Population Dynamics of *Escherichia coli* and *Enterococcus* spp. in Buffalo Bayou and White Oak Bayou

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A comprehensive, multi-year study of the fate and transport of *E. coli* and *Enterococcus* spp. to address several questions:

1. ***Under what conditions can E. coli and Enterococcus spp. bacteria survive for extended periods in the natural waters and sediment in Buffalo Bayou and White Oak Bayou and in the soils in the watersheds?***

2. ***Under what conditions can E. coli and Enterococcus spp. bacteria replicate (grow) in natural waters and soils in Buffalo Bayou and White Oak Bayou?***

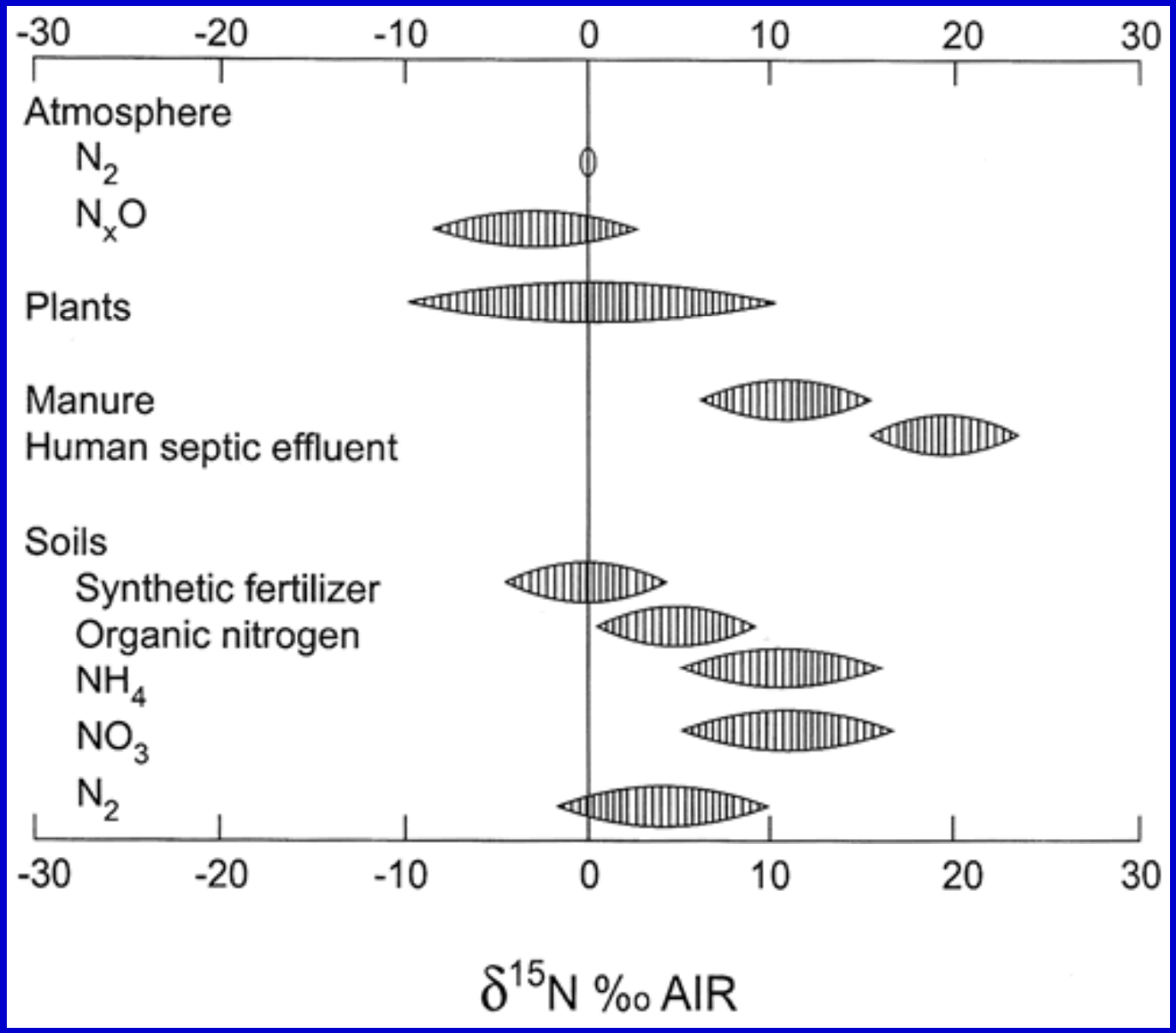
Questions 1& 2 -Sampled from January to August 2008 to assess influence of temperature and water flows

- Analysis of nutrients in water, sediments (from pore water) and soils
- FISH and Micro-FISH (microautoradiography combined with FISH-determines activity) analysis of all substrates with *E. coli* and *Enterococcus* specific DNA probes *(in progress)*
- Traditional EC detection methods for comparison
- Laboratory incubations using substrates to determine if *E. coli* and *Enterococcus* will replicate or are only viable but non culturable (VBNC) (*in progress and focus of FY2009)*
- Determination of false-positives produced with traditional FC detection methods using DNA probes *(in progress)*
- HFERP fingerprinting to determine 'naturalized' indicator bacteria *(in progress)*

3. Under the conditions indicator bacteria are found to replicate in natural waters and sediment, what are they using for growth substrates?

Are they utilizing WWTP effluent derived substrates from chlorine or UV based processes or from other substrates found in the waters and sediments?

- **End member chemical characterization of organic sources (awaiting results from contract lab)**
- **Stable isotopic composition of bacteria that replicate in natural substrates (in progress)**



Typical δ15 N values of various nitrogen sources in the environment.

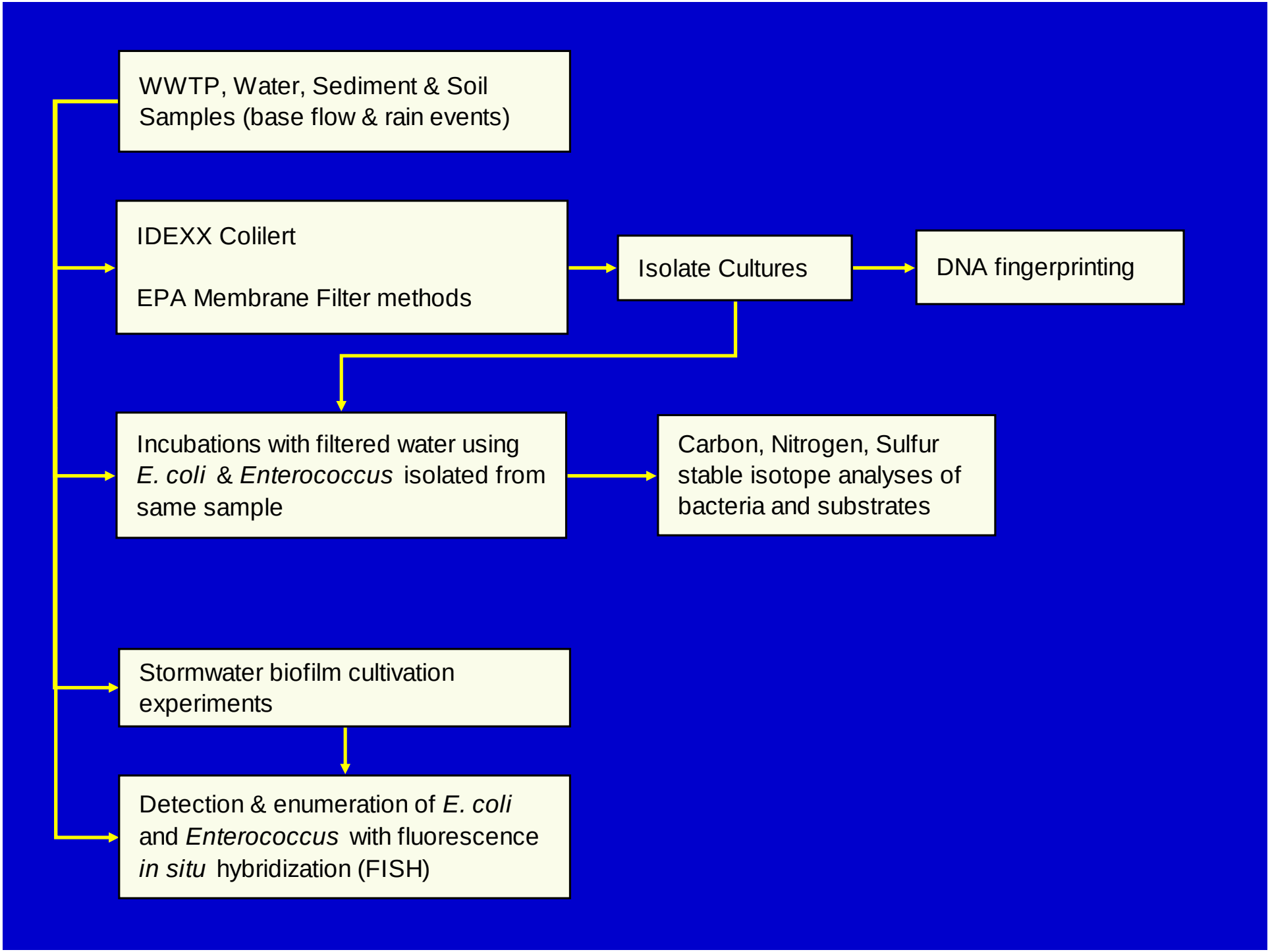
4. ***Are these indicator bacteria attached to particulate matter in the watersheds or surviving on biofilms in discharge conveyance systems and are then flushed into bayous during rain events?***

- **FISH analysis of biofilms and particulate matter collected during storm events (focus of FY2009)**
- **Determination of biofilm growth on microscope slides affixed in discharge conveyance systems (in progress)**

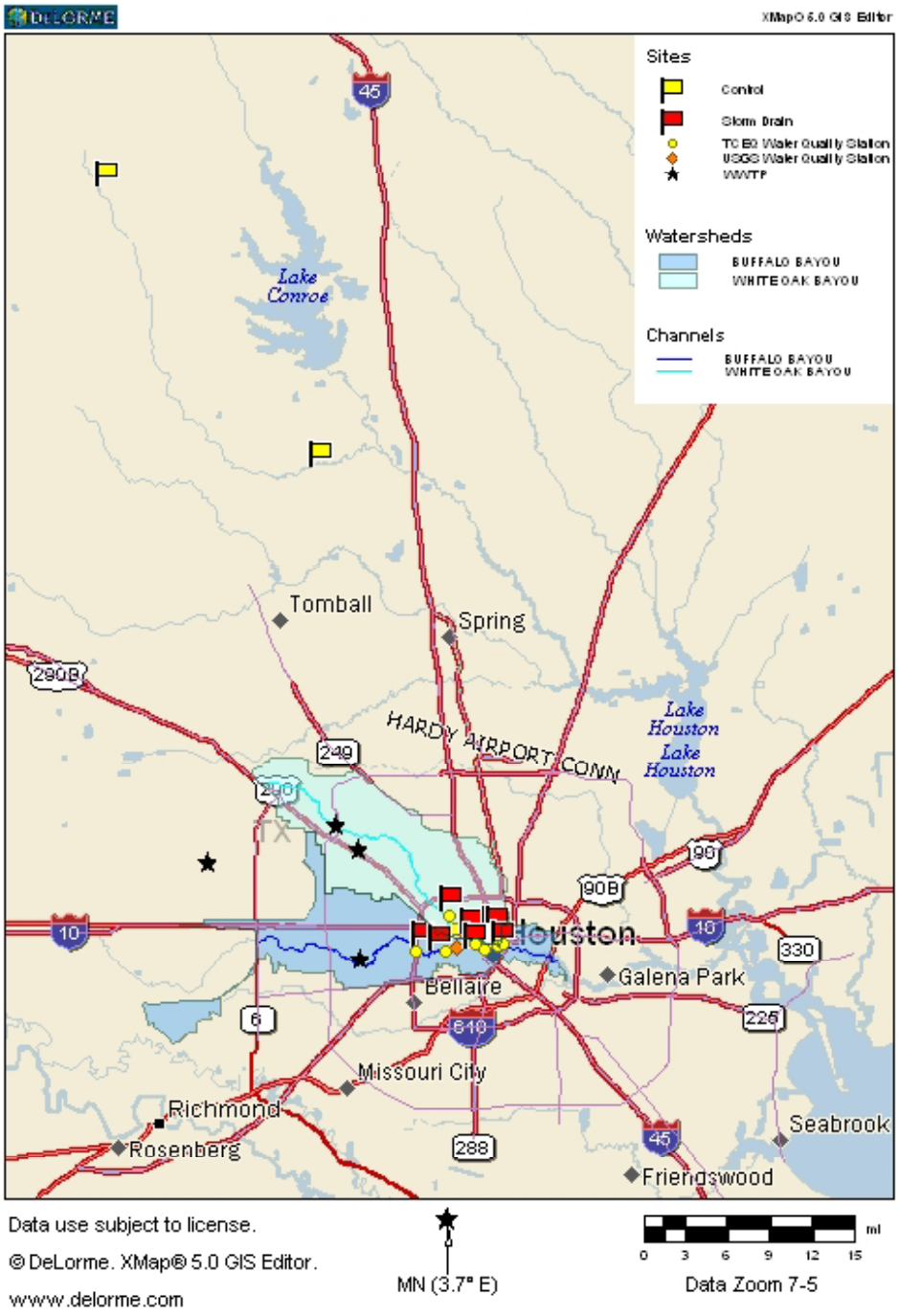
5. *How does the hydrologic cycle influence survival and replication of these indicator bacteria?*

