

# Microbial Source Tracking

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NJDEP Water Monitoring and  
Standards

Bureau of Marine Water  
Monitoring



# OVERVIEW

## *New Jersey's Application of Microbial Source Tracking Techniques*

- New Jersey's tiered approach to microbial source tracking studies
- Laboratory methods used to distinguish sources of fecal contamination
- MST case study at impaired shellfish growing areas
- MST case study at impaired recreational waters

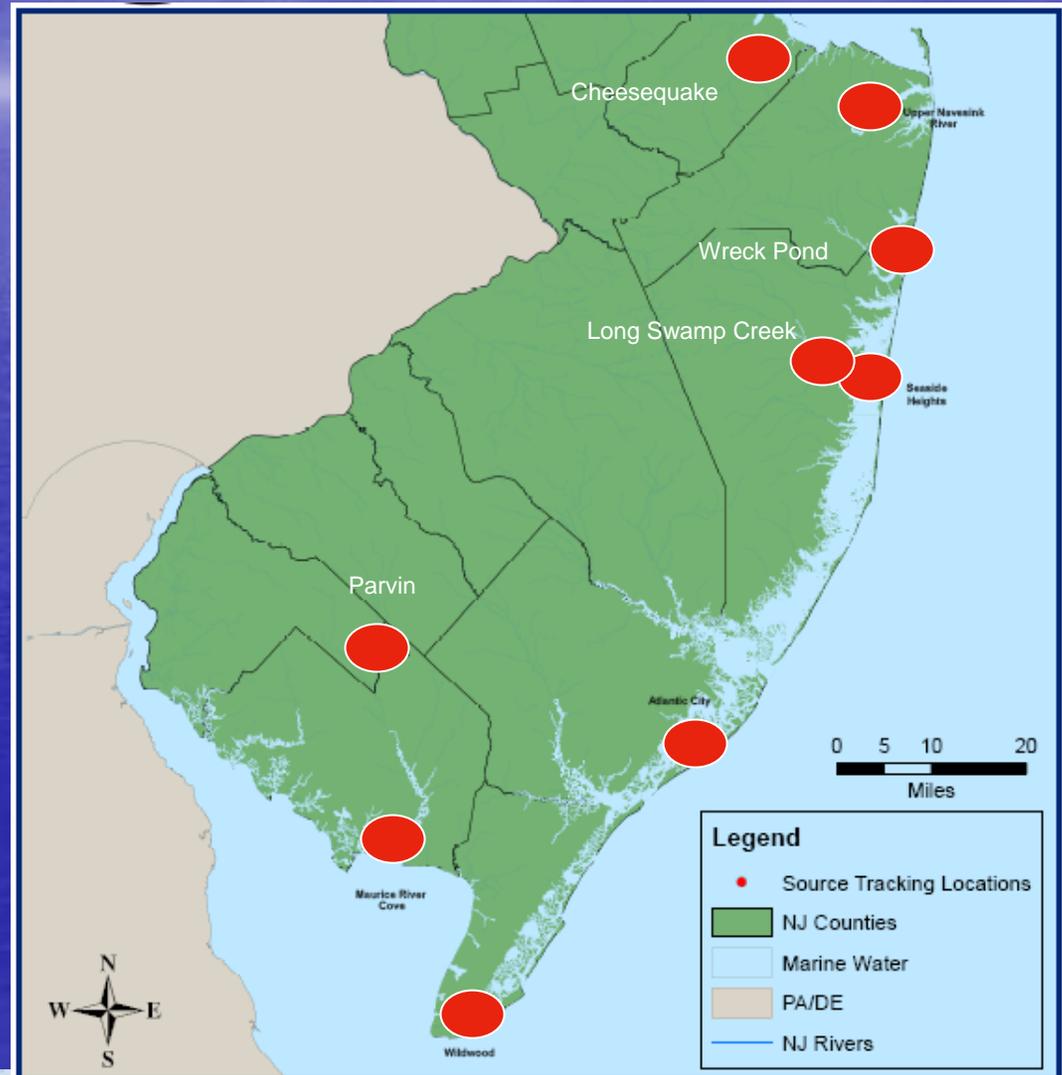


# Nonpoint Source Microbial Source Tracking

A high percentage of NJ's water quality impairments are related to pathogens.

Microbial source tracking efforts are becoming increasingly common in NJ surface waters.

There are currently 9 different source tracking efforts underway in NJ.



NJDEP'S  
MICROBIAL SOURCE  
TRACKING STRATEGY

**Scientific  
Weight  
Of Evidence**

**Utilize MST tools  
including;  
coliphage, ARA, and  
Optical Brighteners**

**Perform intensive monitoring  
under APC regime using conventional  
indicators: FC, Entero, *E.coli*. Sample  
at dry, first flush, hour intervals, next day**

**Perform shoreline survey of the watershed  
Utilize GIS and land use coverage  
Consider sampling logistics**

**Identify impaired areas (i.e. beach closures,  
closed shellfish areas) – based  
on long-term monitoring data analysis**

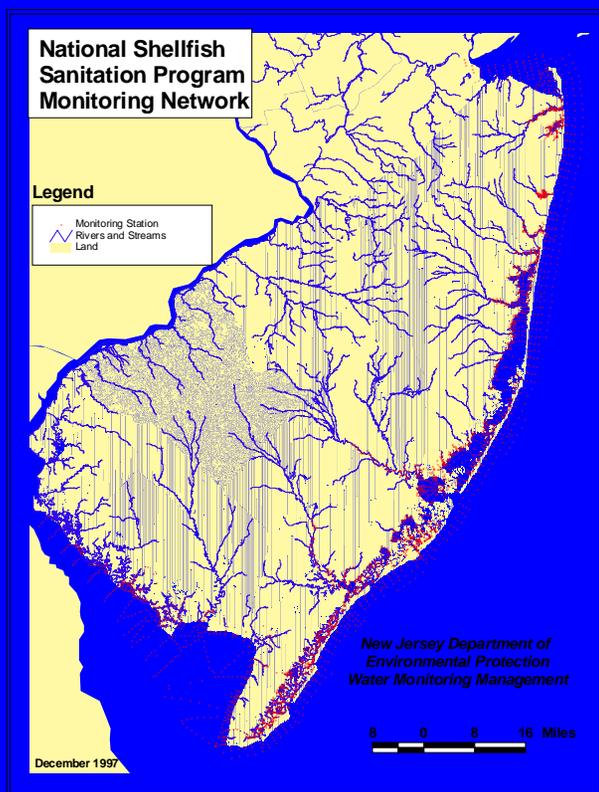


# ADVANCED MICROBIOLOGY LABORATORY AT LEEDS POINT



# NSSP Monitoring Network

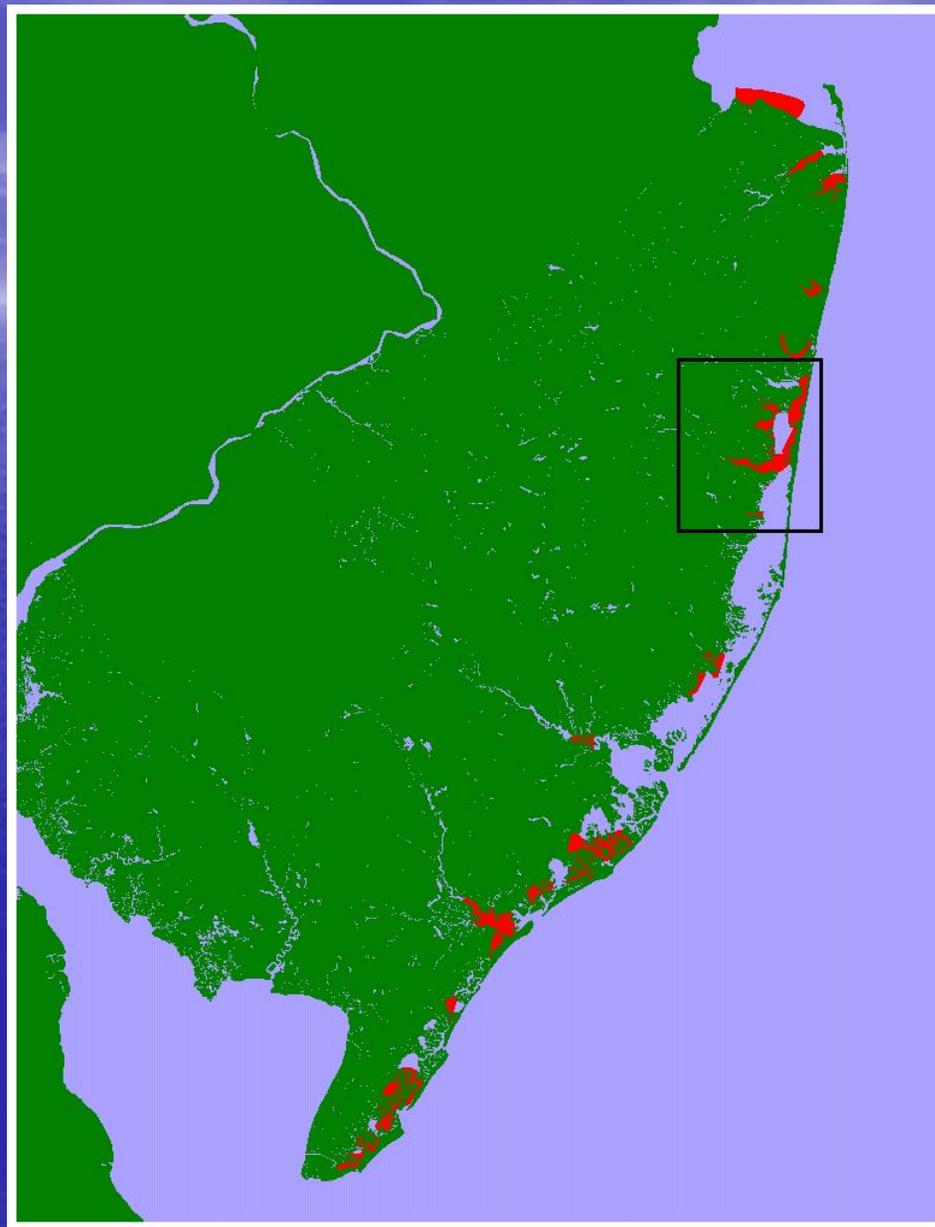
- 2500 active monitoring locations
- Sampling 5-12 times/year
- Total and fecal coliform bacteria
- Approximately 15,000 samples/year



# Stormwater Effects on the Coast

Areas shaded red - coastal  
water quality degraded  
following storm events.

Evaluation based on  
10 years of data - generated  
by Leeds Point staff.



# Questions that need to be answered when designing a MST Study

- Is the Impairment Adequately Defined?
  - beach closures
  - shellfish growing waters
- Has An Adequate Sanitary Survey Been Conducted?
- How Many Sources Were Identified In the Sanitary Survey?



