



### Outline

- Background
- Field
- Aquarium
- Conclusions
- Future work



# Indicator bacteria use for water quality management

- 303 (d) list
- Indicator bacteria include fecal coliform,
  Escherichia coli (E. coli) and Enterococci bacteria
  - Not generally harmful, correlated with pathogenic microorganisms that are present in human & animal digestive systems
  - High levels of indicator bacteria suggest increased risk of exposure to pathogenic microorganisms

# Sources of *E. coli* within urban streams & bayous in Harris County

- Contaminated runoff & stormwater
- Malfunctioning wastewater collection systems
- Improperly functioning wastewater plants
- Treated wastewater effluent
- Wildlife & domesticated animals
- Waterfowl
- Reservoirs of E. Coli include:
  - Algae & periphyton
  - Soils & sediments

### Sources of E. coli

- Source identification is the barrier to effective management
- Warm-blooded organisms such as mammals & also birds are vectors for these bacteria
- Recent studies identified reptiles as potential sources of *E. coli*
- Clark et al 2007, documented the presence of *E. coli* in some pelagic & demersal fishes
  - Origin of bacteria was Canadian geese & human sewage

# E. coli isolate frequency

Mammals	56%
Birds	23%
Crocodile	33%
Turtles	4%
Snakes	2%
Lizards	10%
Frogs	12%
Fish	10%

Omnivores	87.2%
Herbivores	70.0%
Carnivores	57.3%

From Gopee et al. 2000

From Gordon & Cowling, 2003

# POTENTIAL MECHANISMS OF E. COLI PRODUCT & TRANSPORT

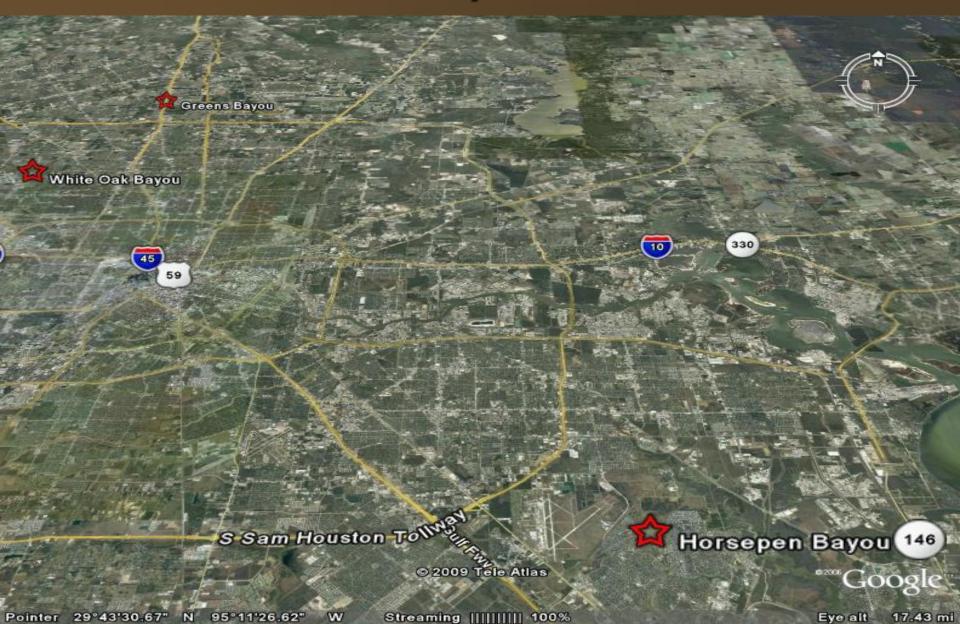
- Fish species can harbor E. coli (Guzman et al. 2004)
- Hansen et al. 2008 Pelagic & Benthic Fish:
  - Percentage of benthic fish harboring fecal coliforms not sig. diff.
    than Pelagic fish
  - Verified E. coli from Benthics 10x that of pelagics (42%, 4%)
  - Source identified for 65% of E. coli isolated
- Fecal coliforms found in every species examined, but not every fish
- Not dominated by a single strain
  - Fish may acquire microorganisms while feeding

### Objectives

To evaluate potential loading of *E. coli* from fish by:

- 1) Determining whether wild caught fish from Harris County waterways representing various species and trophic groups produce feces with detectable levels of indicator bacteria, *E. coli*
- Determining whether farmed fish retained in aquaria transmit *E. coli* bacteria to ambient water