- **Sampling during storm events.**
- **Analyses to include nutrients, DOC, FISH, IDEXX**
- **Comparison of strain genotypes from soils and sediments to cultivated isolates.**

Focus of FY2009

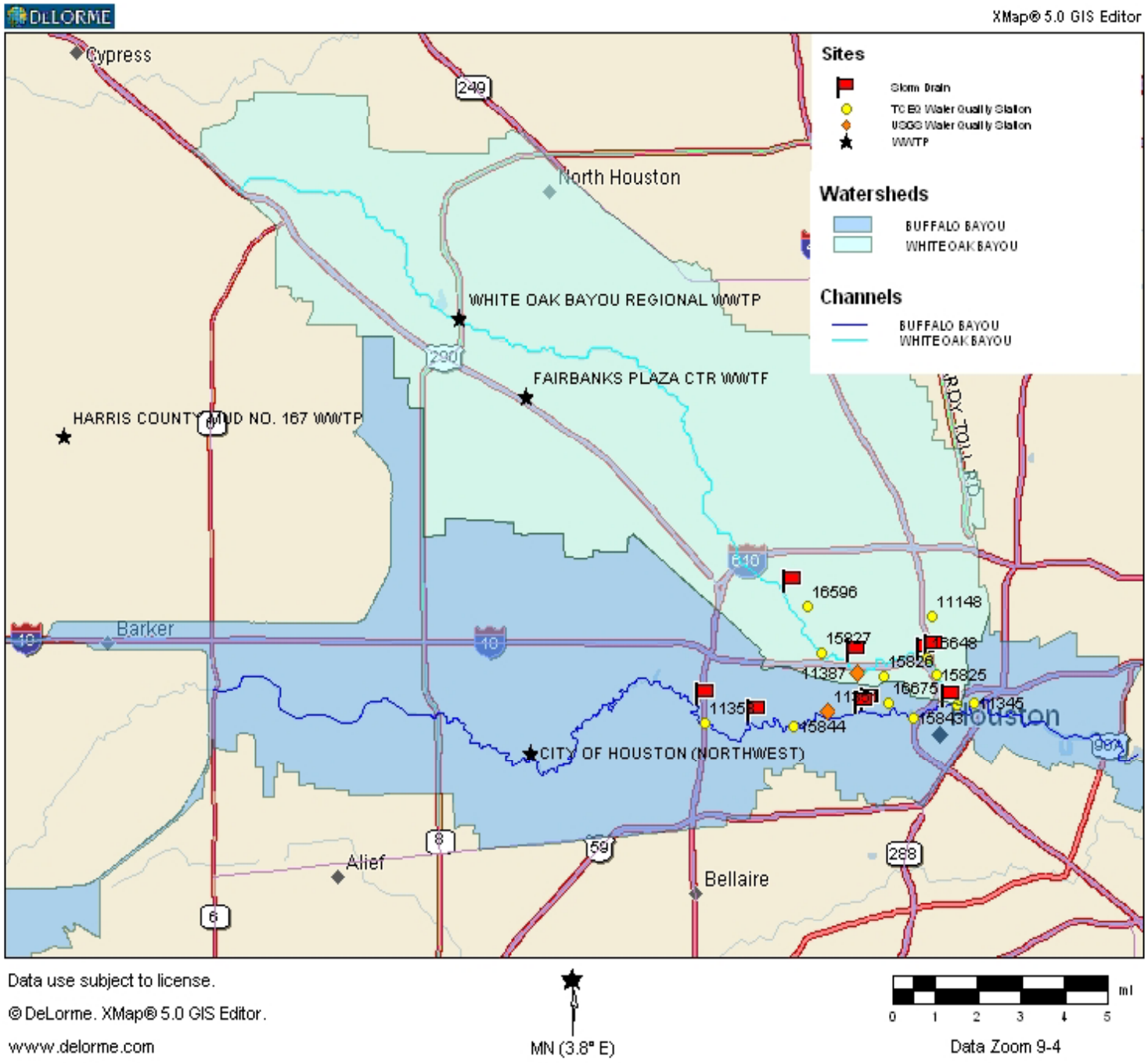

WWTP, Water, Sediment & Soil Samples (base flow & rain events)
IDEXX Colilert
EPA Membrane Filter methods
Isolate Cultures
DNA fingerprinting
Incubations with filtered water using *E. coli* & *Enterococcus* isolated from same sample
Carbon, Nitrogen, Sulfur stable isotope analyses of bacteria and substrates
Stormwater biofilm cultivation experiments
Detection & enumeration of *E. coli* and *Enterococcus* with fluorescence *in situ* hybridization (FISH)

2 Control Sites on Lake Creek in the Caney Creek Watershed



Year 1

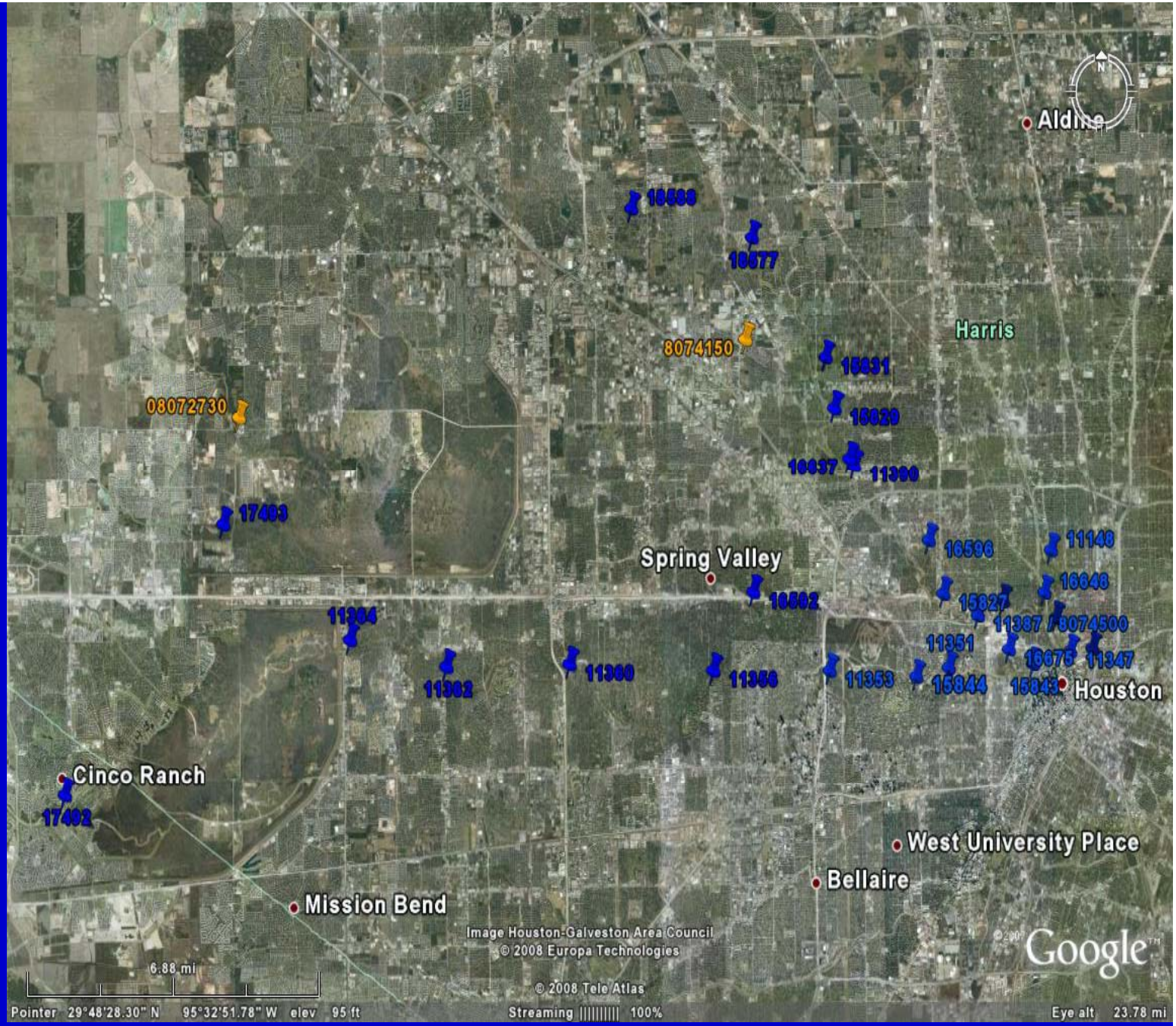
Sampling Sites



In Spring 2008, learned that FY2009 funding would be reduced by more than 50%.

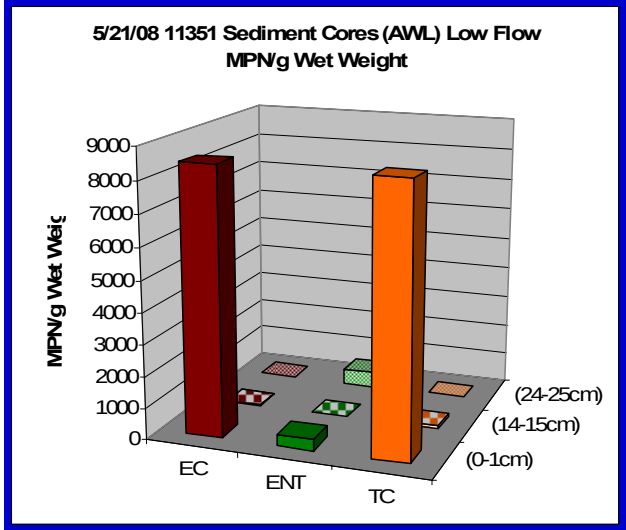
Increased FY2008 budget to include 15 additional sampling sites and QPCR & DNA probing analyses.

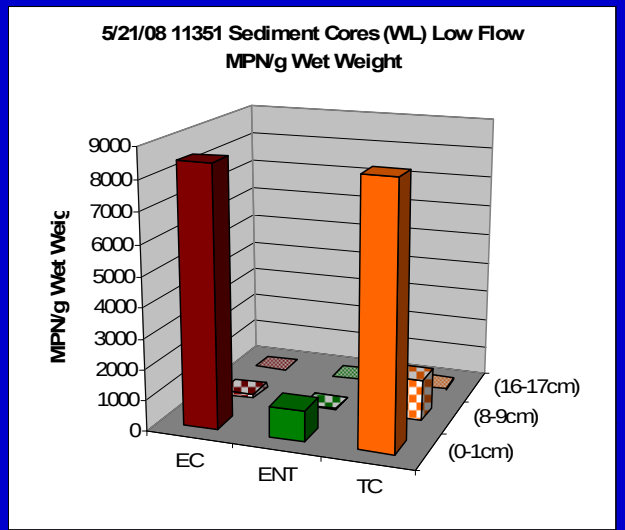
Trade offs- more samples but less time in FY2008 for processing and FY2009 reduction in research staff.