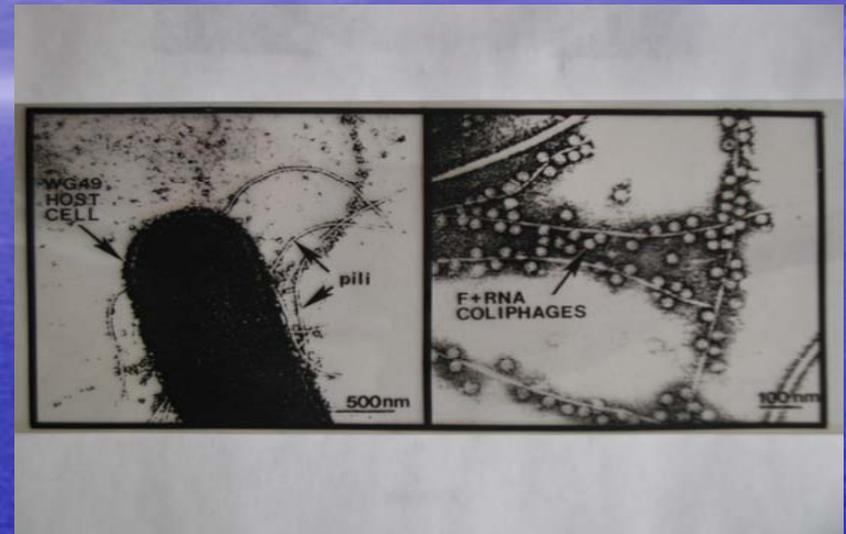
# Questions - continued

- Levels of Discrimination
  - Humans vs. all other sources
  - Species specific (humans, cows, geese, dogs, etc.)
  - Host Group (humans vs. livestock vs. wildlife)
  - Individual hosts ( cows from a certain farm vs. other farms)
  - *Accuracy decreases as level of discrimination increases*



# F+ RNA Coliphages

- Viruses that infect bacterial cells
- Similar in size, shape and morphology to HEV including; HAV and Norwalk therefore: good viral pathogen indicator
- More resistant to chlorination than the conventional indicators therefore: good wastewater effluent indicator



# Coliphage - NJDEP Findings

- Monitoring at known fecal contaminated sites
  - point human - wastewater discharge outfall
  - point animal - wildlife refuge discharge
  - non-point human - malfunctioning septic tank discharge
  - non-point animal - rural creek w/animal population
- Findings:
  - Verified the presence of coliphage at these sites and validated the procedure
  - Genotyping) of the phages provides a promising system for distinguishing human and animal fecal contamination



# Genotyping of coliphages

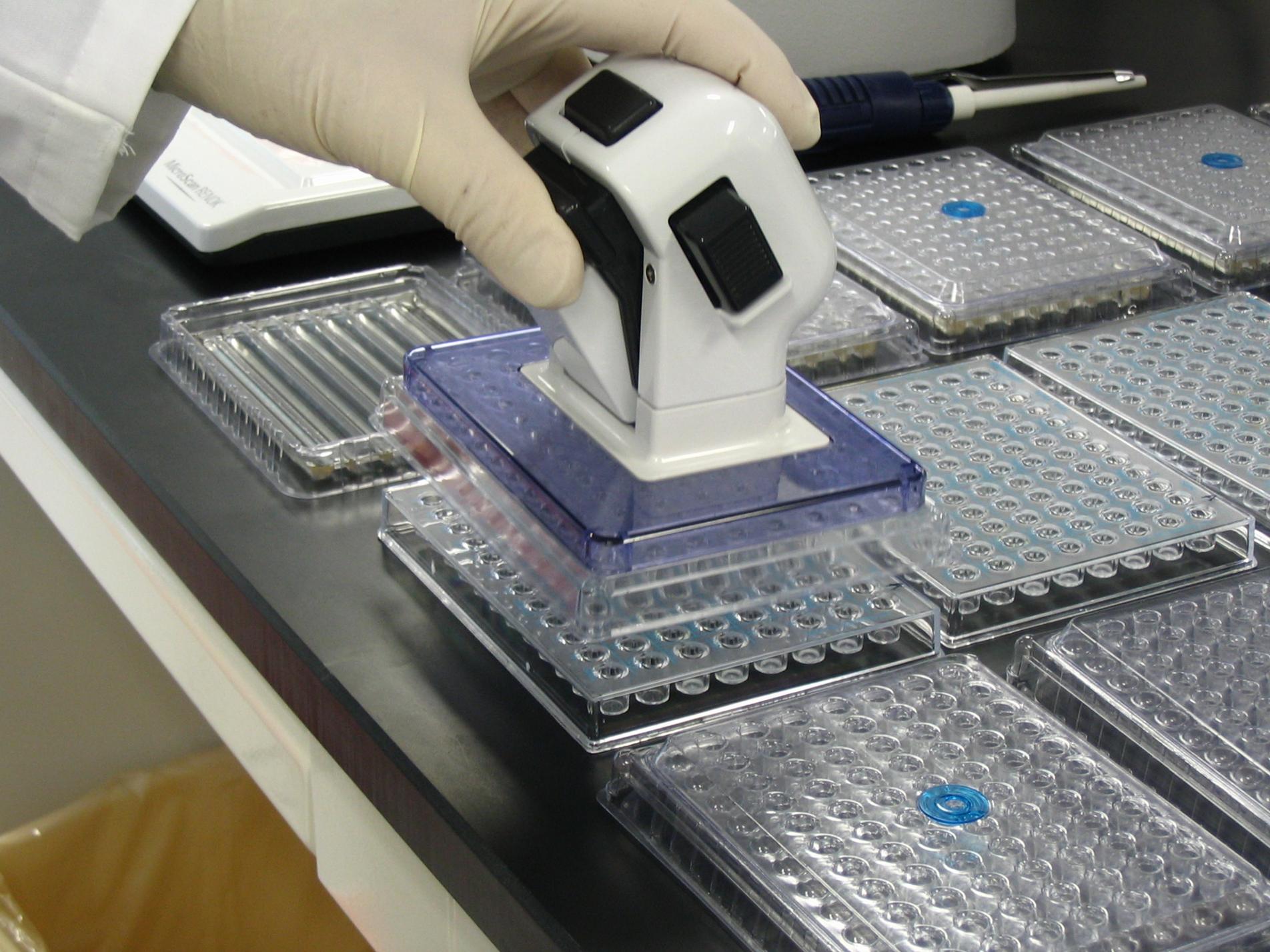
- Group I - Animal
- Group II - Human
- Group III - Human
- Group IV - Animal



# ARA – Antibiotic Resistance Analysis

- **ARA - Antibiotic Resistance Analysis**
  - identifies *E. coli* that are resistant to antibiotics used to treat bacterial infections in humans and domesticated animals.
  - Procedure uses 96 well microplate containing 26 antibiotics typically administered to humans and domesticated animals





# ARA Results

| Antibiotic           | Waterfowl |     | Cattle |     | WWTP |
|----------------------|-----------|-----|--------|-----|------|
| Azithromycin         | Low       | Med | Med    | Med | Med  |
| Erythromycin         | Med       | Med | Med    | Med | Med  |
| Penicillin G or V    | Med       | Med | Med    | Med | Med  |
| Oxytetracycline      | Med       | Med | High   | Med | Med  |
| Tetracycline         | Med       | Med | Med    | Med | Med  |
| Amoxicillin          | Med       | Med | Med    | Med | High |
| Ceftriaxone          | Low       | Med | Med    | Med | Low  |
| Ampicillin           | Med       | Med | Med    | Med | Med  |
| Resistance Intensity | Low       |     | Med    |     | High |





### WILDLIFE

NOAA Panel Date 3-21-06 Coll. Time 11:00 Sample # REFUGE ID # EAST GATE

|           |           |           |           |           |    |            |           |          |           |           |             |
|-----------|-----------|-----------|-----------|-----------|----|------------|-----------|----------|-----------|-----------|-------------|
| <u>G</u>  | 2         | 4         | 8         | 16<br>Imp | 1  | 2          | 4         | 8<br>Mox | 1         | 2         | 4<br>Cp     |
| 1         | 2         | 4         | 8         | 16<br>Mer | 1  | 2          | 4         | 8<br>Op  | 250<br>Sz | 500       | 2           |
| 2         | <u>4</u>  | 16        |           |           |    |            | 8         | 16       | 32<br>C   | <u> </u>  | 4           |
| 4<br>Azi  | <u>8</u>  | 32        | 4         | 8         | 16 | 32<br>Otet | 4         | 8        | 16        | 32<br>NA  | 8<br>T      |
| 8         | <u>16</u> | 64        | 4         | 8         | 16 | 32<br>Te   | 8         | 16       | 32        | 64<br>Ak  | 2/38        |
| 16        | 32<br>E   | 128<br>St | 2         | 4         | 8  | 16<br>Gm   | 16        | 32       | 64        | 128<br>Fd | 4/76<br>T/S |
| 32<br>Apr | 8         | 16        | 32<br>Cfx | 8         | 16 | 32         | 64<br>Cax | <u>4</u> | 8         | 16        | 32<br>Cf    |
| <u>8</u>  | <u>16</u> | 32        | 64<br>P   | 4         | 8  | 16         | 32<br>Amx | 4        | 8         | 16        | 32<br>Am    |

| Vol.      | MODIFIED mTec<br>Colony CFU's/100mLs<br>Count |           |
|-----------|---|-----------|
| <u>30</u> | <u>12</u>                                     | <u>40</u> |

| Phage    | PFU's/100mLs |
|----------|--------------|
| <u>—</u> | <u>&lt;1</u> |





### DOMESTICATED ANIMAL

NOAA Panel Date 3-2-04 Coll. Time \_\_\_\_\_ Sample # \_\_\_\_\_ ID # ALDWAY

|     |    |     |     |     |    |      |     |     |     |     |      |
|-----|----|-----|-----|-----|----|------|-----|-----|-----|-----|------|
| G   | 2  | 4   | 8   | 16  | 1  | 2    | 4   | 8   | 1   | 2   | 4    |
|     |    |     |     | Imp |    |      |     | Mox |     |     | Cp   |
| 1   | 2  | 4   | 8   | 16  | 1  | 2    | 4   | 8   | 250 | 500 | 2    |
|     |    |     |     | Mer |    |      |     | Op  |     |     |      |
| 2   | 4  | 16  |     |     |    |      | 8   | 16  | 32  |     | 4    |
|     |    |     |     |     |    |      |     |     | C   |     |      |
| 4   | 8  | 32  | 4   | 8   | 16 | 32   | 4   | 8   | 16  | 32  | 8    |
| Azi |    |     |     |     |    | Otet |     |     |     | NA  | T    |
| 8   | 16 | 64  | 4   | 8   | 16 | 32   | 8   | 16  | 32  | 64  | 2/38 |
|     |    |     |     |     |    | Te   |     |     |     | AK  |      |
| 16  | 32 | 128 | 2   | 4   | 8  | 16   | 16  | 32  | 64  | 128 | 4/76 |
|     |    | St  |     |     |    | Gm   |     |     |     | Fd  | T/S  |
| 32  | 8  | 16  | 32  | 8   | 16 | 32   | 64  | 4   | 8   | 16  | 32   |
| Apr |    |     | Cfx |     |    |      | Cax |     |     |     | Cf   |
| 8   | 16 | 32  | 64  | 4   | 8  | 16   | 32  | 4   | 8   | 16  | 32   |
|     |    |     | P   |     |    |      | Amx |     |     |     | Am   |

| Vol.  | MODIFIED mTee Colony Count | CFU's/100mLs |
|-------|----------------------------|--------------|
| _____ | _____                      | _____        |

