

While we're waiting ...

- Please complete the:
 - Pre-test (blue)
 - Background information (pink)
 - Photo release (white) – **Hand in**
- Put your name on a piece of paper to win one of the Giant *E. coli* Microbes



E. coli Monitoring in Streams by Volunteers

Barb Liukkonen, U of MN

Eric O'Brien, Iowa DNR

Jerry Iles, Ohio State University

Kris Stepenuck, WI Extension & DNR

Lois Wolfson, Michigan State University



Introduction & Project Summary

- Project funding and partners
- Kits evaluated
- Statistical analysis
- Using the data
- Recommendations



Citizens Monitoring Bacteria

Oct 2003 – Sep 2007

- 6-state research and outreach project
 - IN, IA, MI, MN, OH, WI
- Funding from CSREES
- **Goal:** to test accuracy, reliability, and user satisfaction with test kits



Photo by C. Kowalczak

Project Partners



Water Action Volunteers



Project Team Members



CREATION OF LEVELS
TEC for Views Higher
than 1000 ft. Above

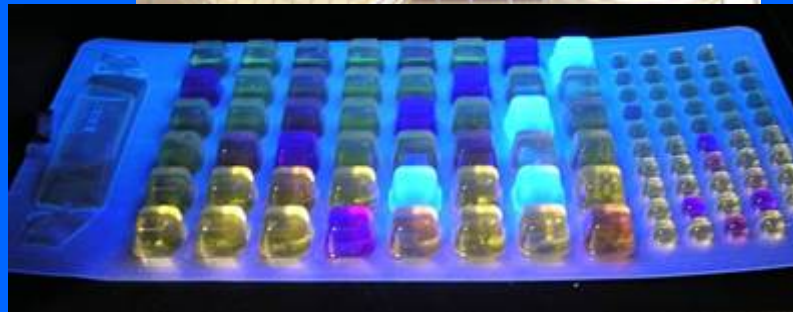
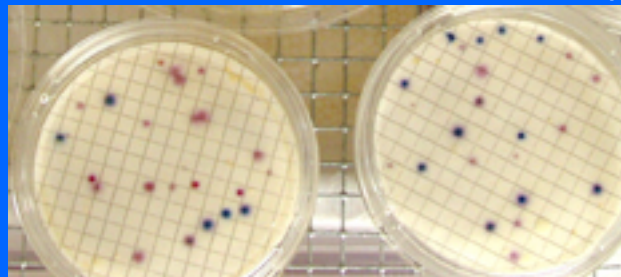


Why research *E. coli* test kits with volunteers?

- Many kits available and being used
- No comparative, independent study
- Cost of lab analysis is high; access to certified labs is problematic
- Citizens are interested and knowledgeable
- Citizens want an easy, reliable, inexpensive test kit

2004 – Year 1

- 5 methods tested in Iowa and Indiana
 - Coliscan[®] Easy Gel (incubated)
 - Coliscan[®] Easy Gel (not incubated)
 - 3M[™] Petrifilm[™]
 - Coliscan[®] MF Method Kit (*IN only*)
 - Colisure[®] Method with IDEXX Quanti-Tray/2000[™] (*IA only*)



Recommended the “best” kit from Year 1

Selected on the basis of:

- **Accuracy**
- **Volunteer satisfaction**
- **Cost**

Test Kits - Years 2 & 3

Coliscan Easygel-incubated



Used in MI, MN, OH, WI;
IA & IN continued others

3M Petrifilm -incubated

Each test costs about \$2



Consistent volunteer training

- **4-5 hour training, covering ...**
 - Background, protocols, QA, practice preparing and interpreting plates
- **Standardized curriculum and manual**
- **Evaluation and tracking**
- **Used same equipment and supplies**

Consistent volunteer training

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Citizens Monitoring Bacteria:

A training manual for monitoring *E. coli*



2nd Edition



A regional partnership between IN, IA, MI, MN, OH and WI

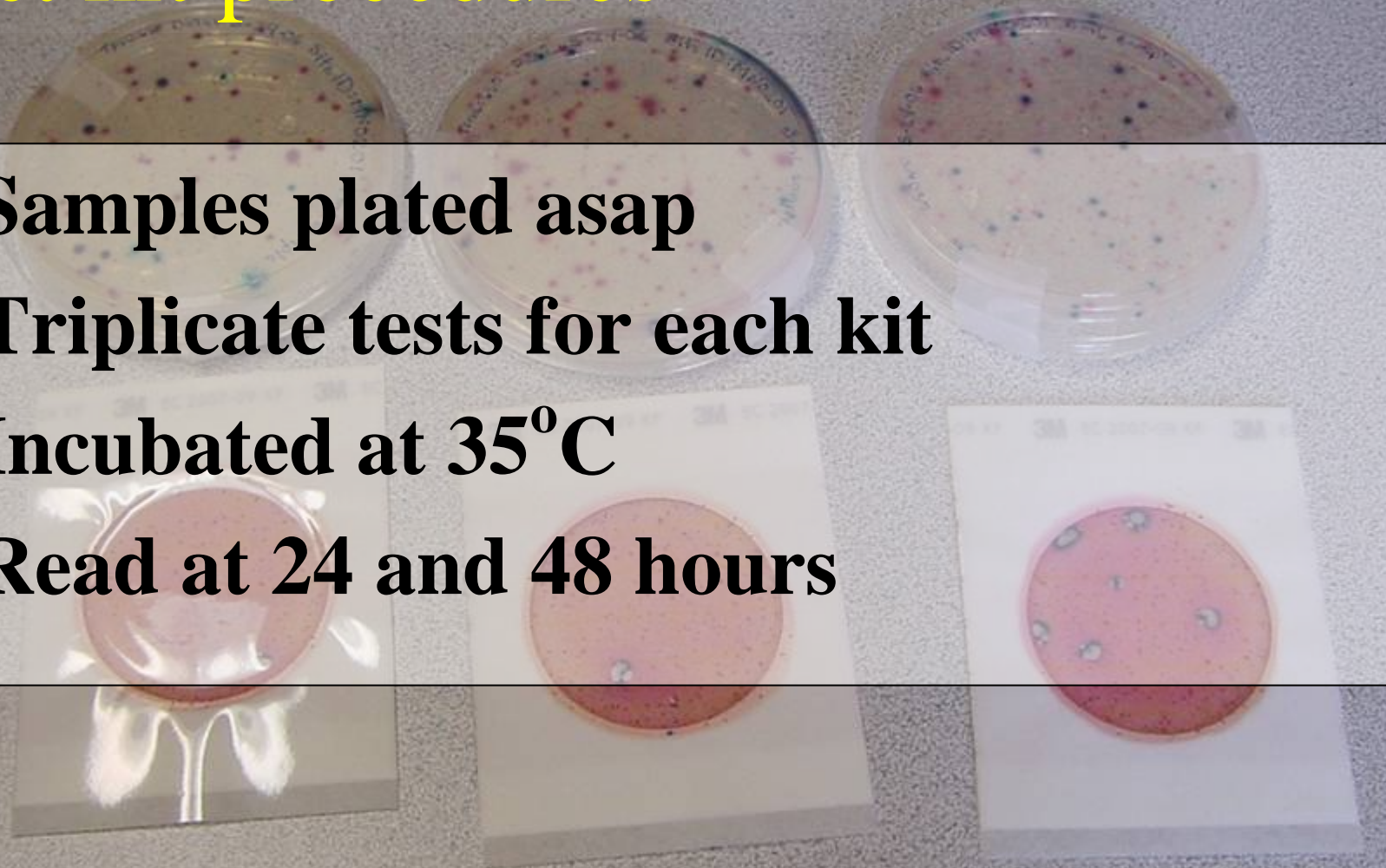


Field Visits

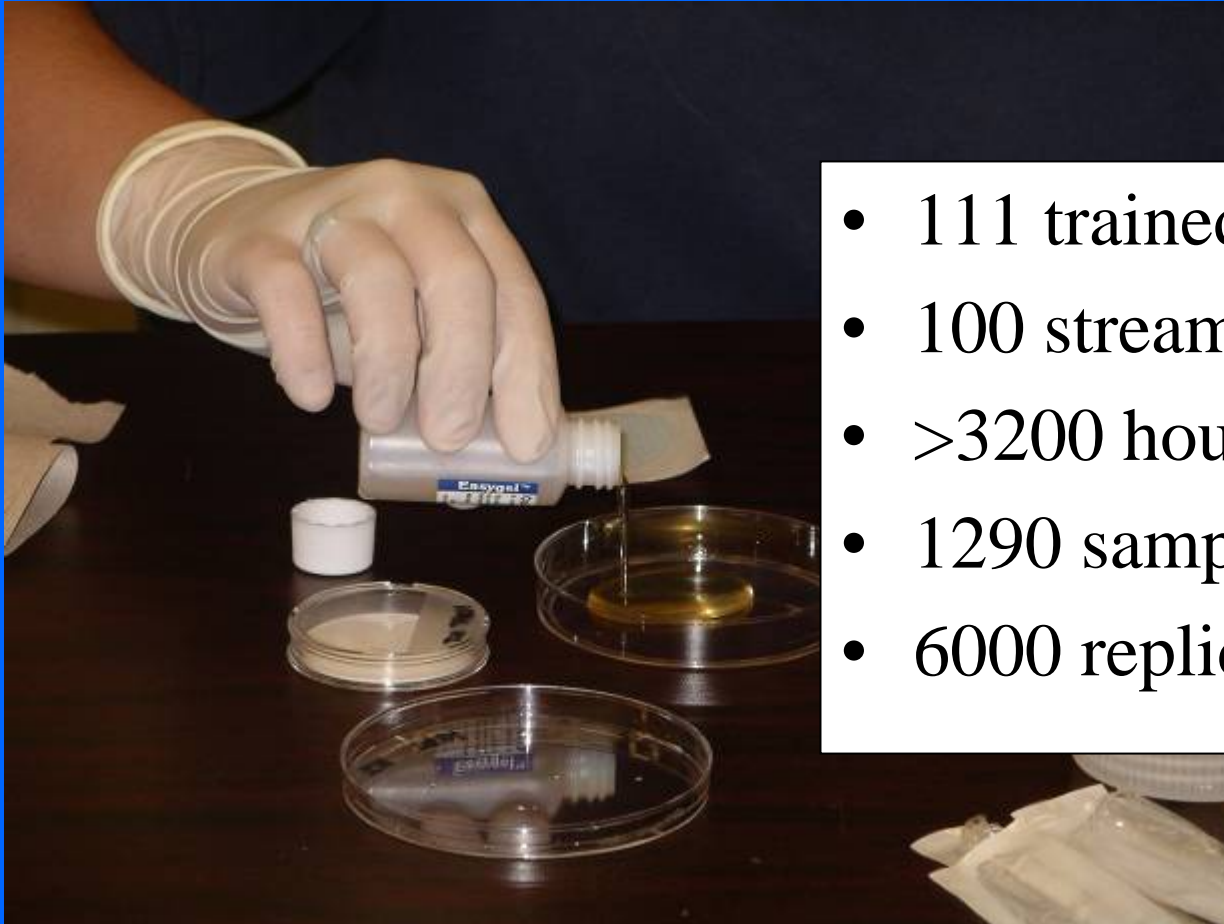
- **Sampled weekly, 1 or 2/month**
- **Recorded field conditions: stream level, weather, temps, T-Tube**
- **Collected one sample – split into 2**
 - 1 sample sent to certified lab
 - 1 tested at home
- **Sample sent to lab on ice, within 24 hrs**

Test kit procedures

- **Samples plated asap**
- **Triplicate tests for each kit**
- **Incubated at 35°C**
- **Read at 24 and 48 hours**



2004 - 2006 in 6 states



- 111 trained volunteers
- 100 stream & lake sites
- >3200 hours contributed
- 1290 samples collected
- 6000 replicates with test kits



So ... how well do they work?