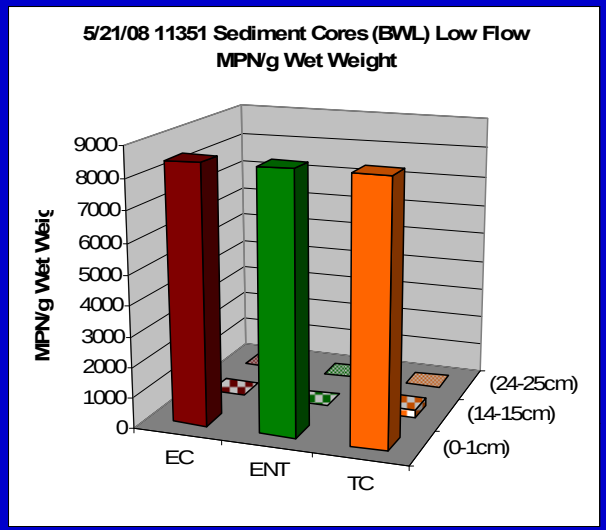


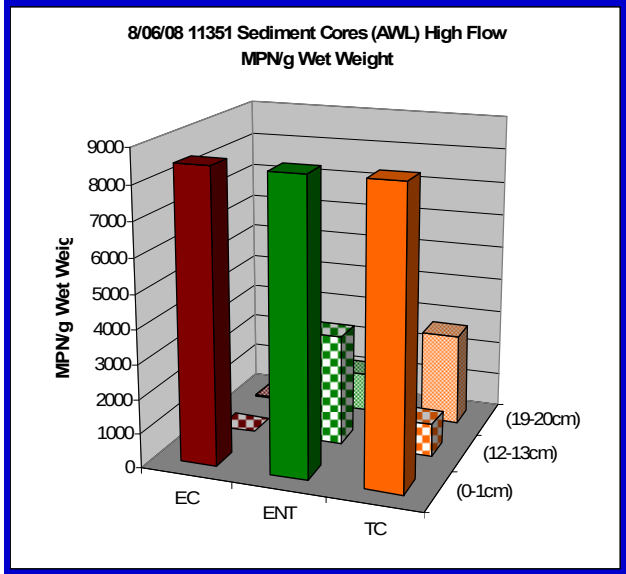

MADE IN

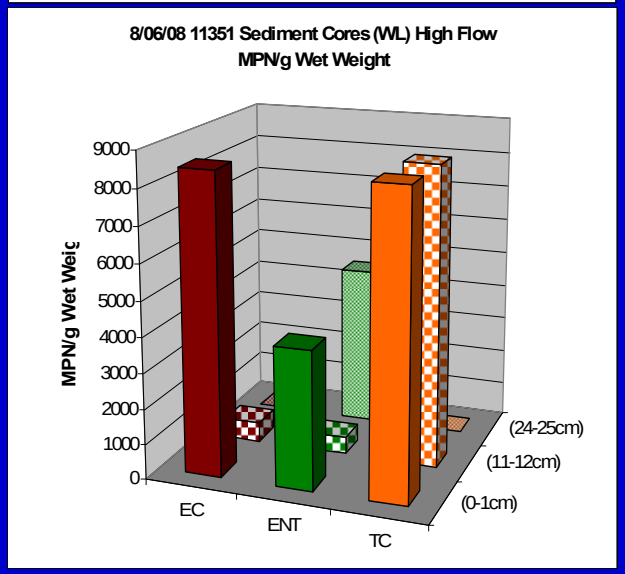
Sediment Cores collected Above the Water Line (AWL), at Water Line (WL) & Below Water Line (BWL). Three horizons sub-sampled: top (0-1cm), mid (variable), bottom (variable).

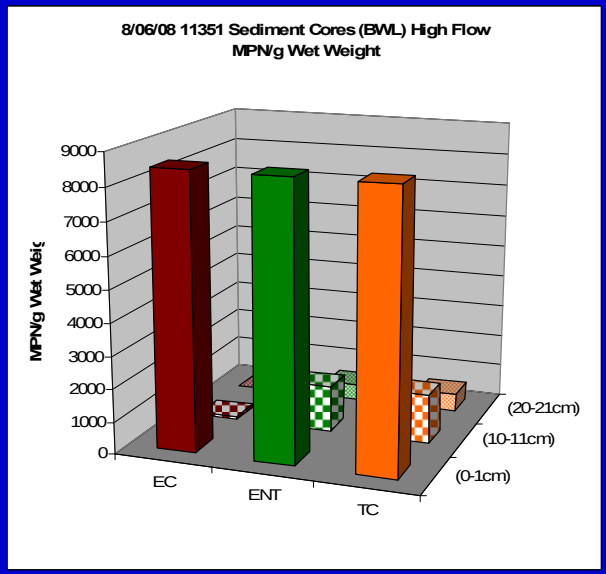






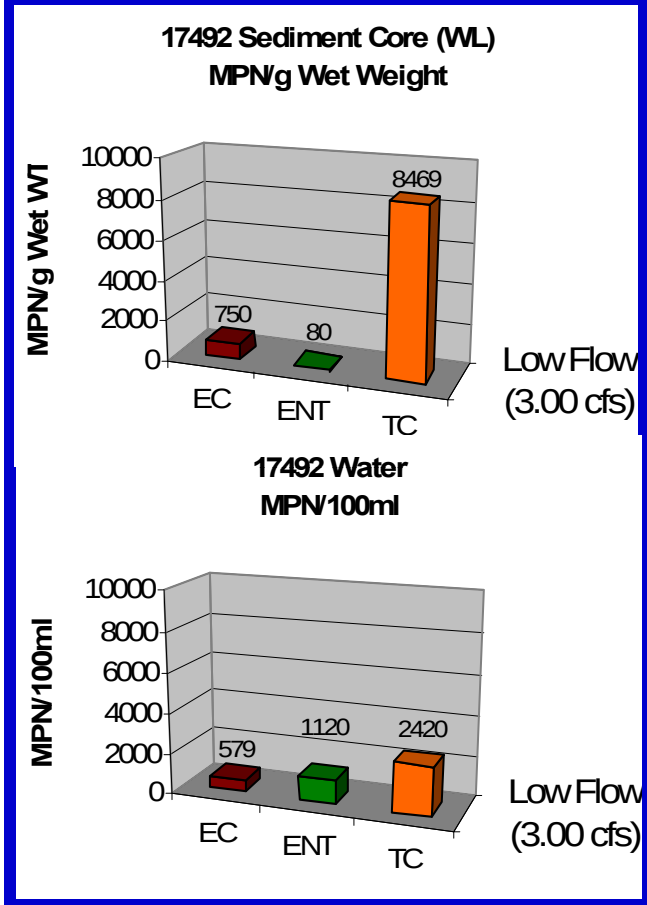


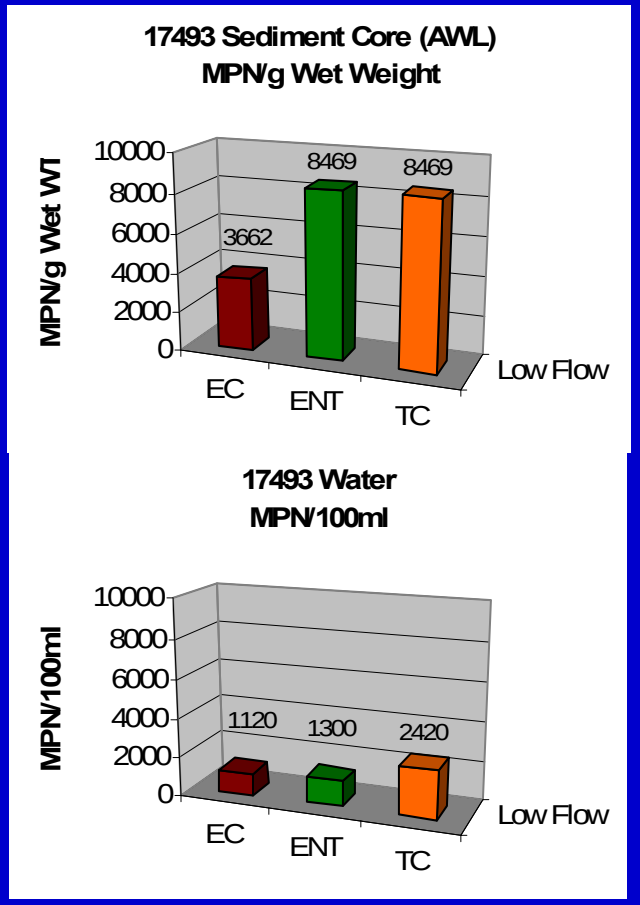


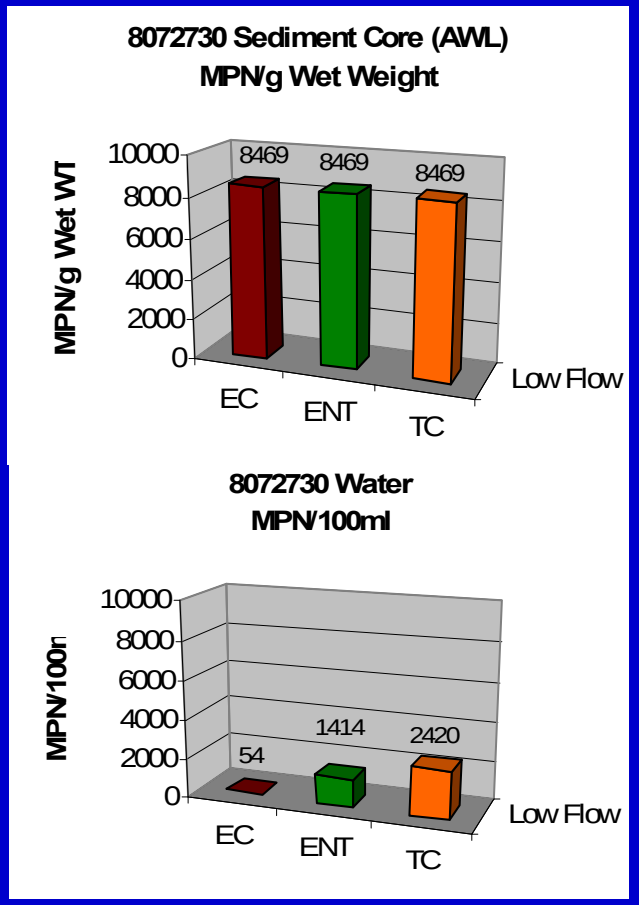


Highest bacterial concentrations typically in top (0-1 cm) horizon

Buffalo Bayou (sediment cores- top horizons)

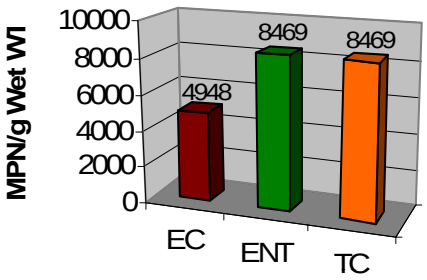






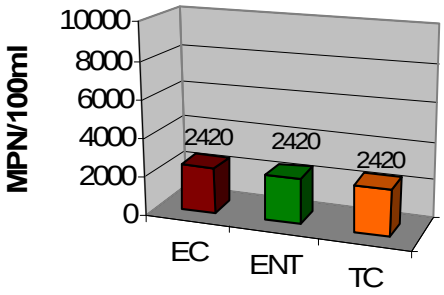
Buffalo Bayou (sediment cores- top horizons)

11364 Sediment Core (WL)
MPN/g Wet Weight



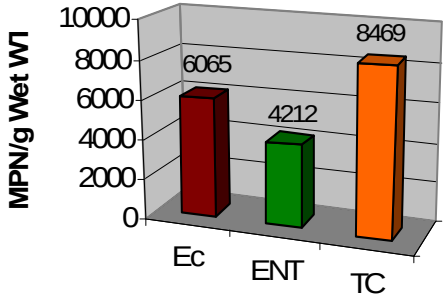
Low Flow (92 cfs)

11364 Water
MPN/100ml



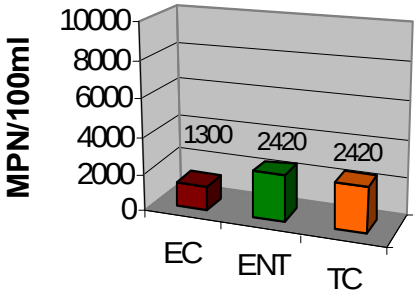
Low Flow (212 cfs)

11362 Sediment Core (WL)
MPN/g Wet Weight



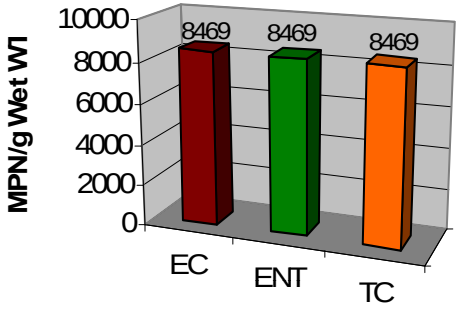
Low Flow (227 cfs)

11362 Water
MPN/100ml



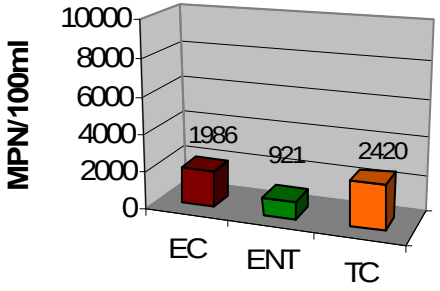
Low Flow (227 cfs)

11360 Sediment Core (AWL & WL)
MPN/g Wet Weight



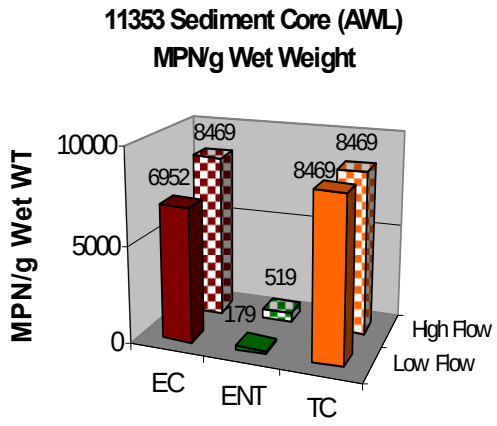
Low Flow (202 cfs)