| Phage | PFU's/100mLs |
|-------|--------------|
| _____ | _____        |





## HUMAN

NOAA Panel    Date 3-27-04    Coll. Time \_\_\_\_\_    Sample # ACUA    ID # EFFLUENT

|     |    |     |     |     |    |      |     |     |     |     |      |
|-----|----|-----|-----|-----|----|------|-----|-----|-----|-----|------|
| G   | 2  | 4   | 8   | 16  | 1  | 2    | 4   | 8   | 1   | 2   | 4    |
|     |    |     |     | Imp |    |      |     | Mox |     |     | Cp   |
| 1   | 2  | 4   | 8   | 16  | 1  | 2    | 4   | 8   | 250 | 500 | 2    |
|     |    |     |     | Mer |    |      |     | Opf |     |     |      |
| 2   | 4  | 16  |     |     |    |      | 8   | 16  | 32  |     | 4    |
|     |    |     |     |     |    |      |     | C   |     |     |      |
| 4   | 8  | 32  | 4   | 8   | 16 | 32   | 4   | 8   | 16  | 32  | 8    |
| Azi |    |     |     |     |    | Otet |     |     |     | NA  | T    |
| 8   | 16 | 64  | 4   | 8   | 16 | 32   | 8   | 16  | 32  | 64  | 2/38 |
|     |    |     |     |     |    | Te   |     |     |     | Ak  |      |
| 16  | 32 | 128 | 2   | 4   | 8  | 16   | 16  | 32  | 64  | 128 | 4/76 |
|     |    |     |     |     |    | Gm   |     |     |     | Fd  | T/S  |
| 32  | 8  | 16  | 32  | 8   | 16 | 32   | 64  | 4   | 8   | 16  | 32   |
| Apr |    |     | Cfx |     |    |      | Cax |     |     |     | CF   |
| 8   | 16 | 32  | 64  | 4   | 8  | 16   | 32  | 4   | 8   | 16  | 32   |
|     |    |     | P   |     |    |      | Amx |     |     |     | Am   |

|            |                      |              |
|------------|----------------------|--------------|
|            | <b>MODIFIED mTec</b> |              |
| Vol.       | Colony Count         | CFU's/100mLs |
| <u>100</u> | <u>17</u>            | <u>17</u>    |

|       |               |
|-------|---------------|
| Phage | PFU's/100mLs  |
| _____ | <u>11,280</u> |



# Optical Brighteners

- Fluorescent Whitening Agents (FWAs) are compounds that can be measured and studied as an indicator of human sources of pollution
- Laundry detergents contain FWAs and are discharged in substantial quantities with household wastewater
- Fluorometric Detection – Turner Fluorometer



# CASE HISTORY-IMPAIRED SHELLFISH GROWING WATERS and Recreational Waters

- Study area in bay waters adjacent to Seaside Park, NJ
- Small cove with impaired shellfish waters and recreational waters
- Area typical of many N.J. barrier island coastal towns – aging infrastructure and numerous stormwater discharges



# Seaside Storm Water Project

## Prior to Rainfall

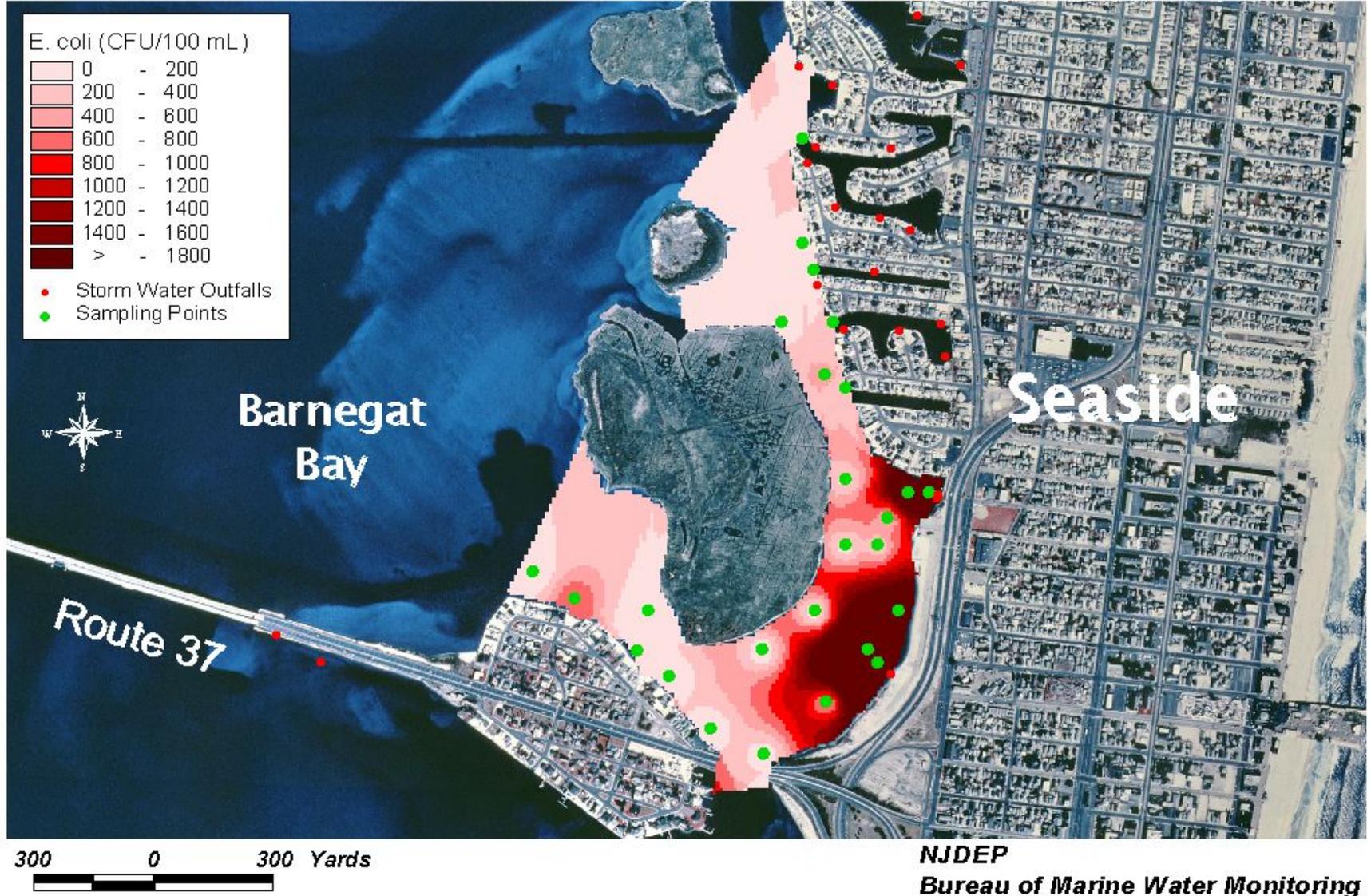


**NJDEP**  
**Bureau of Marine Water Monitoring**



# Seaside Storm Water Project

1 Hour After Storm Event Began

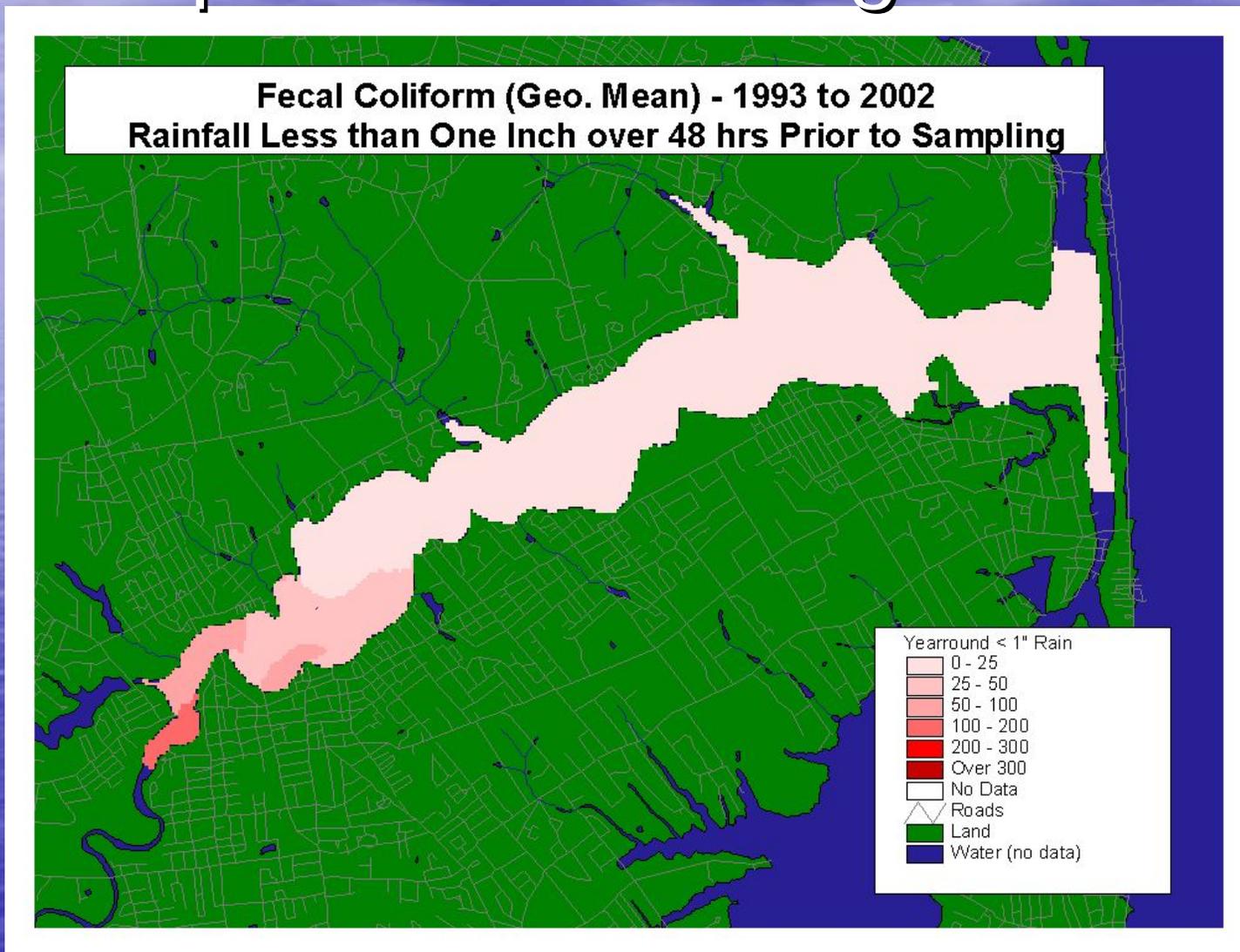


# CASE HISTORY-IMPAIRED SHELLFISH GROWING WATERS

- Navesink River – Monmouth County, N.J.
- Tidal river approx. 8 miles long with numerous small creeks – draining an area of 95 square miles
- Contains significant hard clam resource – approved waters and special restricted waters
- Upper portion of the river recently downgraded to prohibited status – prompting MST study to determine sources of bacterial contamination

