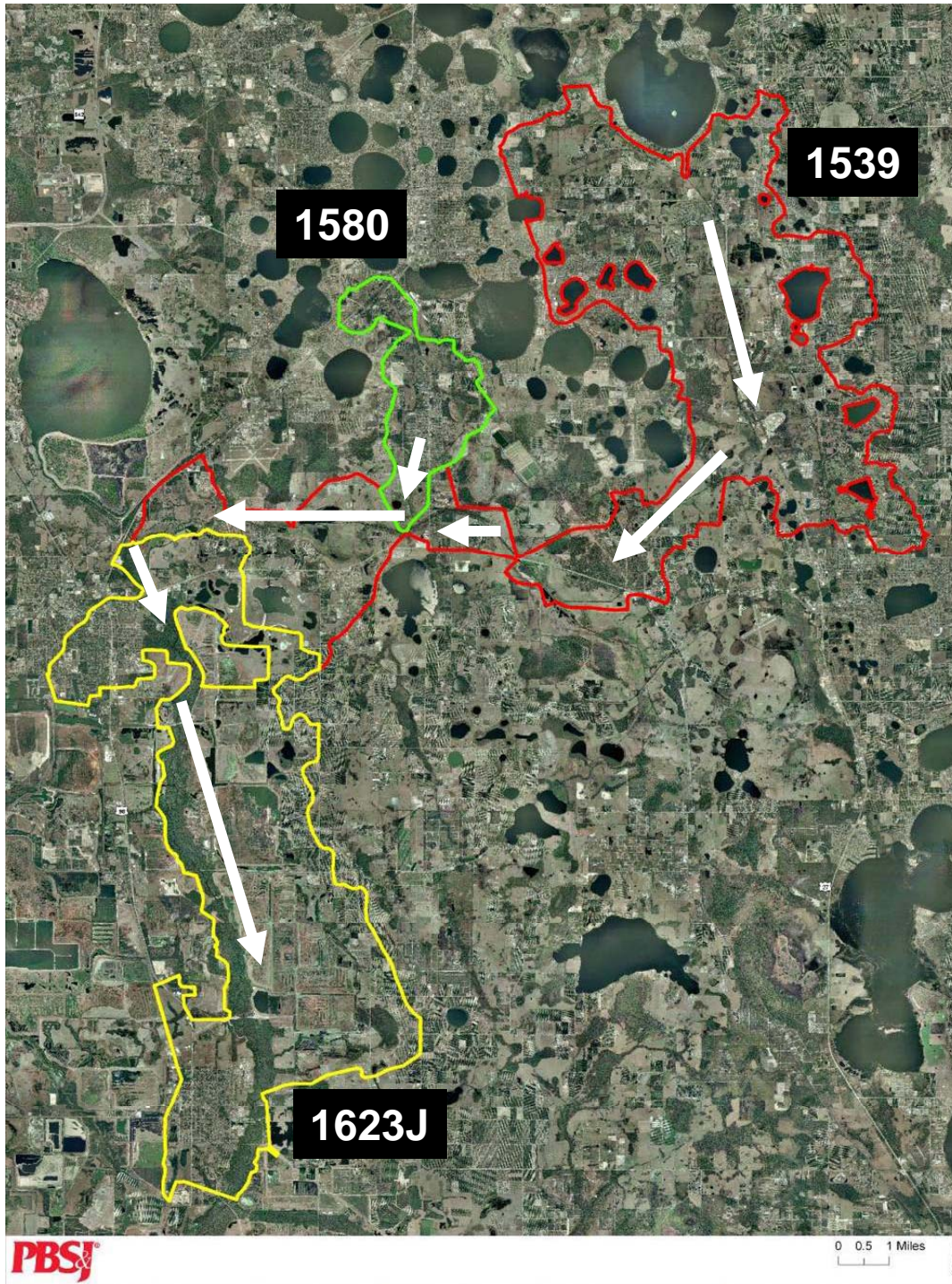
- Fecal Coliform
- *E. coli*
- Enterococci
- - - Fecal Coliform Regulatory Limit
- - - *E. coli* Regulatory Limit
- - - Enterococci Regulatory Limit
- Horse *Bacteroides* (Present)
- Horse *Bacteroides* (Absent)
- Ruminant *Bacteroides* (Present)
- Ruminant *Bacteroides* (Absent)
- ▲ *esp* (Present)
- △ *esp* (Absent)
- Human *Bacteroides* (Present)
- Human *Bacteroides* (Absent)
- ◆ HPyV (Present)
- ◇ HPyV (Absent)