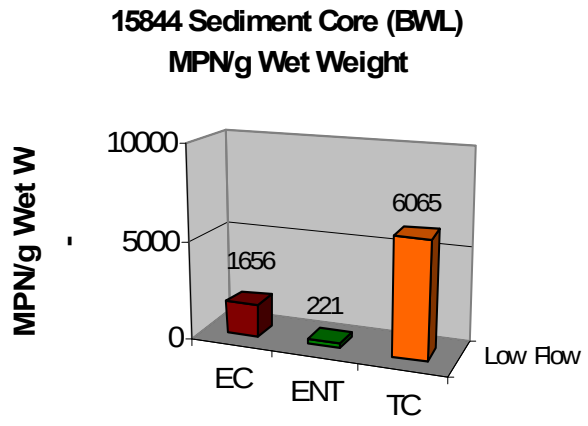
11360 Water
MPN/100ml

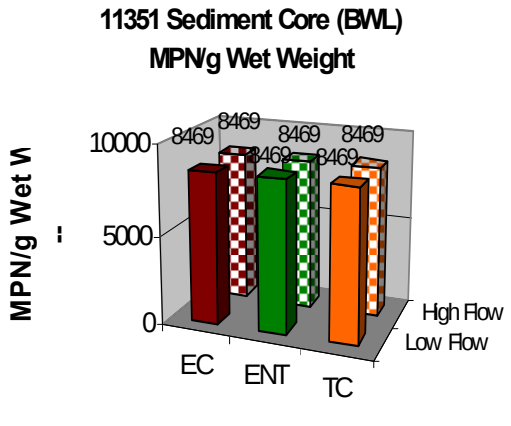


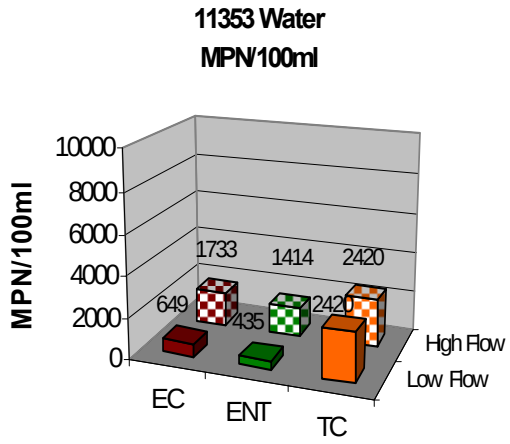
Low Flow (202 cfs)

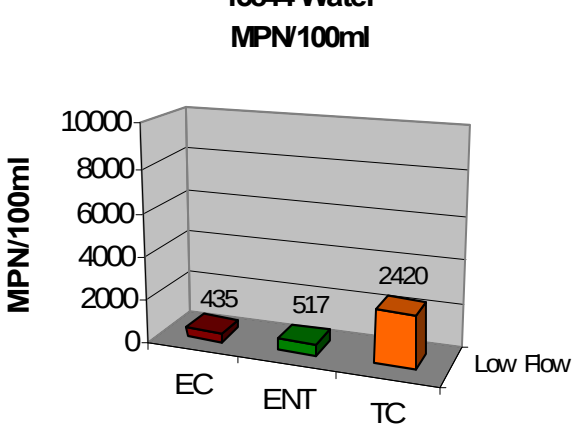
Buffalo Bayou (sediment cores- top horizons)

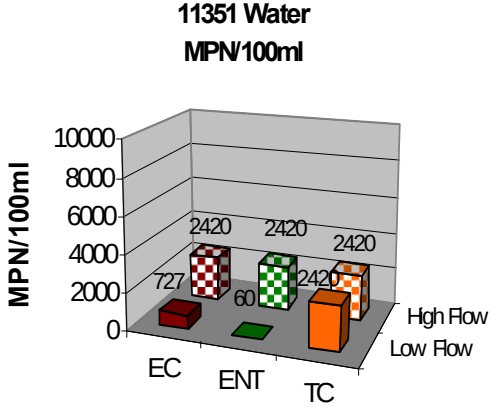










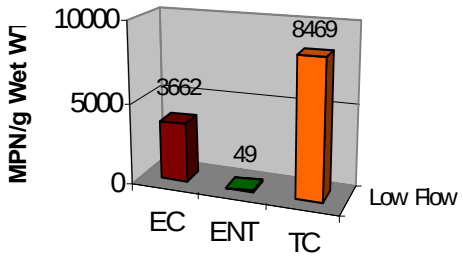


BB at 610

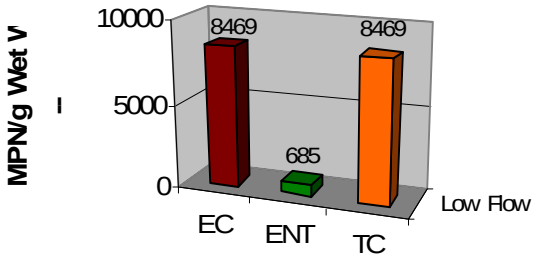
BB at Shepherd

Buffalo Bayou (sediment cores- top horizons)

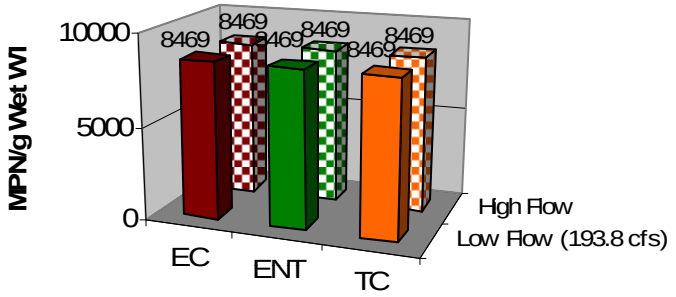
16675 Sediment Core (WL)
MPN/g Wet Weight



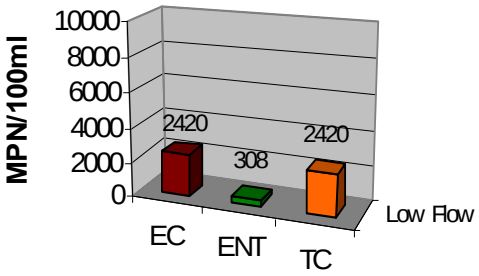
15843 Sediment Core (BWL)
MPN/g Wet Weight



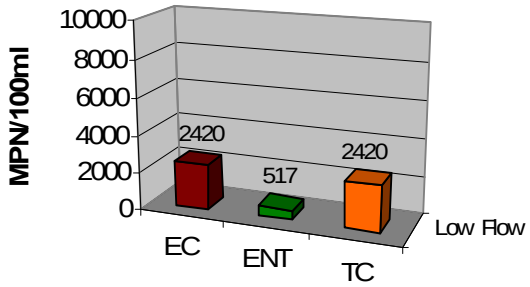
11347 Sediment Core (WL)
MPN/g Wet Weight



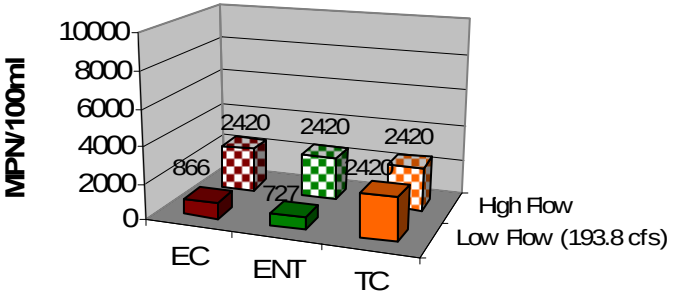
16675 Water
MPN/100ml



15843 Water
MPN/100ml

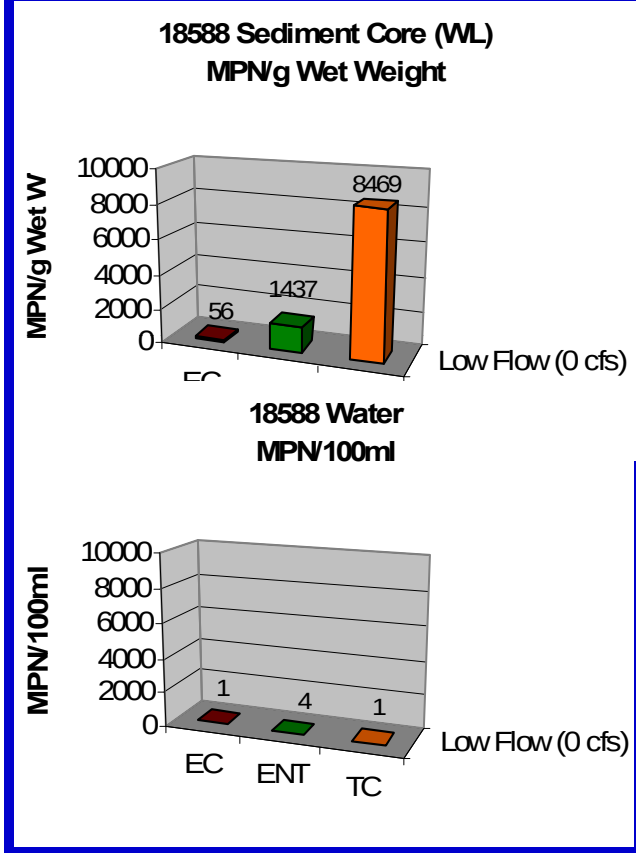


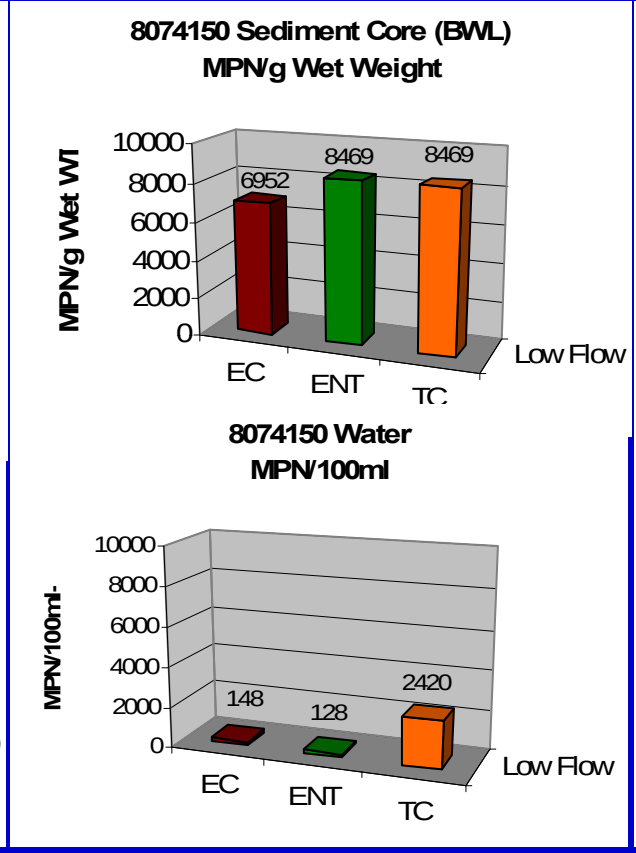
11347 Water
MPN/100ml

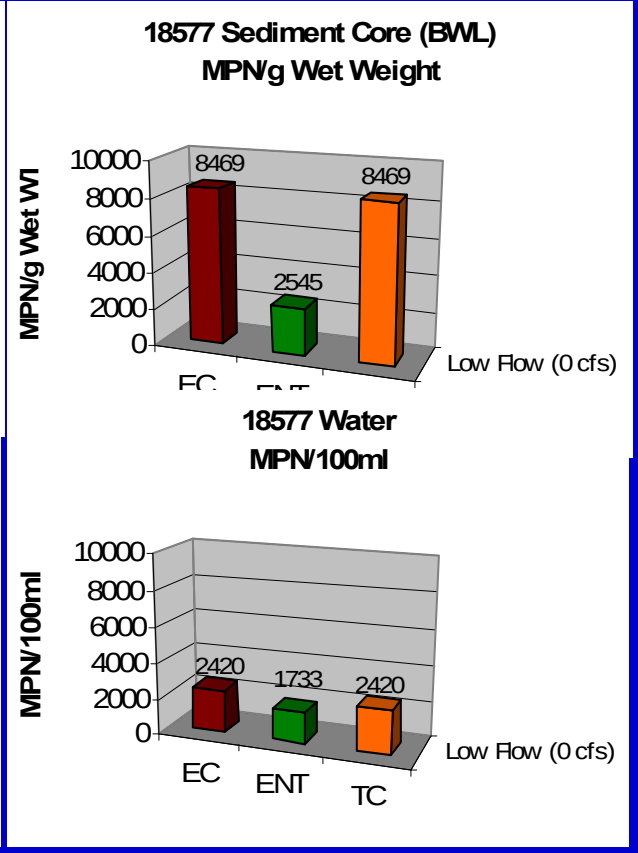


BB at Main St.