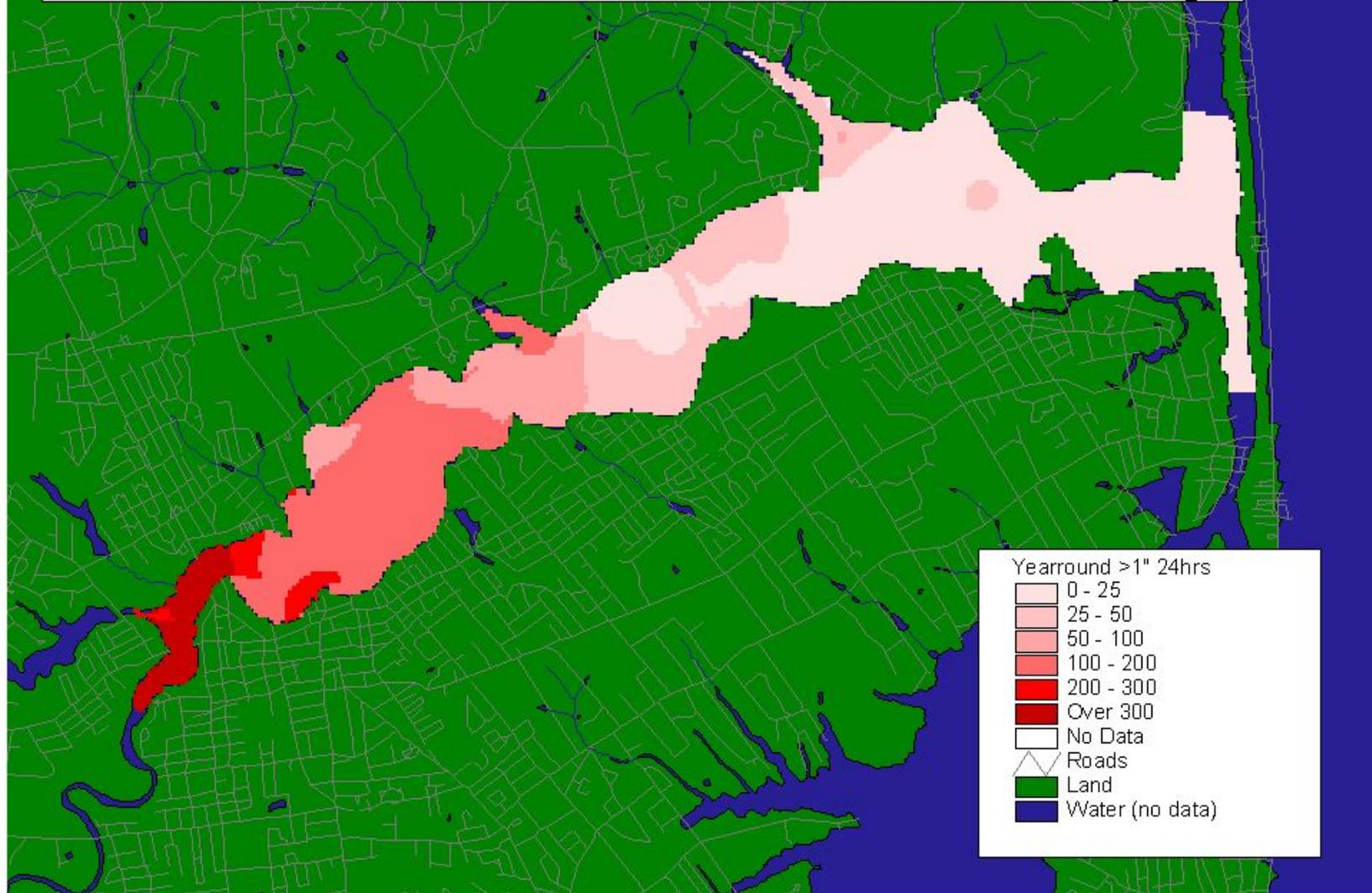


# First Step – Evaluate long-term data

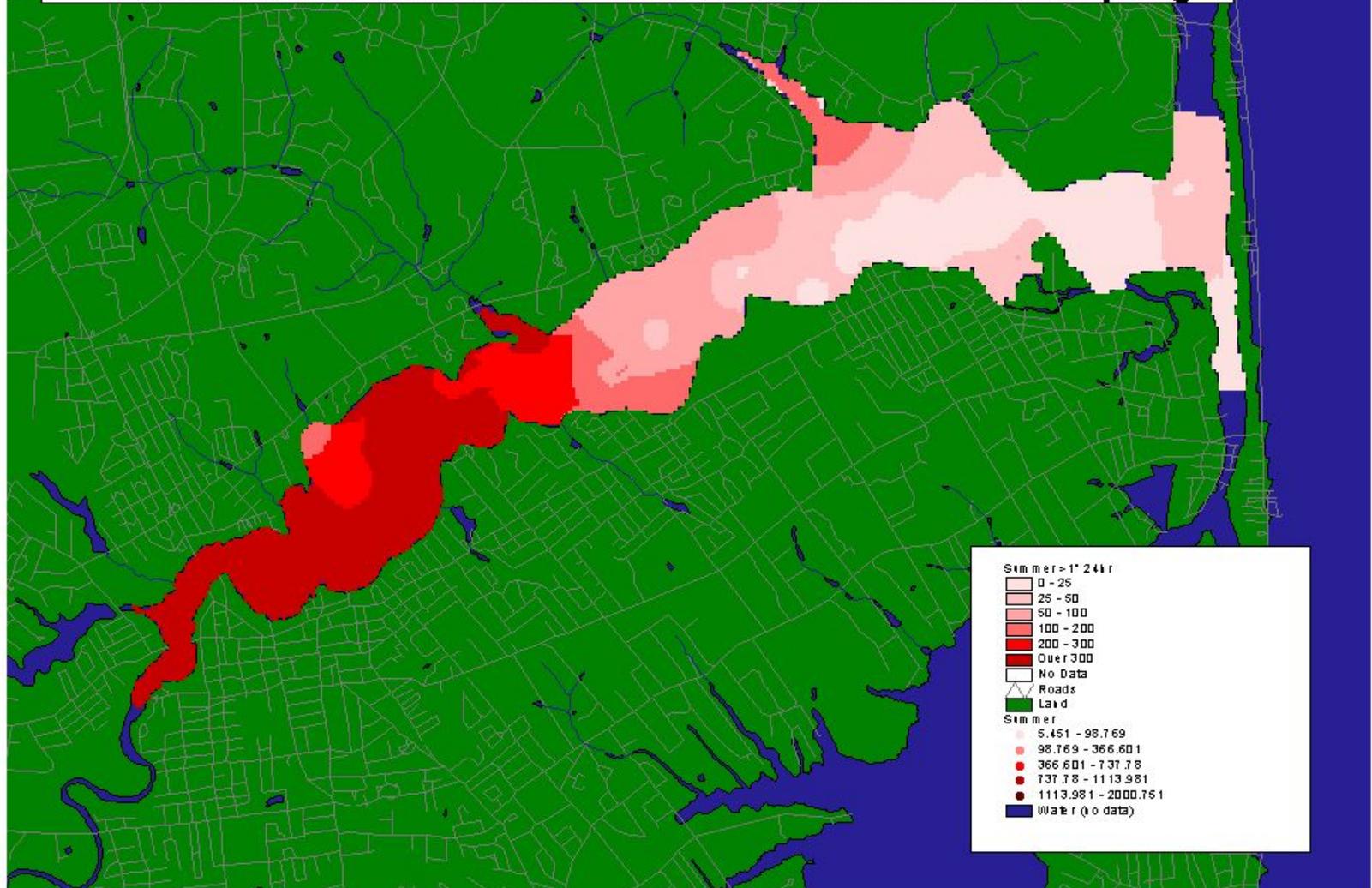


# Fecal Coliform (Geo. Mean) - 1993 to 2002

## Rainfall More than One Inch over 24 hrs Prior to Sampling



# Fecal Coliform (Geo. Mean) - 1993 to 2002 (Summer) Rainfall More than One Inch over 24 hrs Prior to Sampling



# Designing the Study: Actual and Potential Sources of Pollution



# Oyster Point



# Stormdrain @ Newman Springs Rd.



# Stormdrain Upstream Swimming River

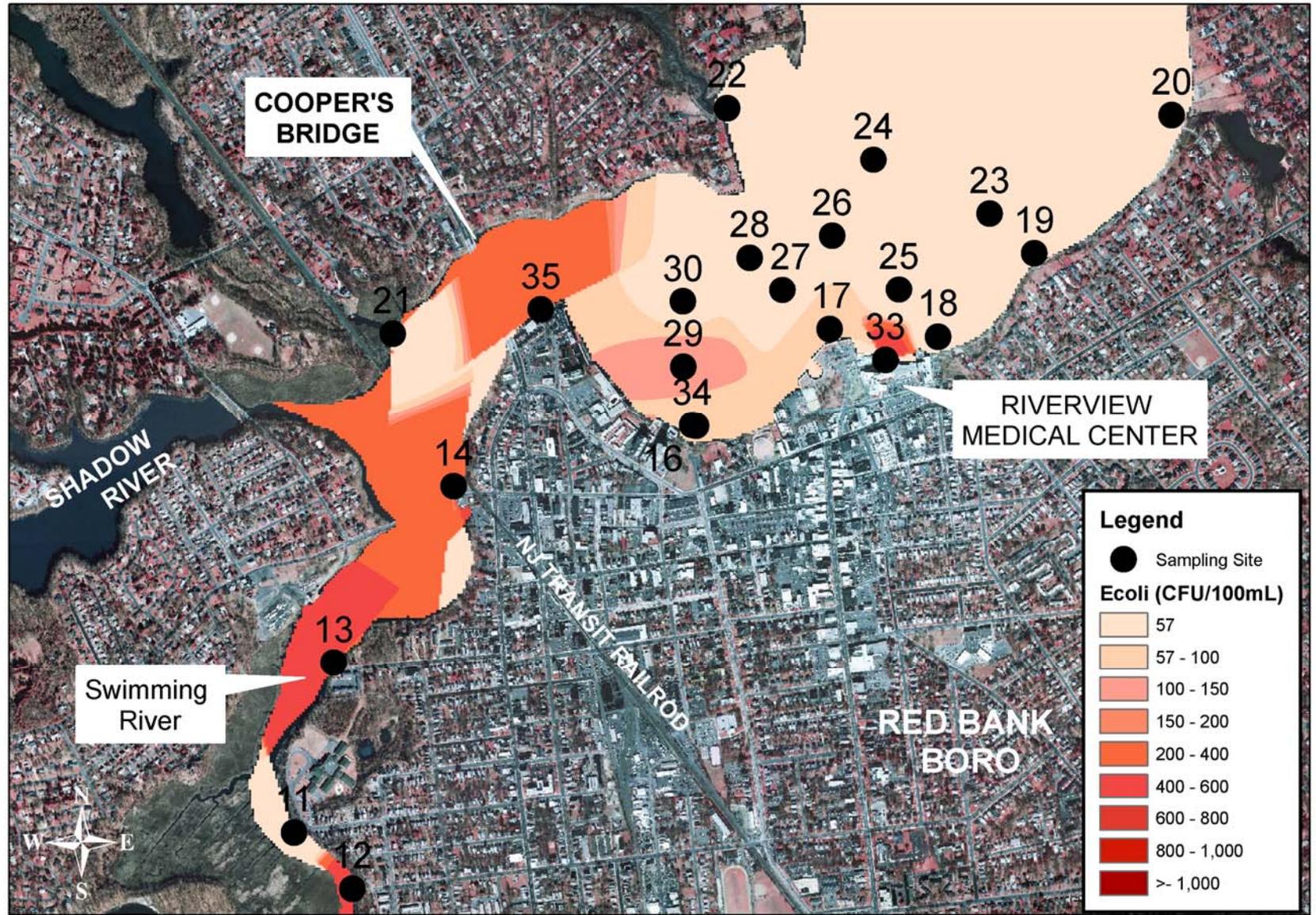


# Marsh Area in Vicinity of GSP



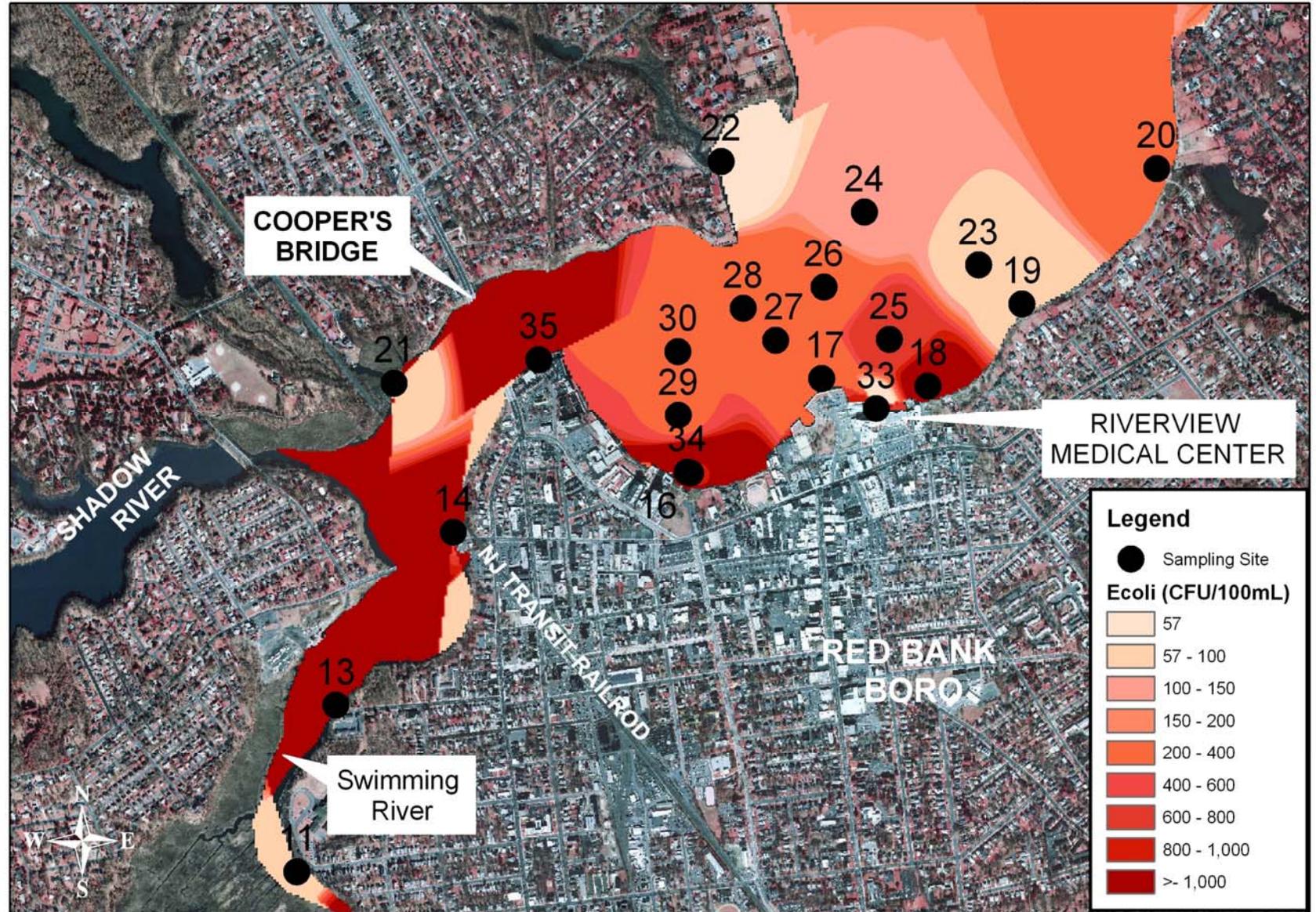