**Exceedances likely due to upstream human source**

# Case Studies

## Impairment at Station Level

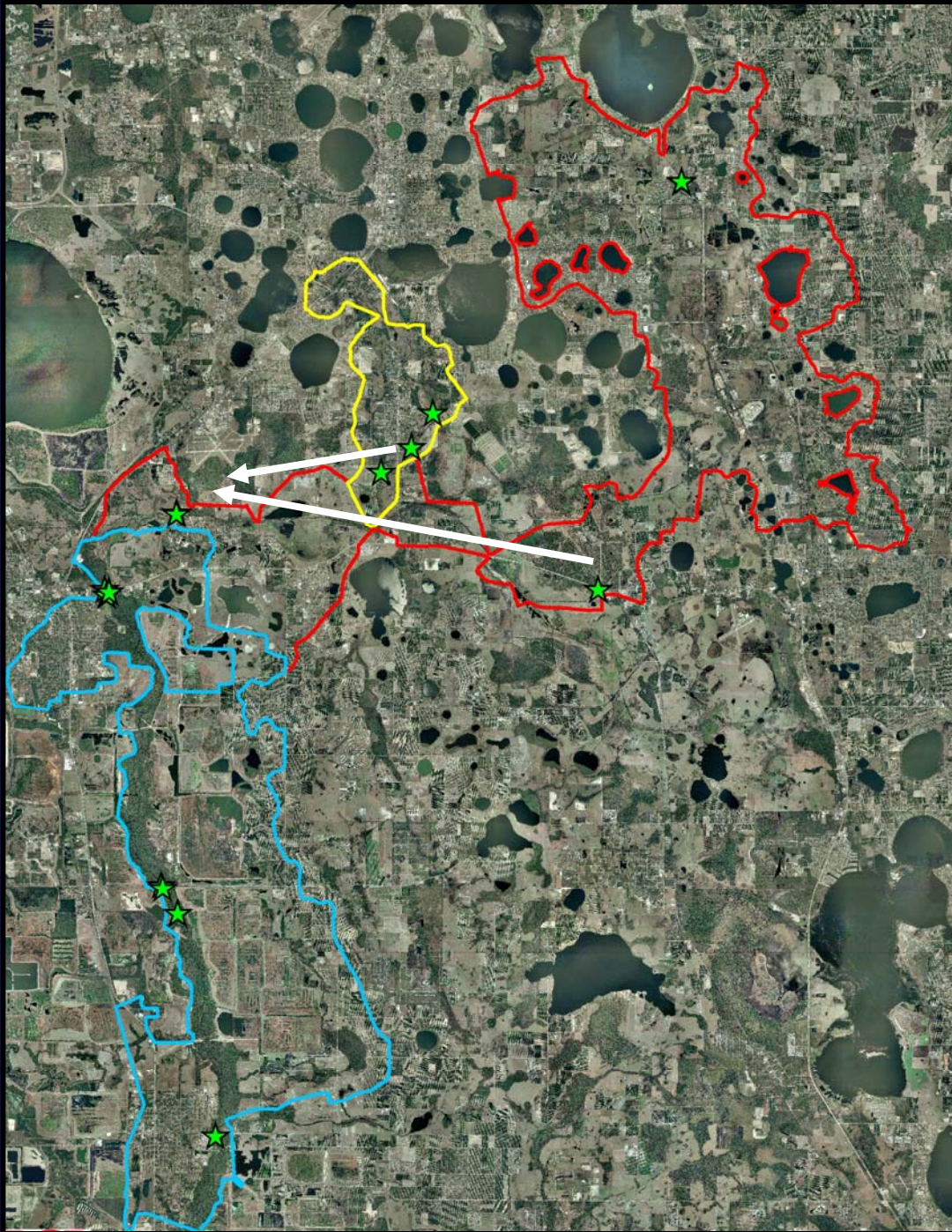
### Upper Peace Creek



**Part of an interconnected system which receives flow from upstream watersheds and transfers flow downstream**

**Redefine sampling methodology to better determine potential for transport**





# Transport?

## Statistical Comparison of fecal coliform concentrations between stations

- Upstream to Midstream- No significant relationship
- Midstream to Downstream- significant increasing
- Wahneta Farm Drain Canal downstream to Downstream- significant increasing



# **Case Studies**

## **Collaboration with Local Stakeholders**

### **Upper Peace Creek**

# City of Winterhaven

## Tour of WWTP #3



# FDACS

- Dissemination of key educational information to project team
- Leadership in education of stakeholders in implementing BMPs
- Facilitation for meetings and accessing local ranches
- Inclusion of Draft Summary of Agricultural Operations and Practices in Final Report



# Case Studies

**Limitations due to lab  
capacity and incorporation of  
improved MST assays**

**Lower St. Johns River**



# Academic MST Lab

## PROs

1. Reputable lab
2. Lower cost
3. Collaboration in field
4. Collaboration in evaluation and synthesis of information
5. Report review
6. Excellent, trustworthy working relationship

## CONs

1. Limited capacity
2. Limited funding for updating equipment and assays

# Commercial MST Lab

## PROs

1. Reputable lab
2. Higher capacity
3. More funding for updating equipment and assays

## CONs

1. Higher cost
2. Lack of collaboration in field
3. Lack of data synthesis
4. Working relationship not yet established

# **Suggestions**

- 1. Work to increase lab capacity and sharing of data among labs**
- 2. Lab inter-calibration / method validation project**