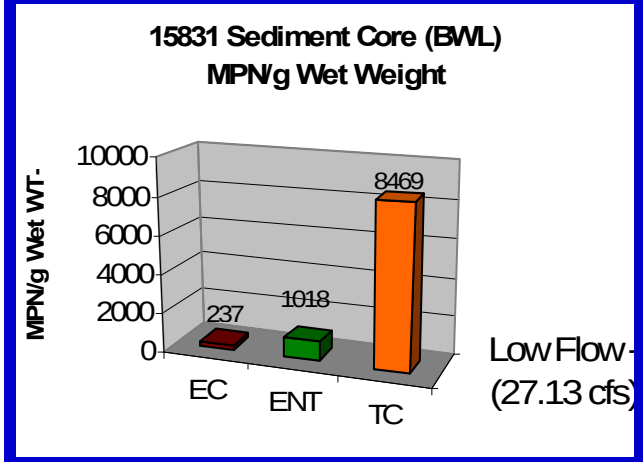
White Oak Bayou (sediment cores- top horizons)

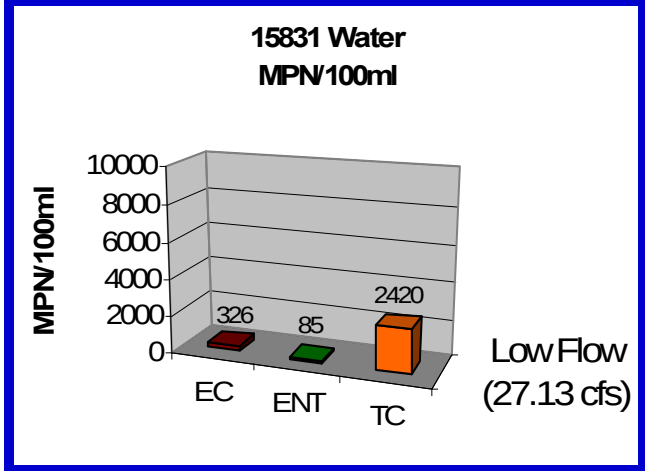


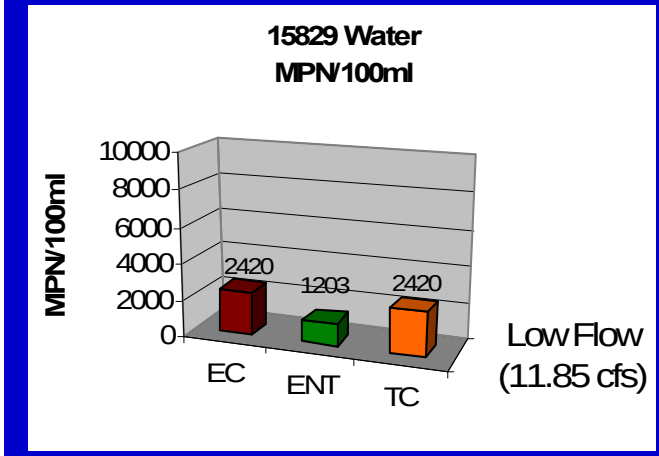


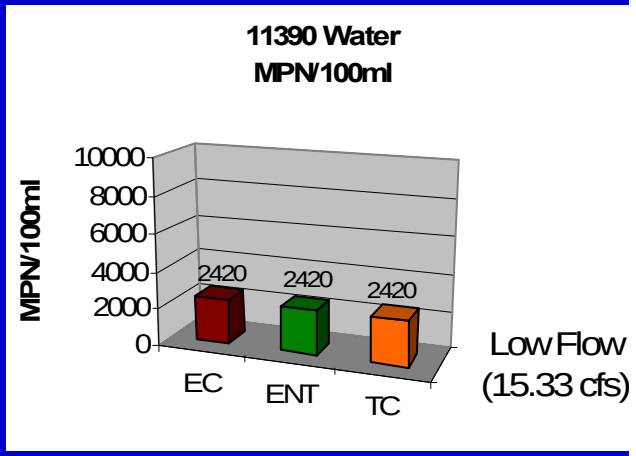


White Oak Bayou (sediment cores- top horizons)



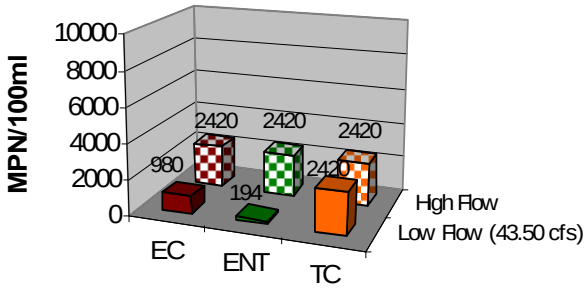




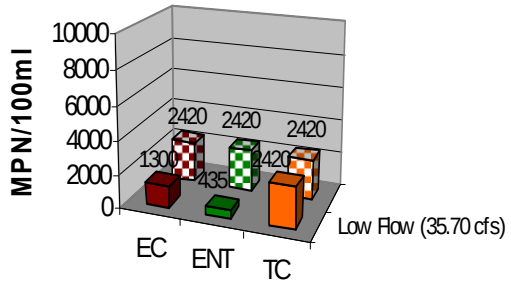


White Oak Bayou (sediment cores- top horizons)

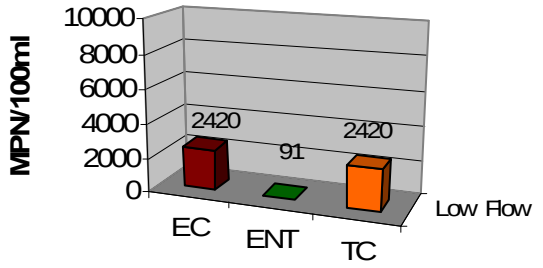
15827 Water
MPN/100ml



11387 Water
MPN/100ml

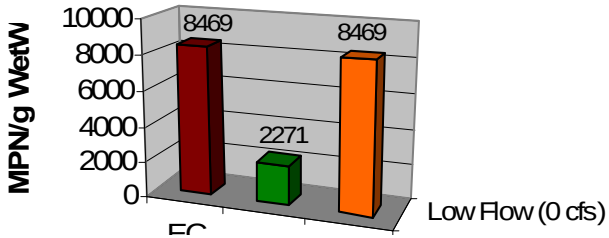


15825 Water
MPN/100ml

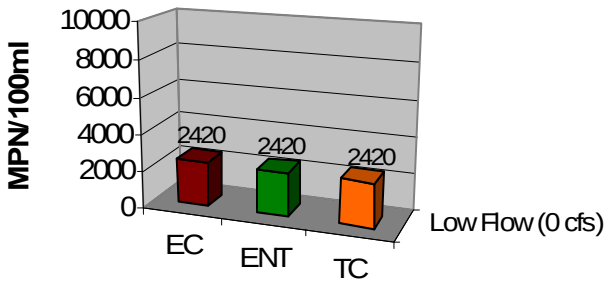


Little White Oak Bayou (sediment cores- top horizons)

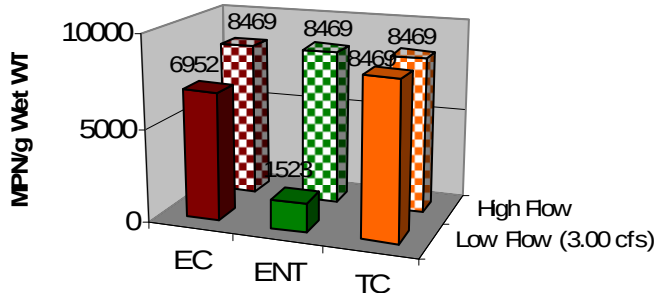
11148 Sediment Core (AWL)
MPN/g Wet Weight



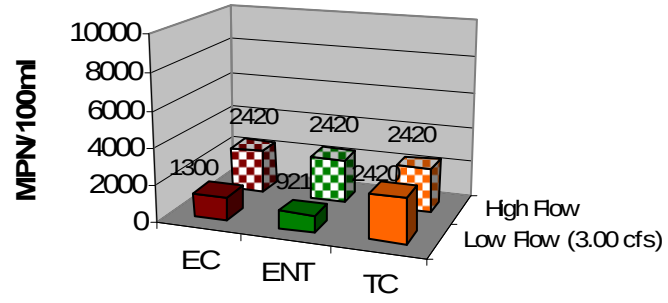
11148 Water
MPN/100ml



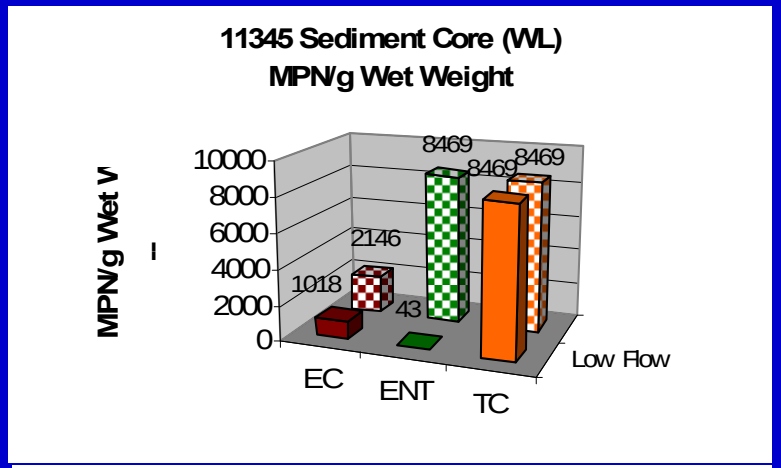
16648 Sediment Core (WL)
MPN/g Wet Weight

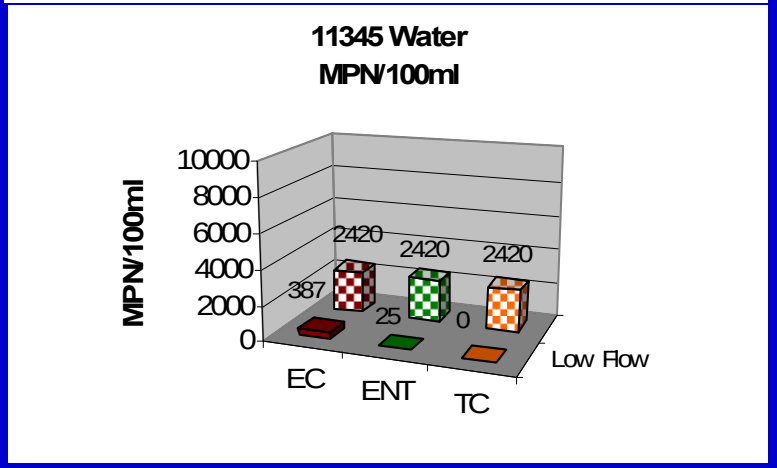


16648 Water
MPN/100ml

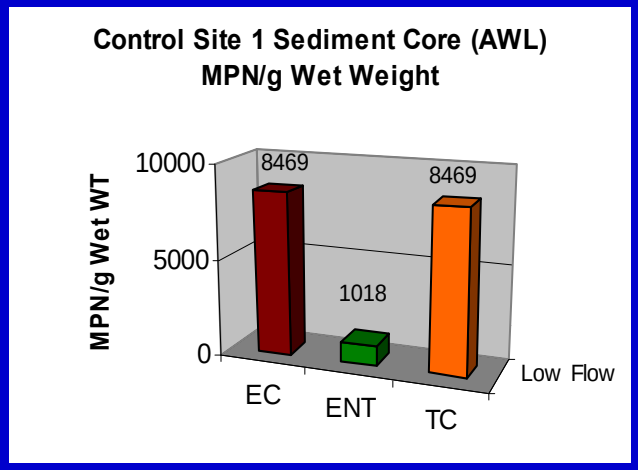


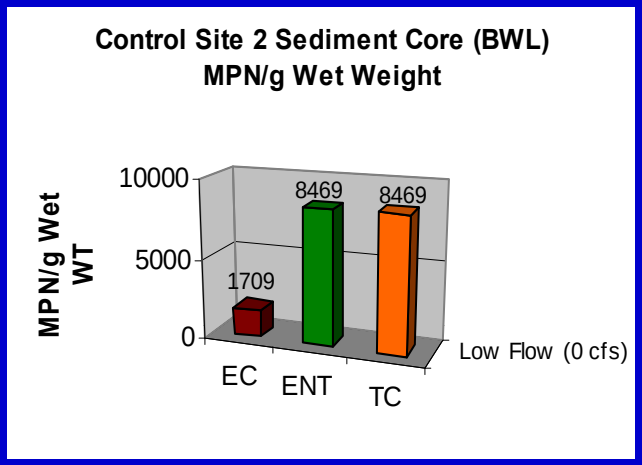
Buffalo Bayou below confluence with White Oak Bayou