# Navesink River NPS Study

## Average Ecoli At Initial Sampling



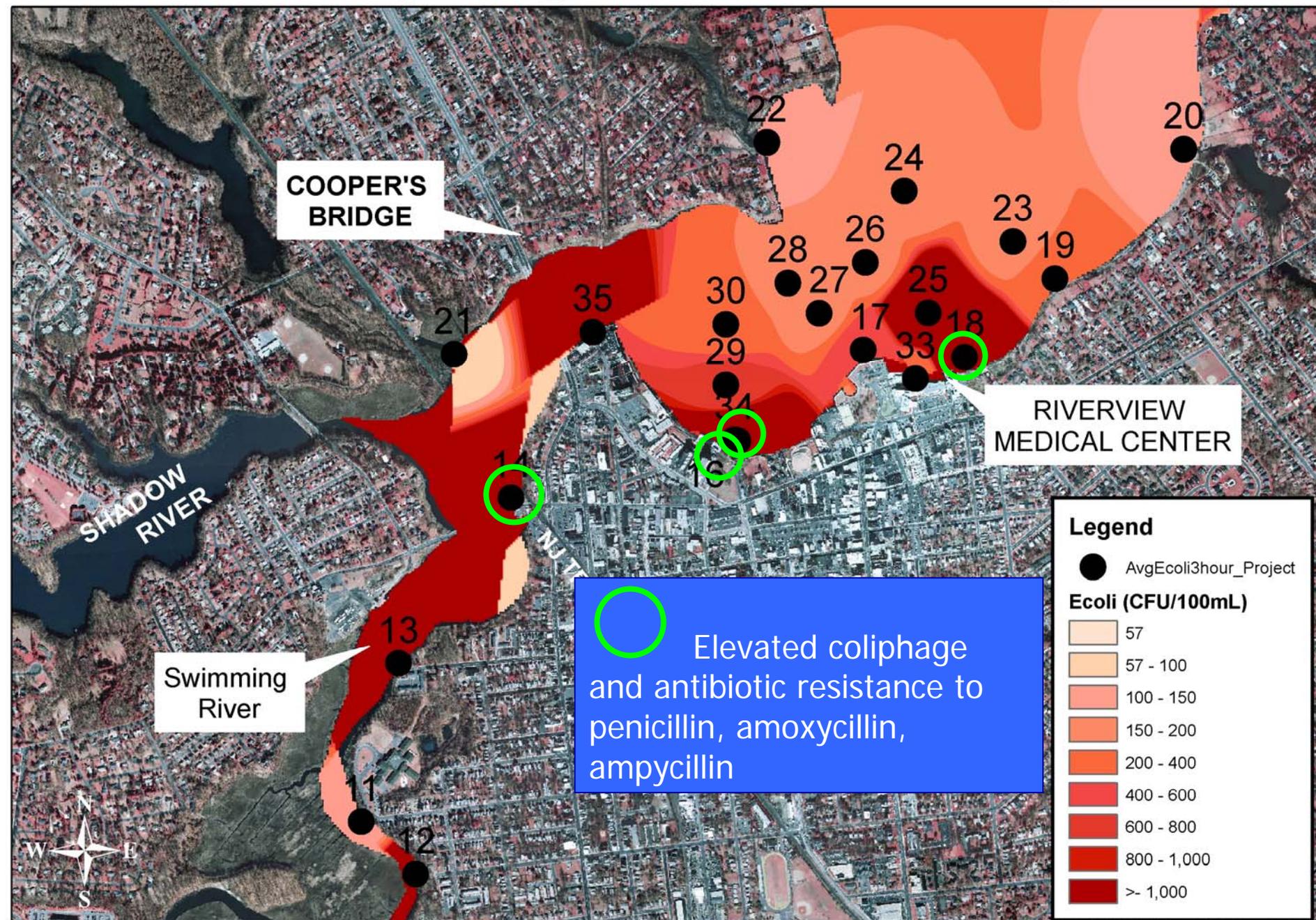
# Navesink River NPS Study

## Average Ecoli @ 1 Hour After Storm Event Began



# Navesink River NPS Study

## Average Ecoli @ 3 Hours After Storm Event Began



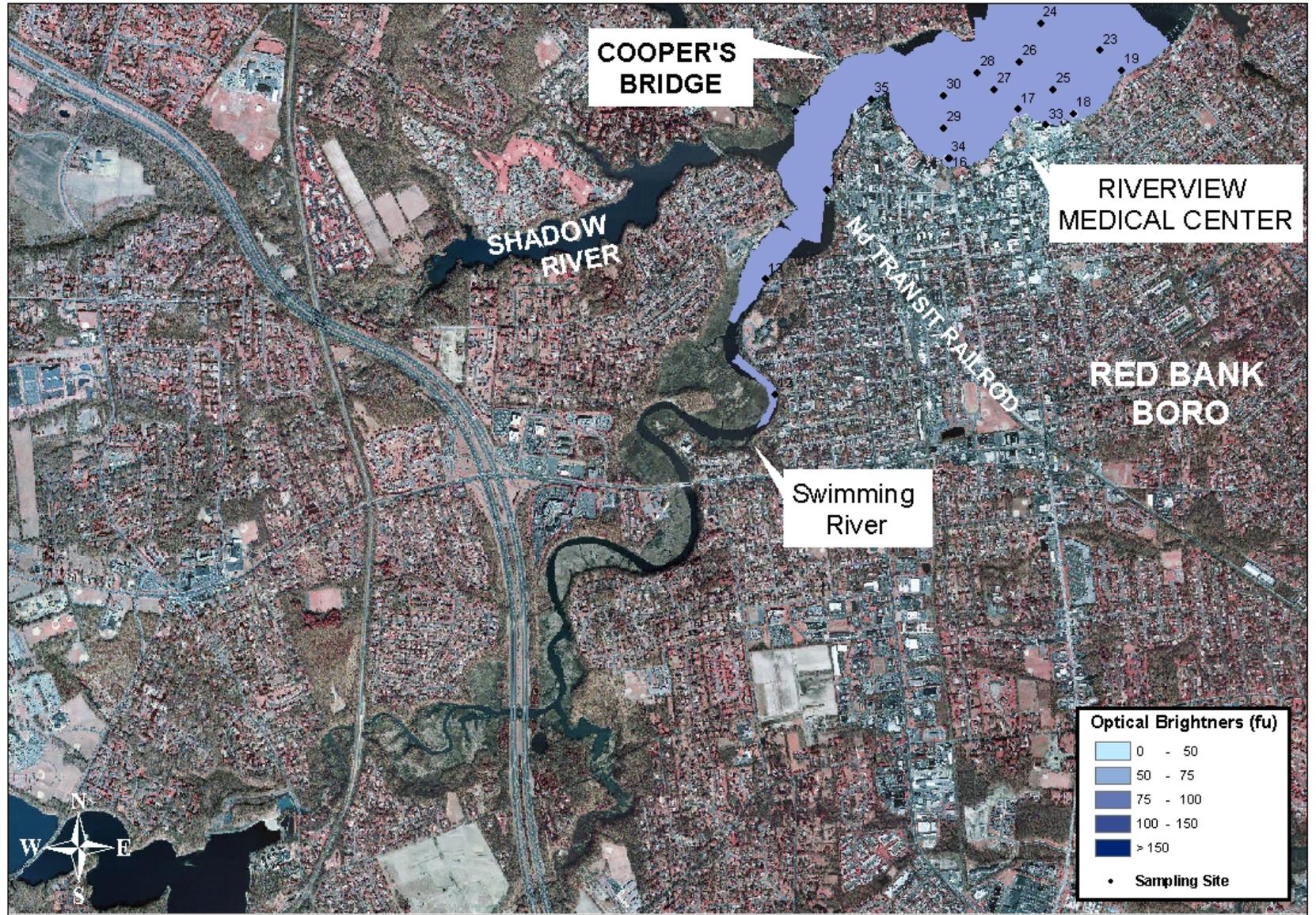
# Optical Brighteners

- Fluorescent Whitening Agents (FWAs) are compounds that can be measured and studied as an indicator of human sources of pollution
- Laundry detergents contain FWAs and are discharged in substantial quantities with household wastewater
- Fluorometric Detection – Turner Fluorometer