# Field Study Locations





# Target Species

<b>Trophic Group</b>	Candidate Species
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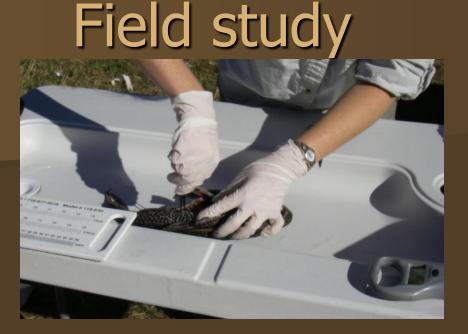
Herbivore	Armored Catfish
	Grass Carp
	Striped mullet
Benthic omnivore	Channel catfish
	Carp
	Smallmouth buffalo
Insectivore	Bluegill
	Rio Grande Cichlid
	Redear Sunfish
	Longear Sunfish
Omnivore	Gizzard or Threadfin shad
	Gulf Menhaden
	Tilapia
Piscivore	Largemouth Bass
	Spotted Bass
	Green Sunfish Spotted Gar
Benthic predator	Blue catfish



# Field study

- Fish Collection
  - Tote barge Electroshocker
  - Euthanized in MS-222







- Fecal Material Extraction
  - Fish measured & weighed
  - Large intestine removed
  - Fecal material extracted into pre-weighed
    Bacti-bottles with 100 ml of sterile water



Fecal Matter Extraction

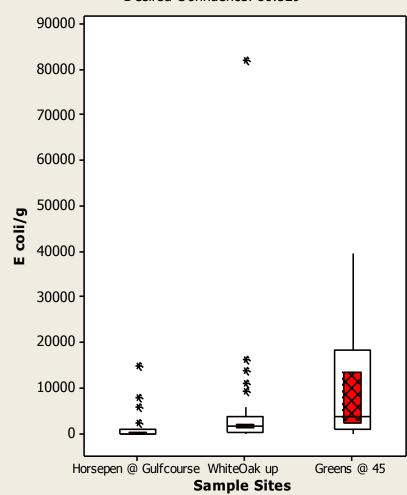


Laboratory Processing:

**IDEXX Method** 

# Kruskal-Wallis Multiple Comparisons with Dunn's Test

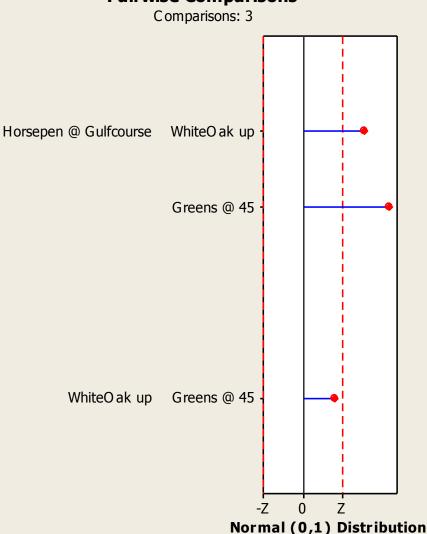




#### Family Alpha: 0.2

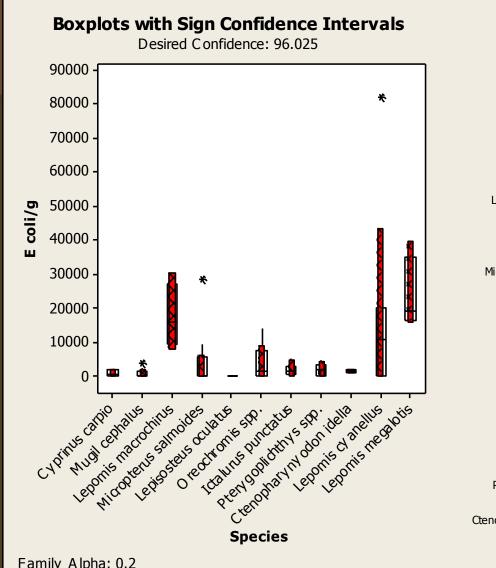
Bonferroni Individual Alpha: 0.067

**Pairwise Comparisons** 



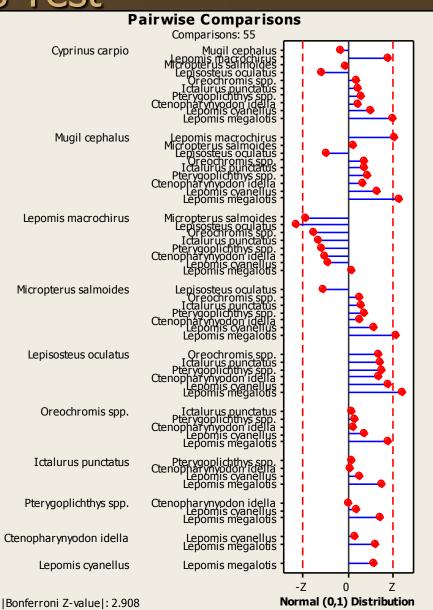
|Bonferroni Z-value|: 1.834

#### Kruskal-Wallis Multiple Comparisons with Dunn's Test



Family Alpha: 0.2

Bonferroni Individual Alpha: 0.004



# Kruskal-Wallis Multiple Comparisons with Dunn's Test

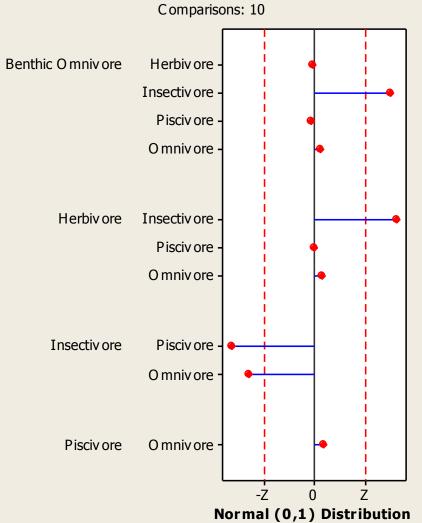
#### **Boxplots with Sign Confidence Intervals**

**Trophic Level** 

Family Alpha: 0.2

Bonferroni Individual Alpha: 0.02

#### **Pairwise Comparisons**



|Bonferroni Z-value|: 2.326

## Aquarium Study

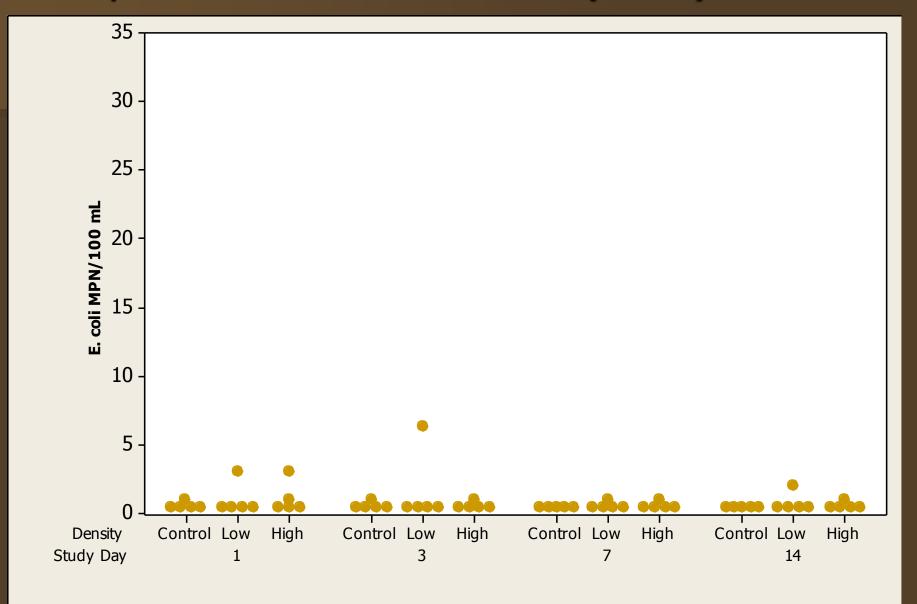
- 1<sup>st</sup> round used Bluegill (*Lepomis macrochirus*)
  - 2<sup>nd</sup> round still in progress with channel catfish
- 15 15 gal. aquaria
  - 5 replicates
    - Control
    - Low density (1 fish/tank)
    - High Density (3 fish/ tank)
- Basic WQ parameters monitored daily
  - Additional parameters measured on bacteria sampling days

# Aquarium Study

- Bacteria sampled
  - Pre-stocking
  - 1d post-stocking
  - 3d post-stocking
  - 7d post-stocking
  - 14d post-stocking



### Aquarium Water *E. coli* (MPN) Values



#### Conclusions

- Fish in the stream study seem to be transporters of *E.coli*
- Supported by aquarium study using farmed fish
  - Showed no increase in *E.coli* levels due to stocking density or over time
  - Fish fecal matter tested after 14d aquarium study showed low levels of *E. coli*
- Diet plays an important role in amount of *E. coli* in fish fecal matter

### Conclusions/Future Implications

- Bacteria in fish maybe indicators of bacteriological pollution in the waterbodies they inhabit
- Fish seem to absorb the bacteria from their food as well as their environment
- Therefore fish maybe significant transport mechanisms not captured by current modeling efforts
- Future studies on isolating bacteria strains need to be conducted to determine source loadings

#### Future Work

- Spring & Summer field sampling
  - Determine if there is a seasonal difference in bacteria levels in the fish fecal matter
- Aquarium Study with channel catfish
  - Difference in fish species
  - Preliminary results similar to bluegill
    - After 7d all tanks have <1 MPN of E. coli</p>