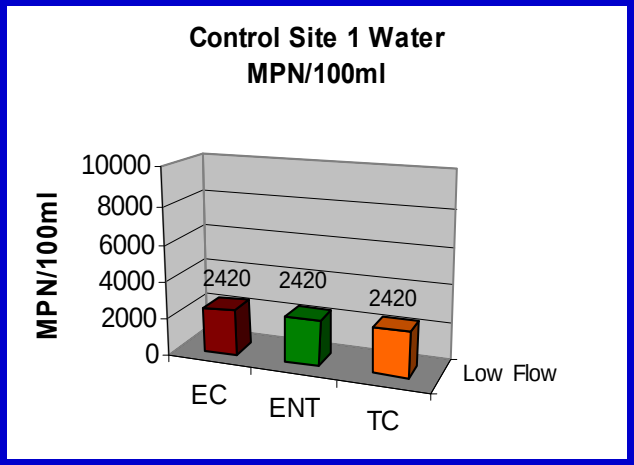


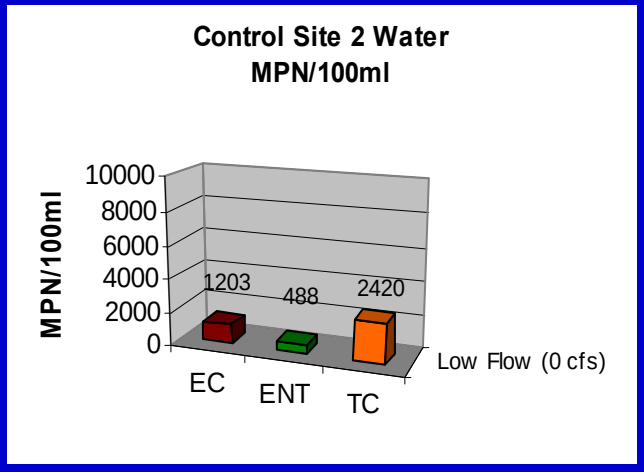


Control Sites- MPN values in sediment and water

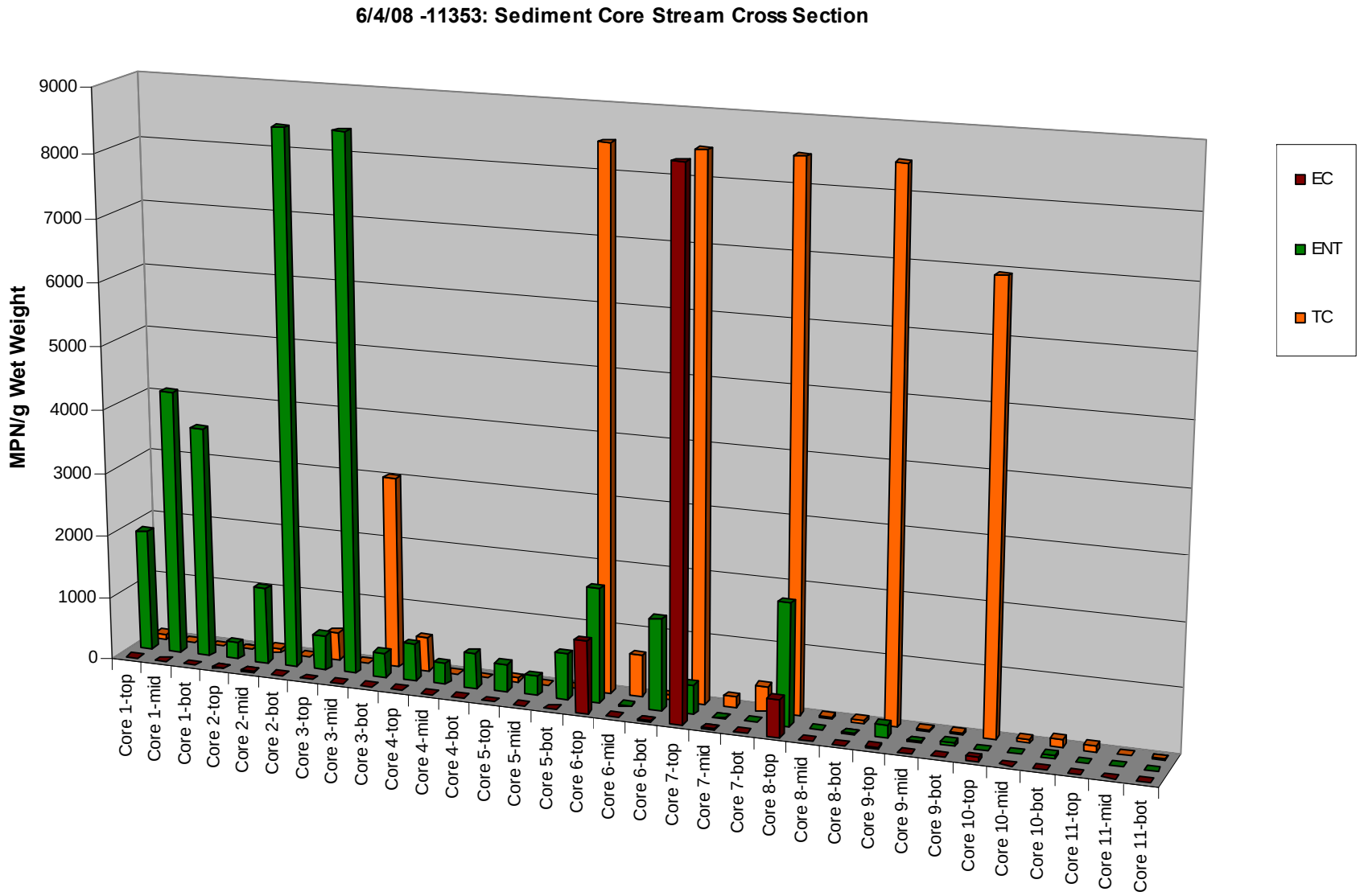






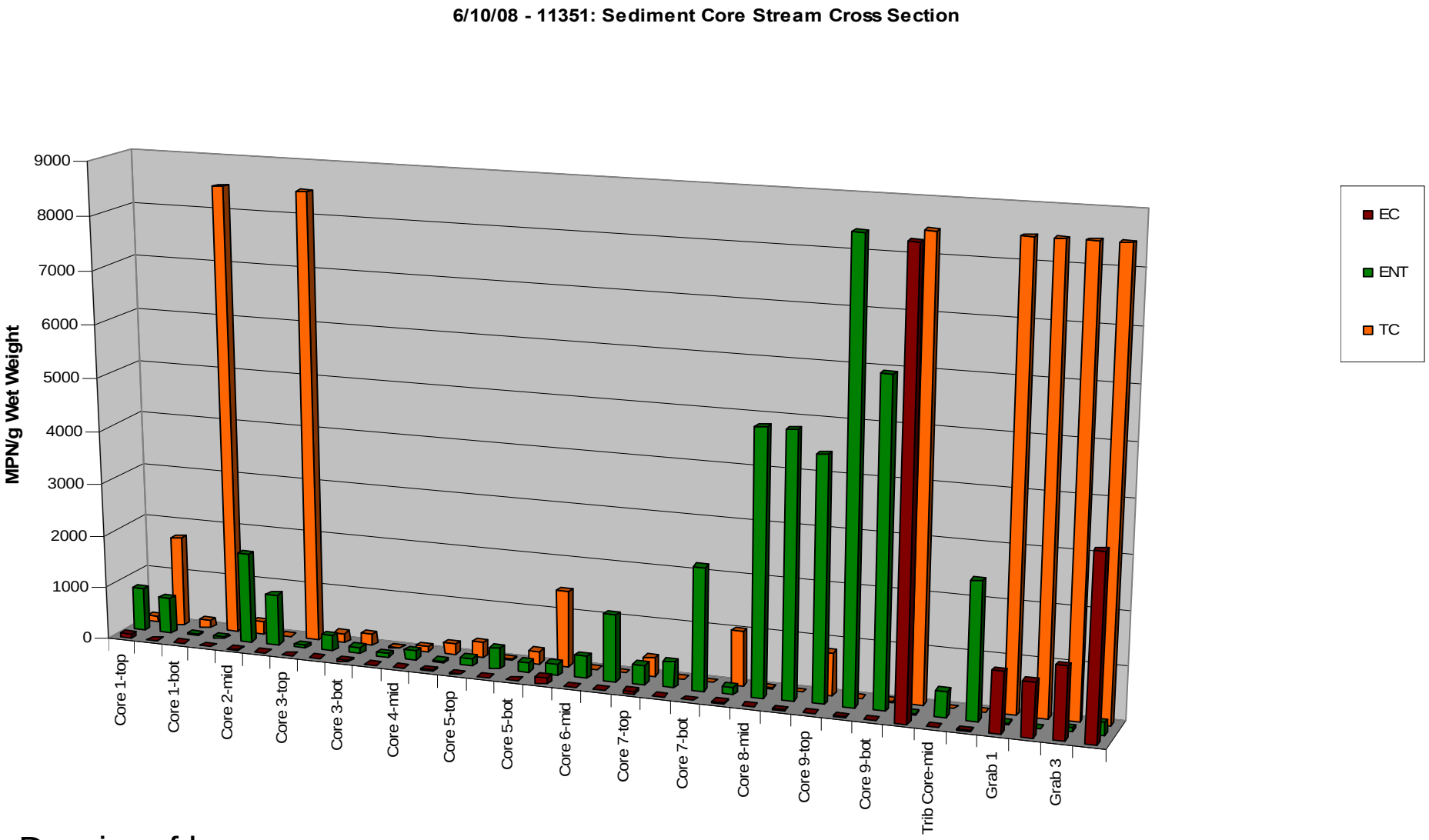


Station 11353 Buffalo Bayou at 610- Cross section analysis of sediments



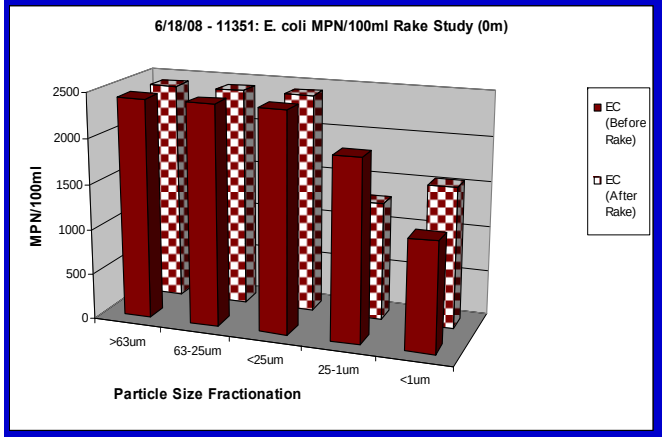
Dry rim of bayou ⟶ Midway in bayou

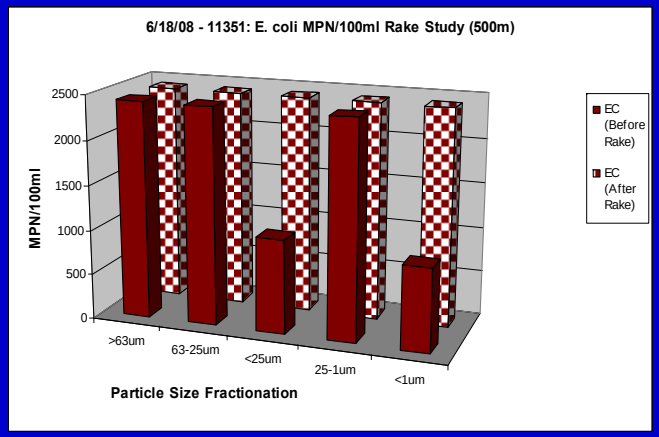
Station 11351 Buffalo Bayou at Shepherd- Cross section analysis of sediments

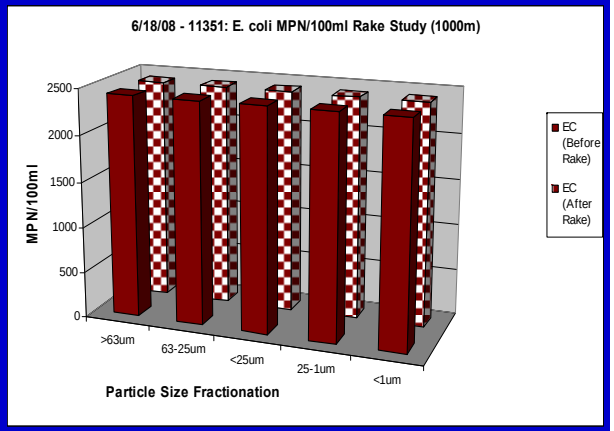


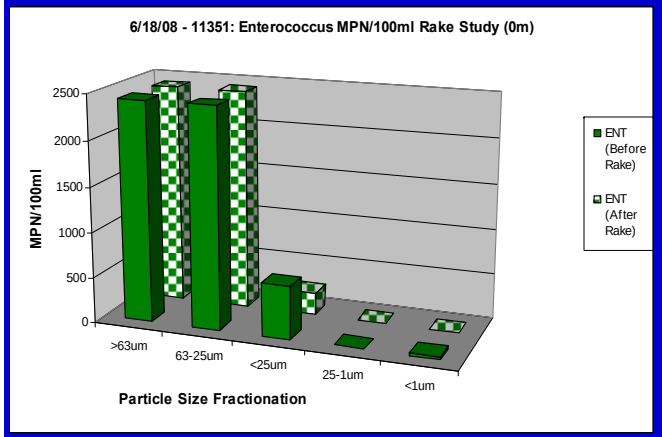
Dry rim of bayou → Midway in bayou

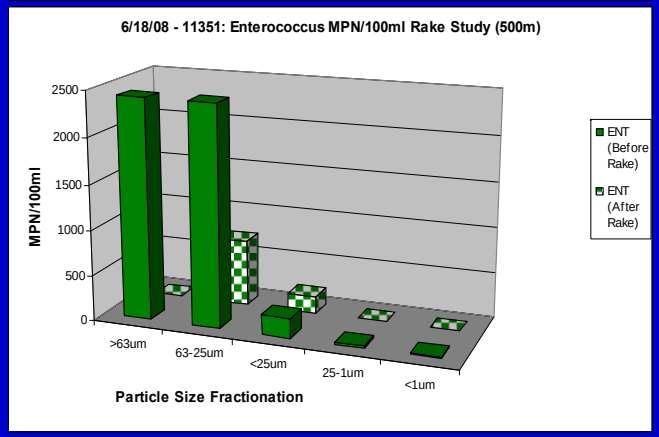
‘Rake Study’ Disturbed sediments at 11351 particle size fractionation & transport analysis