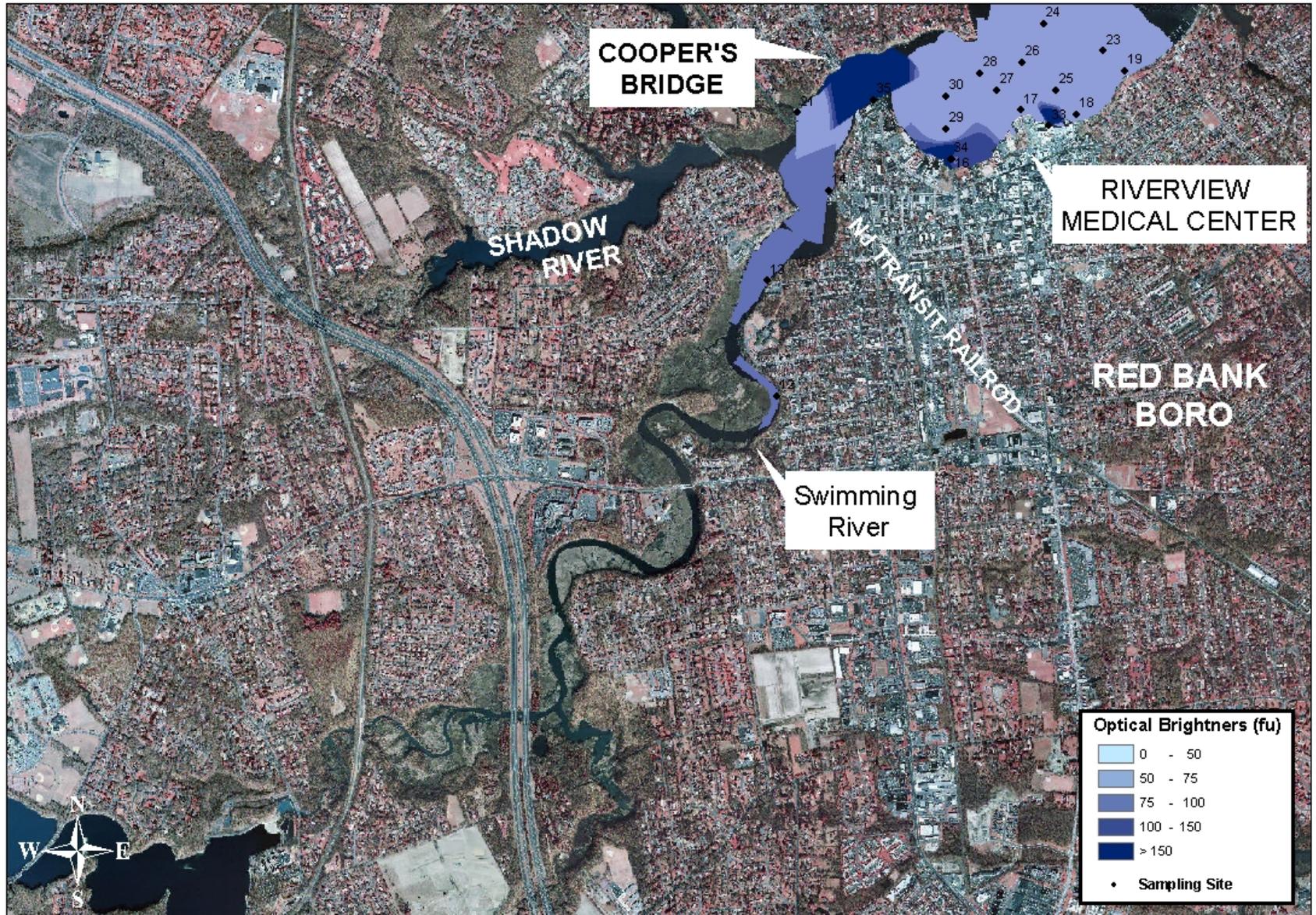
# Navesink River NPS Study

## Optical Brighteners Average Value (Background)



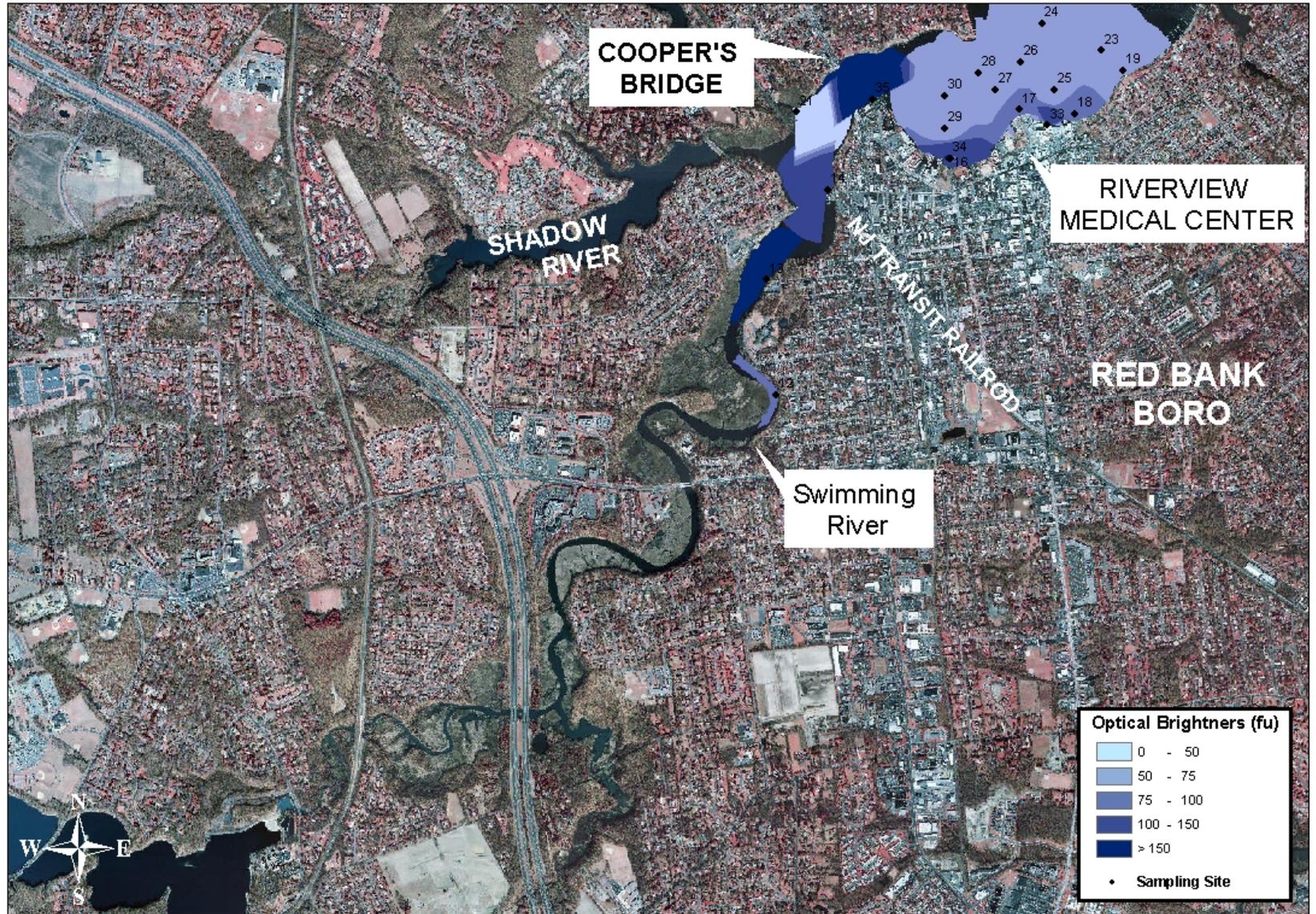
# Navesink River NPS Study

Optical Brightners Average Value (1 Hour)



# Navesink River NPS Study

Optical Brighteners Average Value (3 Hours)



# CASE HISTORY-IMPAIRED RECREATIONAL BATHING WATERS

- Wreck Pond – discharges via a spillway and 300' outfall to the ocean
- Elevated bacterial levels (enterococcus) following rainfall impacts ocean bathing beaches in the vicinity of the outfall
- This has resulted in a “precautionary closure” of these beaches following rain events  $>0.1$ ”





# Potential Pollutant Sources

- Upper Watershed
- Wreck Pond
- Stormwater discharges
- Resuspension of bacteria in sediments
- Offshore Impacts