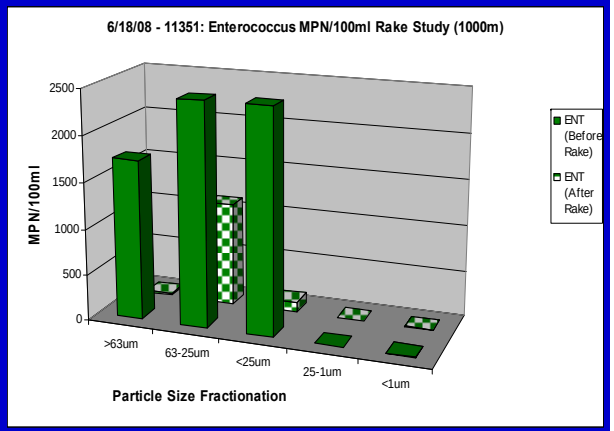


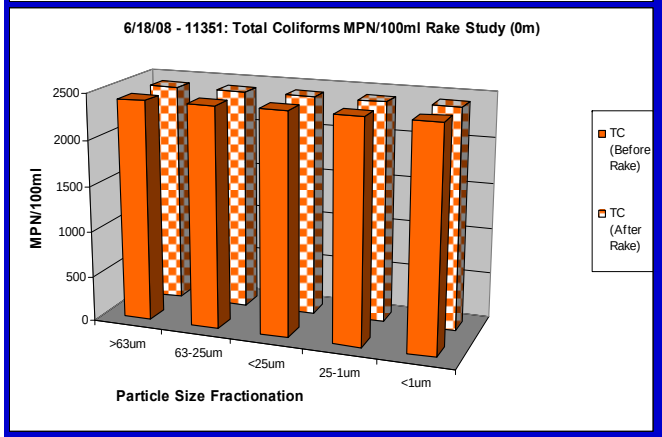


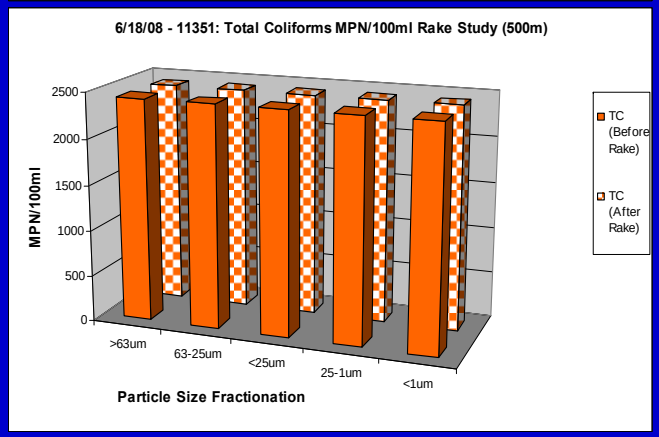


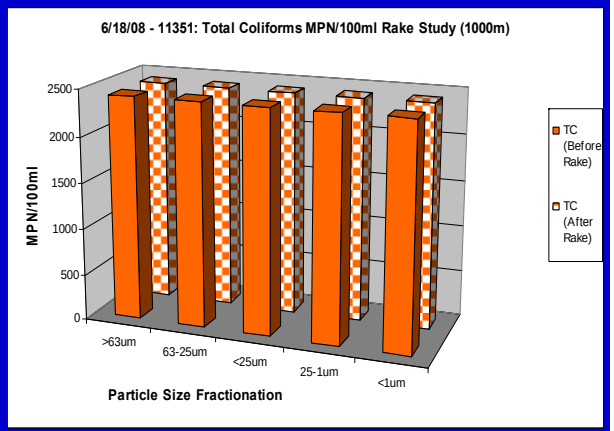






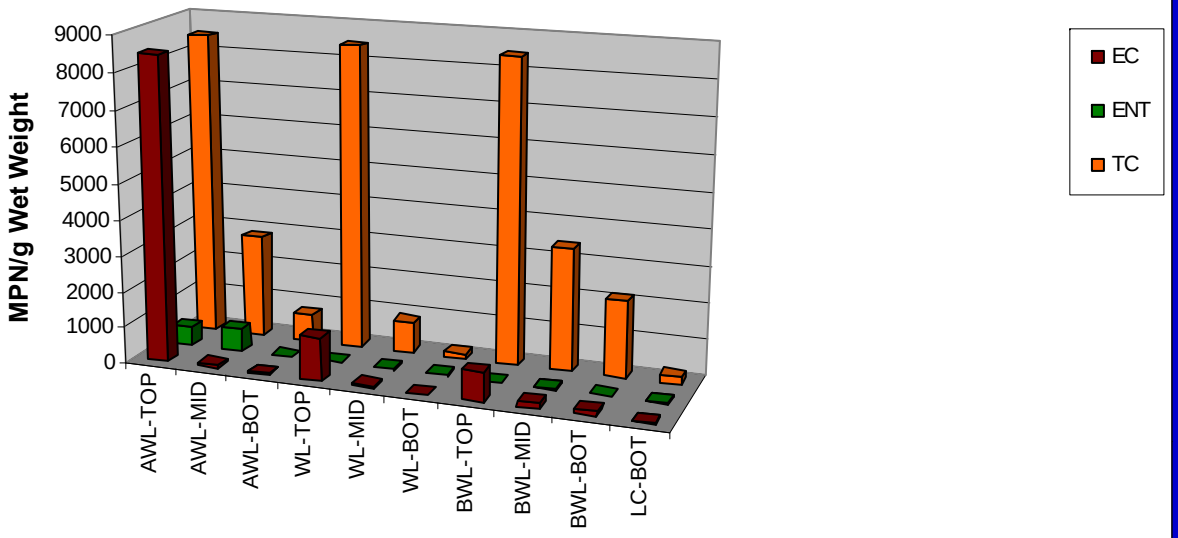




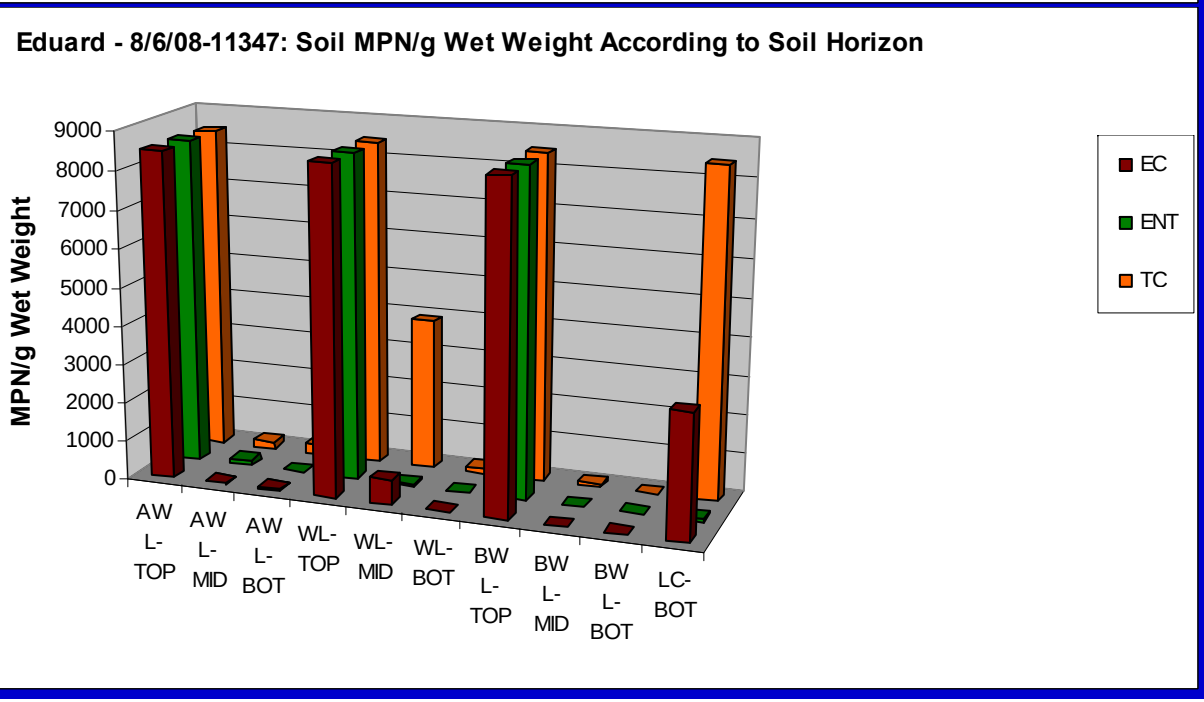


Sediment analysis after Hurricane Eduard

BB at 610

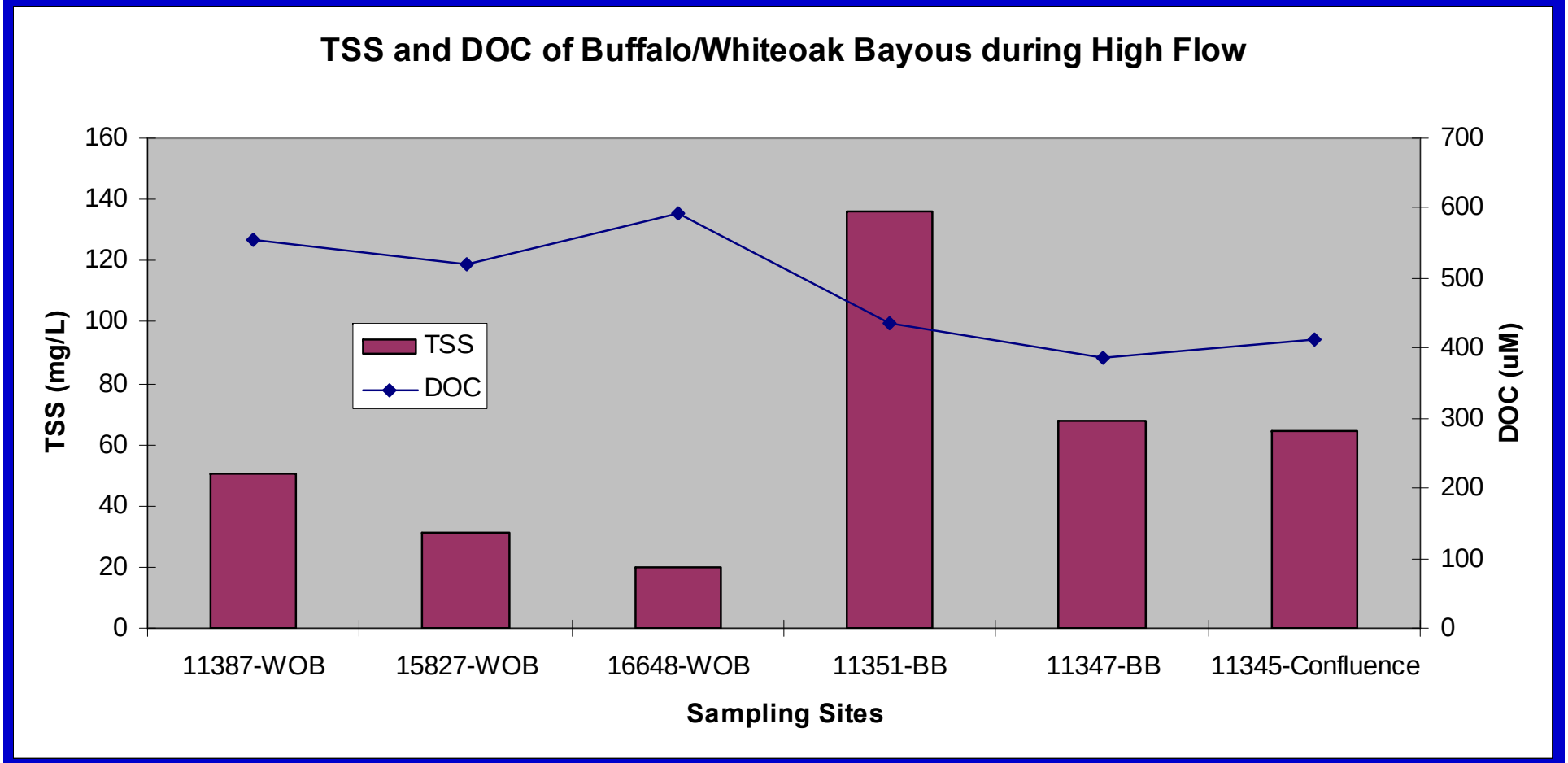


BB at Main St

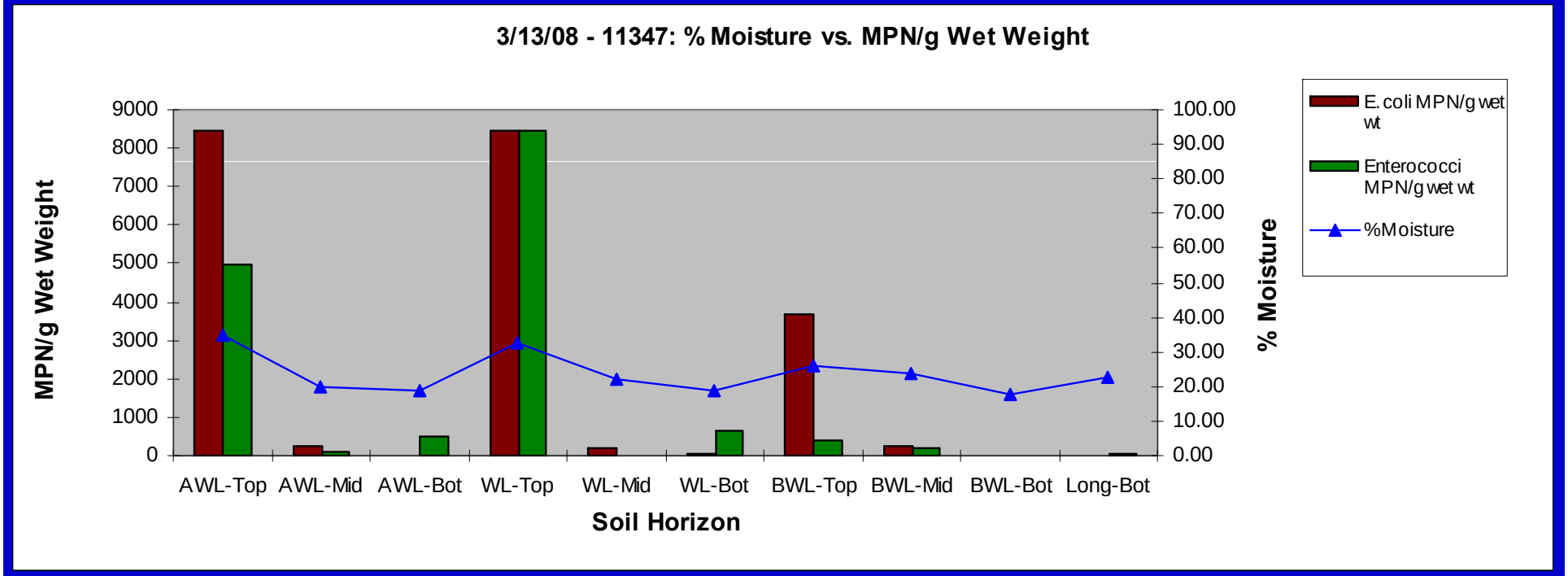


Total Suspended Solids (63-25 μm) and Dissolved Organic Carbon

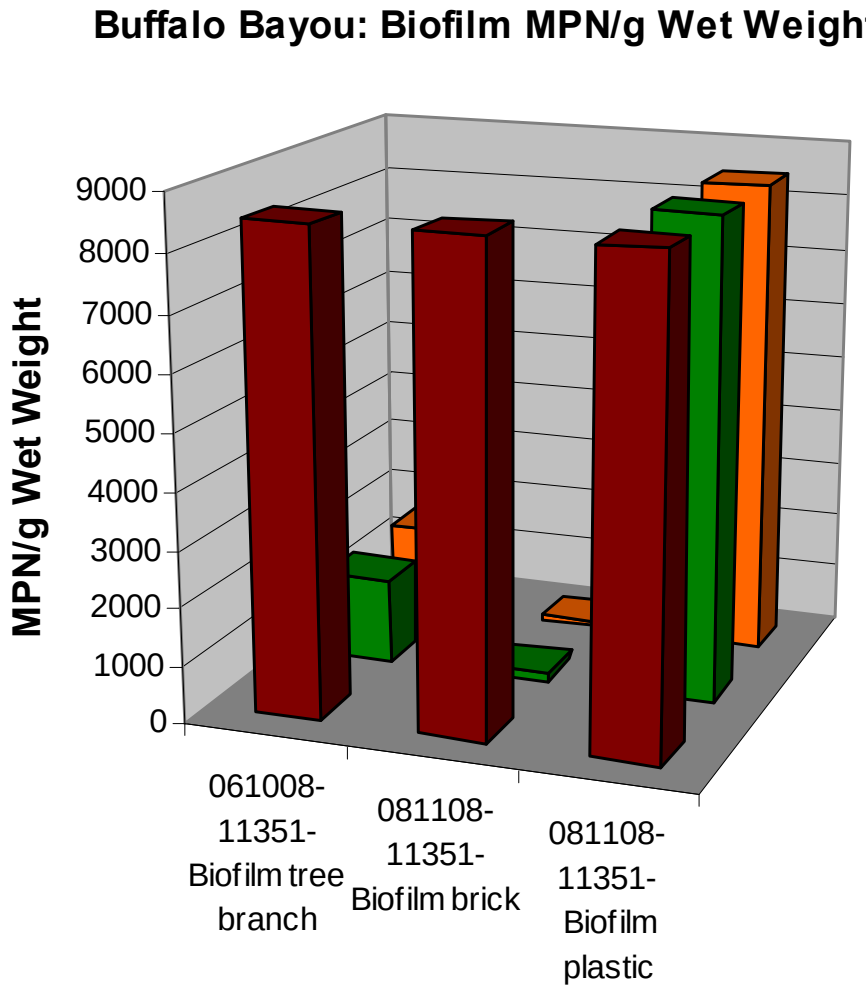
TSS higher in Buffalo Bayou- carbon and stable isotope analysis of particle size Fractionation in progress.



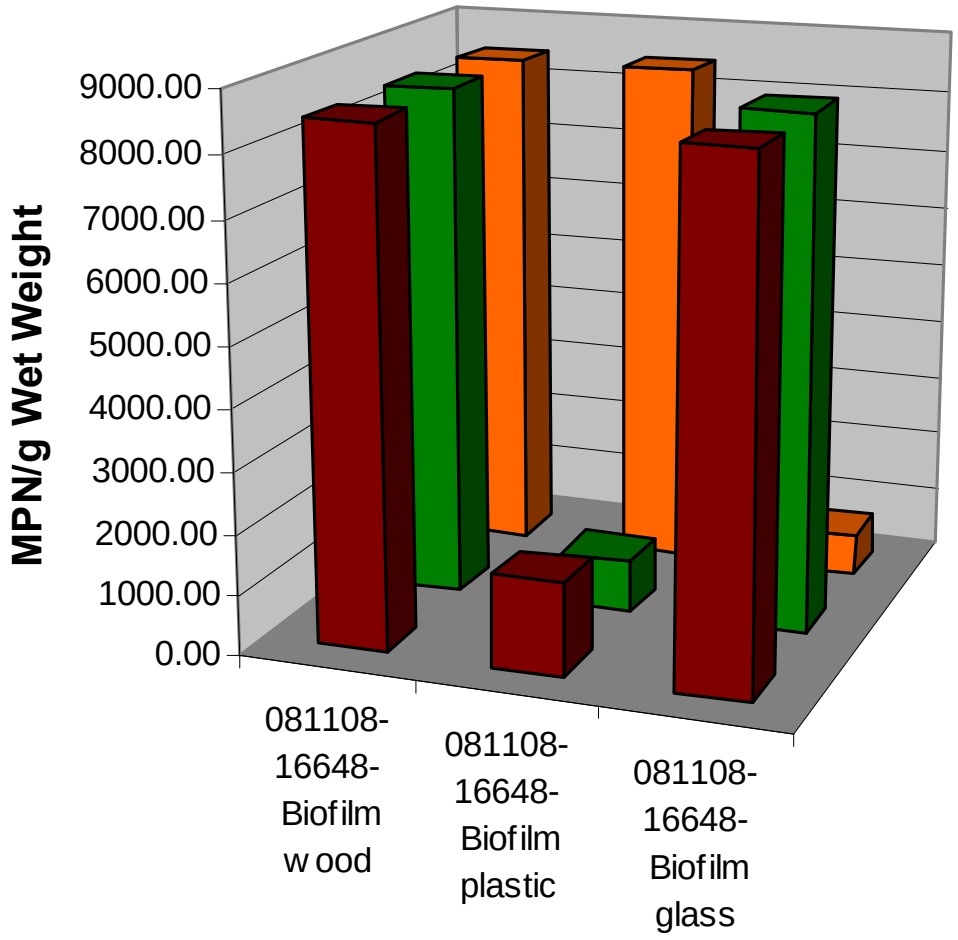
Percent sediment moisture vs MPN/g Wet Weight



Analysis of Biofilms

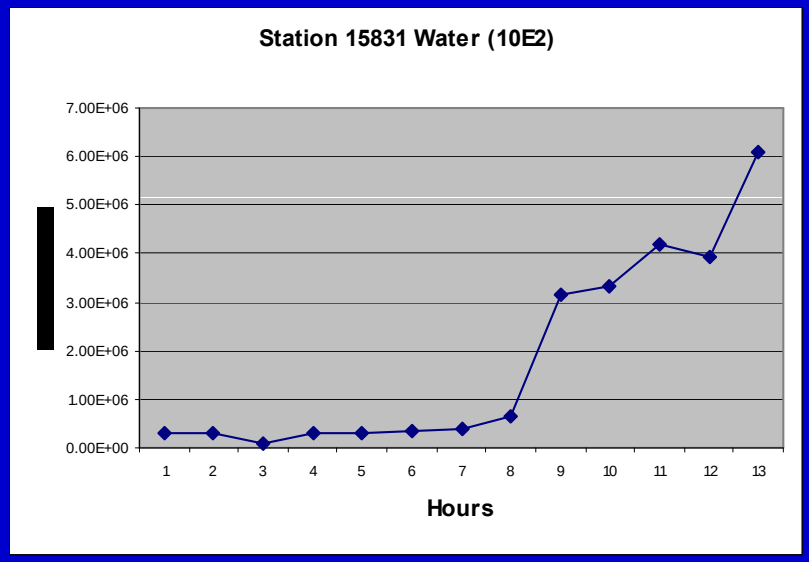


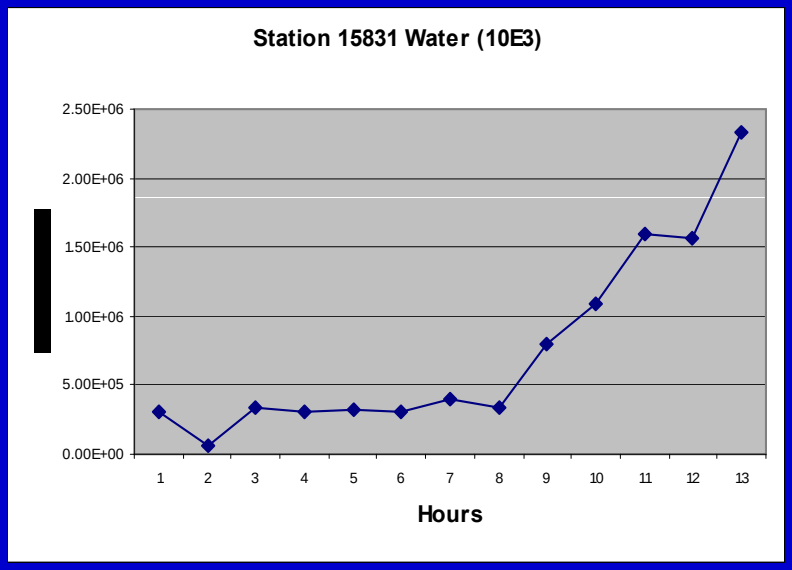
White Oak Bayou: Biofilm MPN/g Wet Weight



E. Coli Incubation Experiments in bayou water

Very preliminary – need to be repeated and confirmed with FISH-DNA probing anlaysis





Sediment incubation studies in progress using gamma irradiated samples from 6 sites.

FY2009 Focus on storm drains, biofilms, and incubation studies