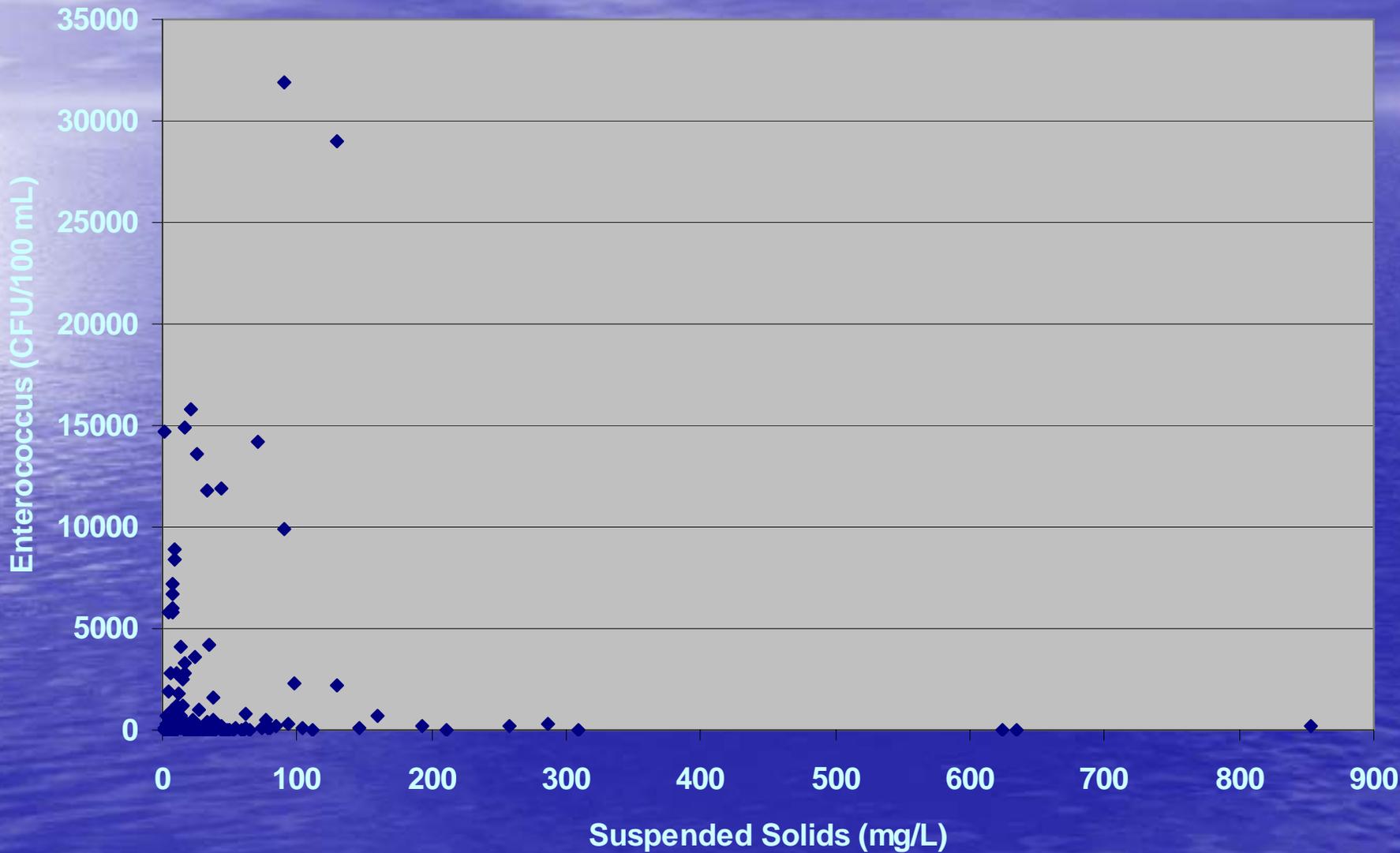


# Our Evaluation of Offshore/Ocean Impacts

- NJDEP Monitoring – 0.25 – 3.0 nm offshore
- >3,800 samples collected in the vicinity of Wreck Pond from 1993 – 2007
- Maximum level found was 95 FC MPN/100 mL
- 99.8% of the results were < 10 FC MPN/100 mL



# Enterococcus vs Suspended Solids



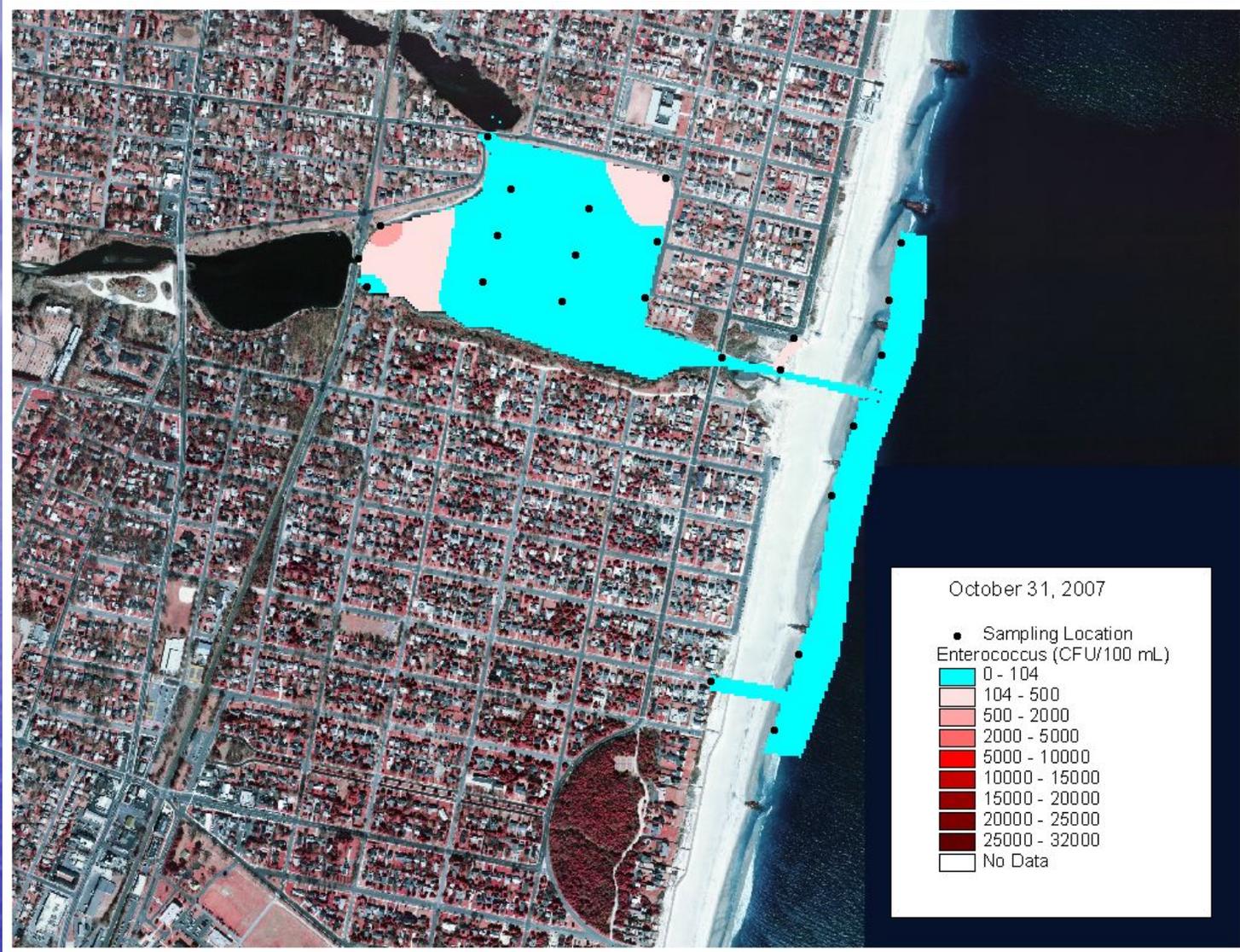
# Sampling



# Enterococcus Levels

Spring Lake /  
Sea Girt

10/31/2007  
@ 10 AM



No appreciable rainfall 3 days prior.

Wind Speed: ~10 kts

Wind Direction: SE

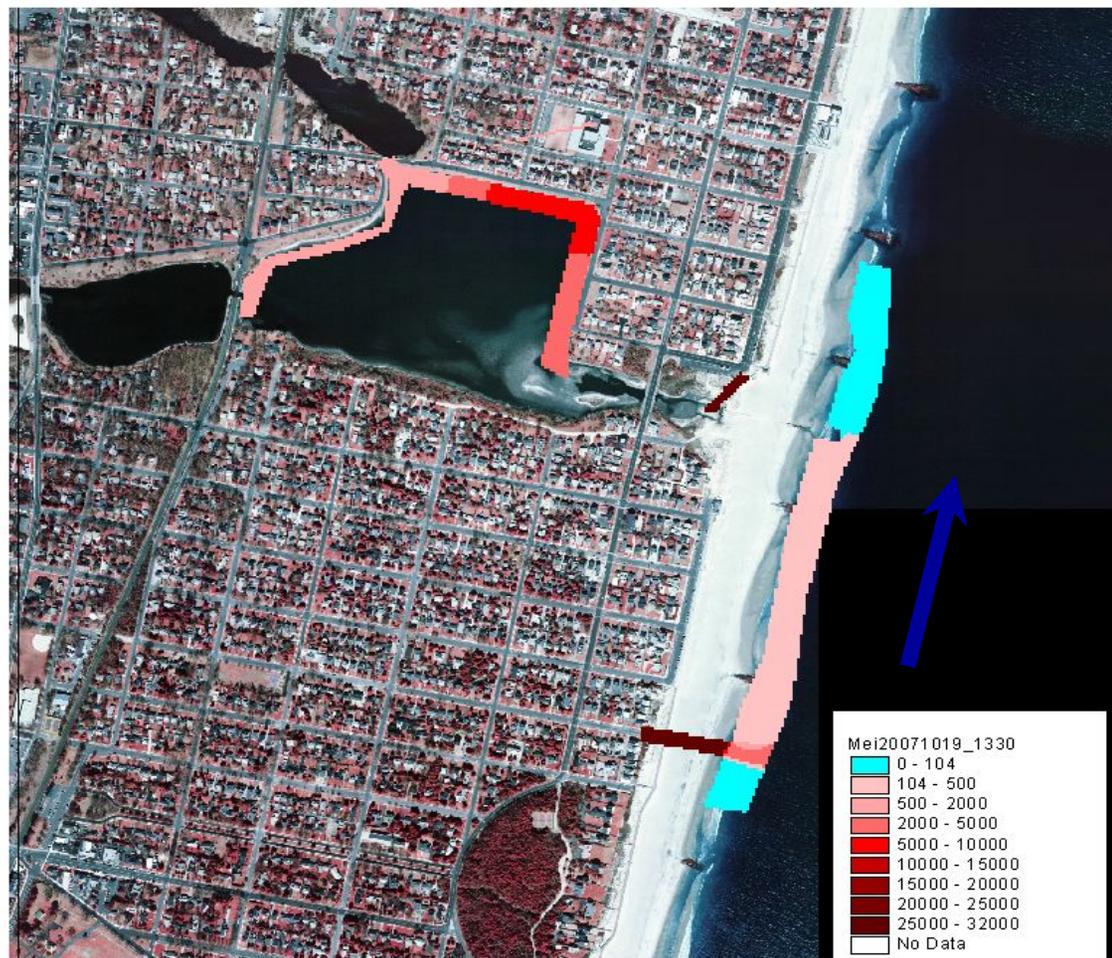
Total 24 hr. rainfall: 0.0"



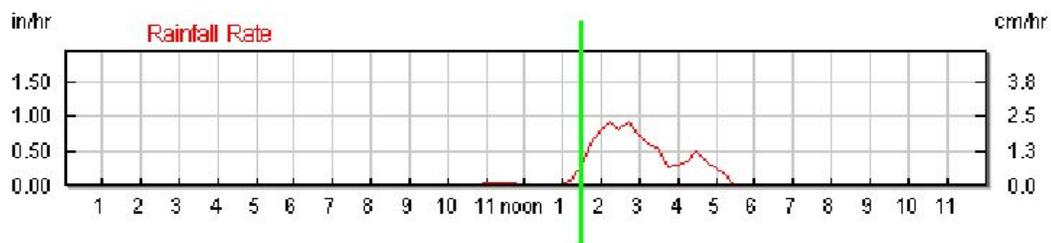
# Enterococcus Levels Spring Lake / Sea Girt 10/19/2007 @ 1:30 PM

- Early in storm event
- Elevated levels in Wreck Pond near discharge from Spring Lake
- Very high levels at Baltimore Ave. stormwater discharge
- Predominant nearshore ocean current is south to north.

Wind: S  
Tide: 2.58 ft > MLW  
24 hr. Rainfall: 2.20"



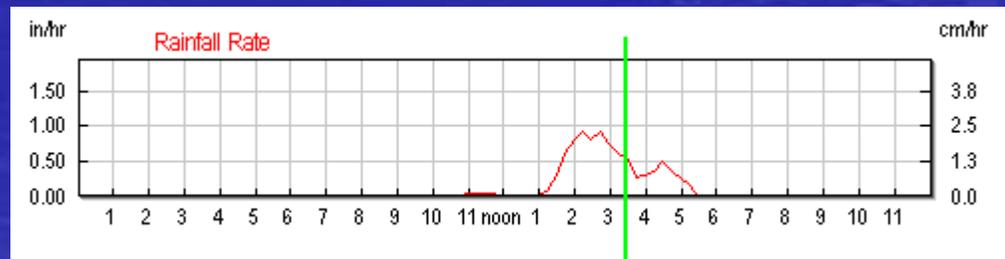
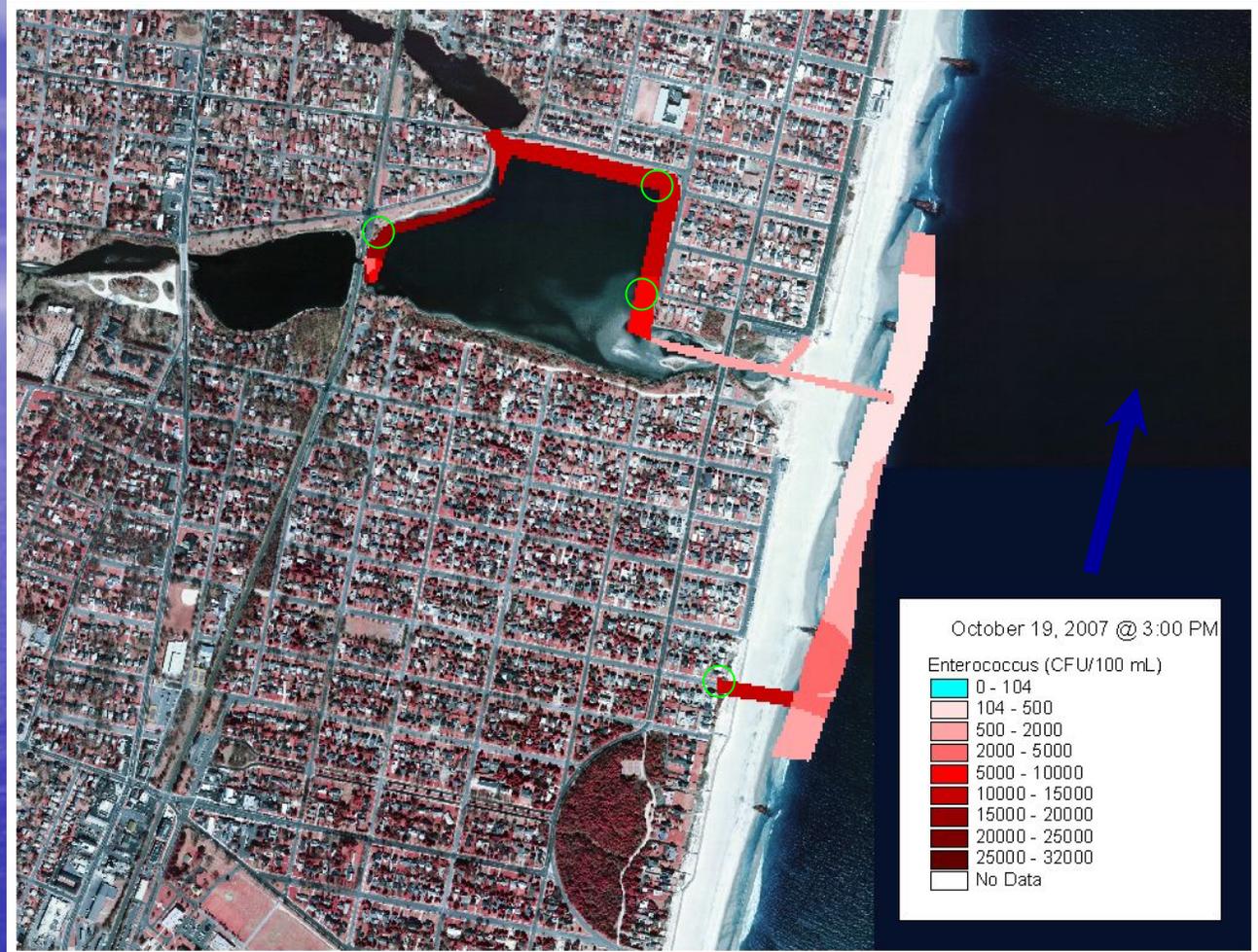
2007, NJDEP, Bureau of Marine Water Monitoring Levels reported as Enterococcus CFU/100 mL



Enterococcus Levels  
Spring Lake / Sea Girt  
10/19/2007 @ 3:30 PM

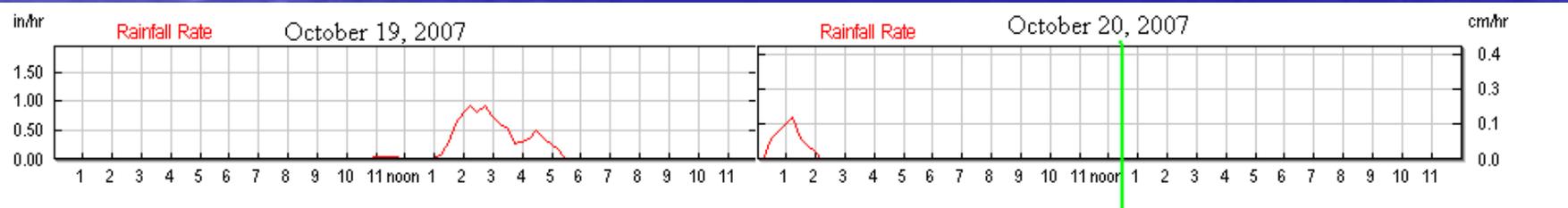
Wind: S  
Tide: 4.13 ft > MLW

Green Circles show  
where ARA suggests  
human source.



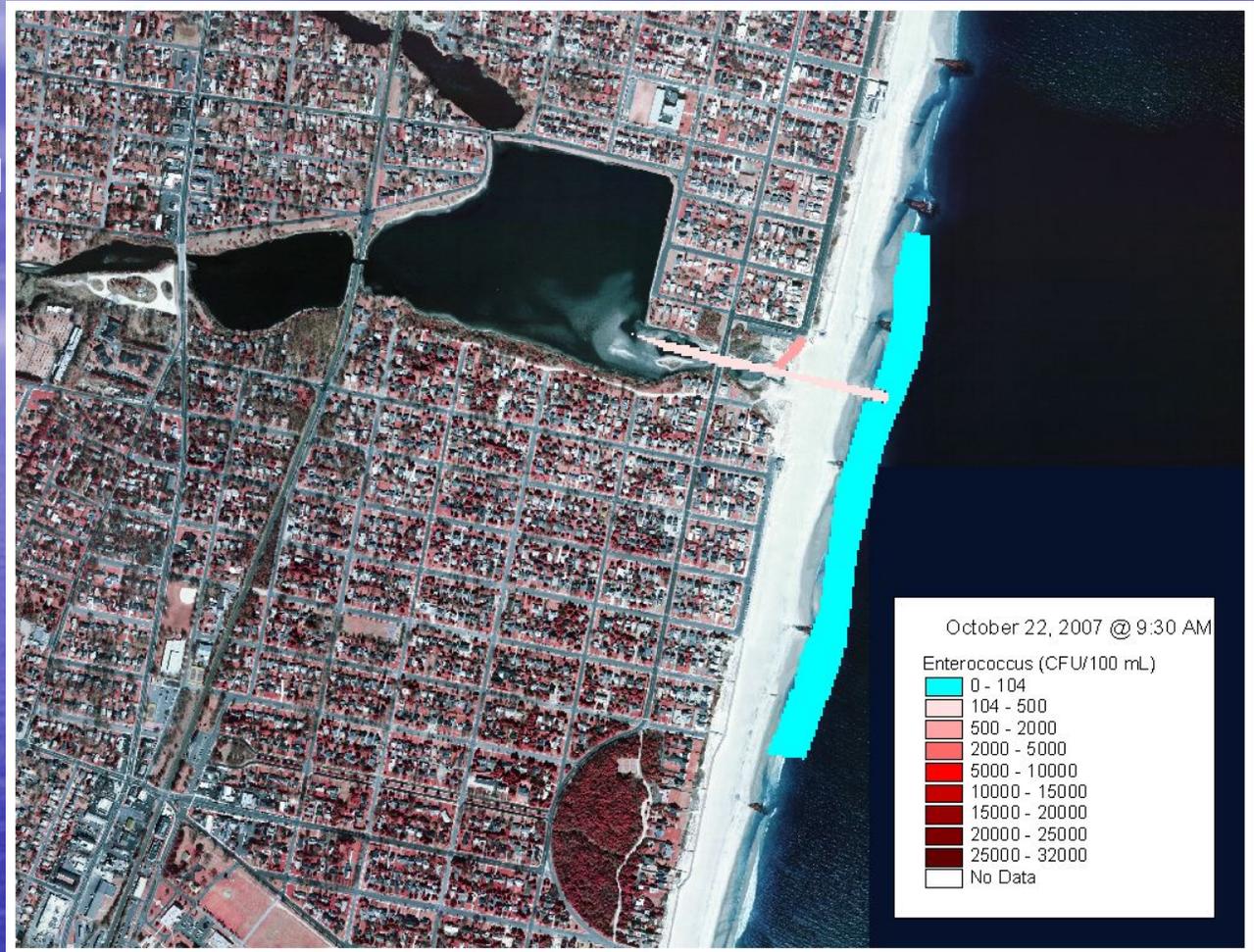
Enterococcus Levels  
Spring Lake / Sea Girt  
10/20/2007 @ 12:30 PM

Tide: 2.22 ft > MLW



Enterococcus Levels  
Spring Lake / Sea Girt  
10/22/2007 @ 9:30 AM

Tide: 4.31 ft > MLW



# Enterococcus Sources

- Upper Watershed – not impacting within 24 hrs.
- **Wreck Pond (stormwater discharges to it)**
- **Stormwater discharges (Baltimore Ave)**
- ~~Resuspension~~ of bacteria in sediments – no evidence of this as a major impact to beaches.
- Offshore Impacts – no evidence of this as a major impact to beaches.



# LESSONS LEARNED

- Importance of the shoreline survey (site selection, sampling logistics)
- Need for intensive monitoring including APC, first flush, temporal and spatial considerations
- No "silver bullet" Need to use multiple MST lab methods
- Evaluation of the data collectively to build "Scientific Weight of Evidence"



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MICROBIAL SOURCE  
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**Scientific  
Weight  
Of Evidence**

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Optical Brighteners**

**Perform intensive monitoring  
under APC regime using conventional  
indicators: FC, Entero, *E.coli*. Sample  
at dry, first flush, hour intervals, next day**

**Perform shoreline survey of the watershed  
Utilize GIS and land use coverage  
Consider sampling logistics**

**Identify impaired areas (i.e. beach closures,  
closed shellfish areas) – based  
on long-term monitoring data analysis**



# Contact Information

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