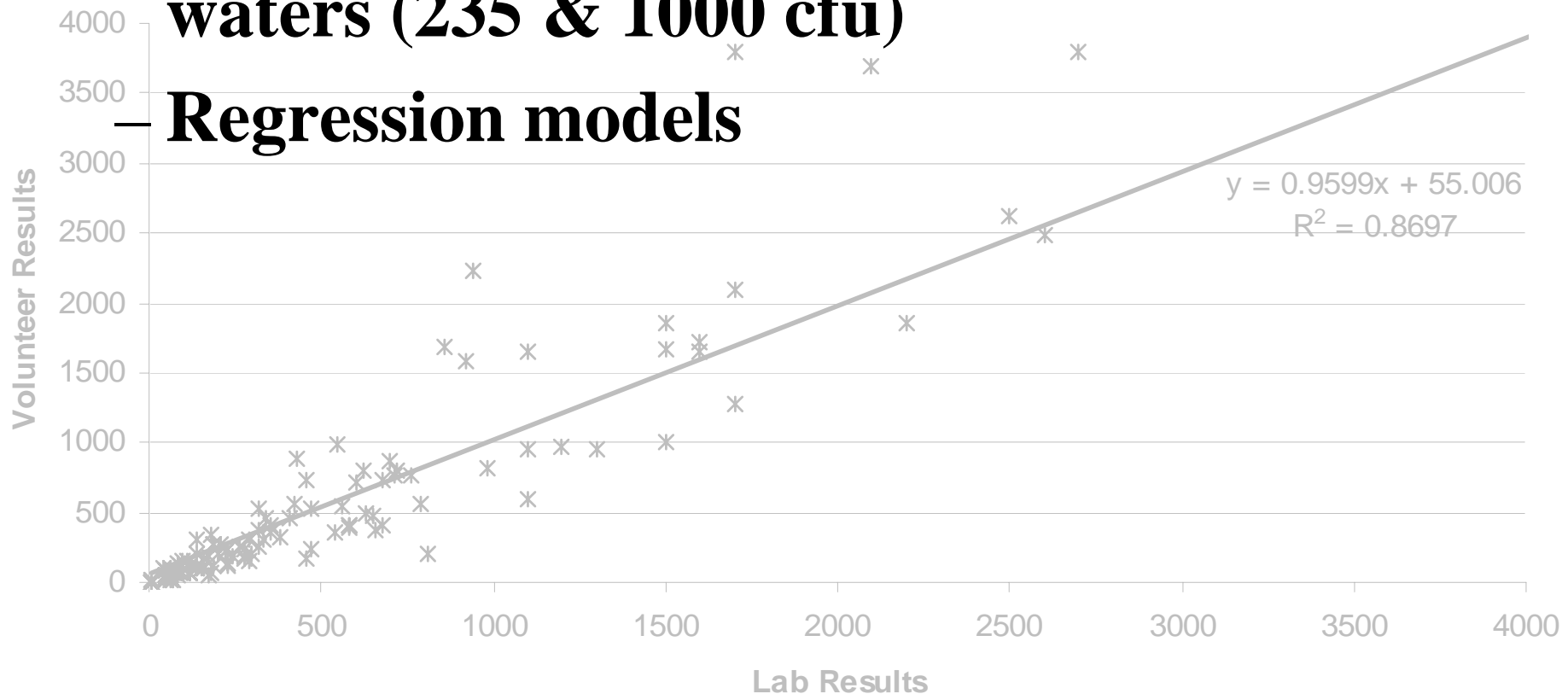


Data Analysis

- Evaluation of:

- Ability to make distinctions on impaired waters (235 & 1000 cfu)

- Regression models



Threshold levels

Petrifilm @24 hrs, 2006

Percent of samples with test kit and lab values **both** either above or below the 235cfu value

80.9% agreement

lab \ kit	< 235 cfu	> 235 cfu
< 235 cfu	64.6 %	6.1
> 235 cfu	12.9	16.3

Threshold levels

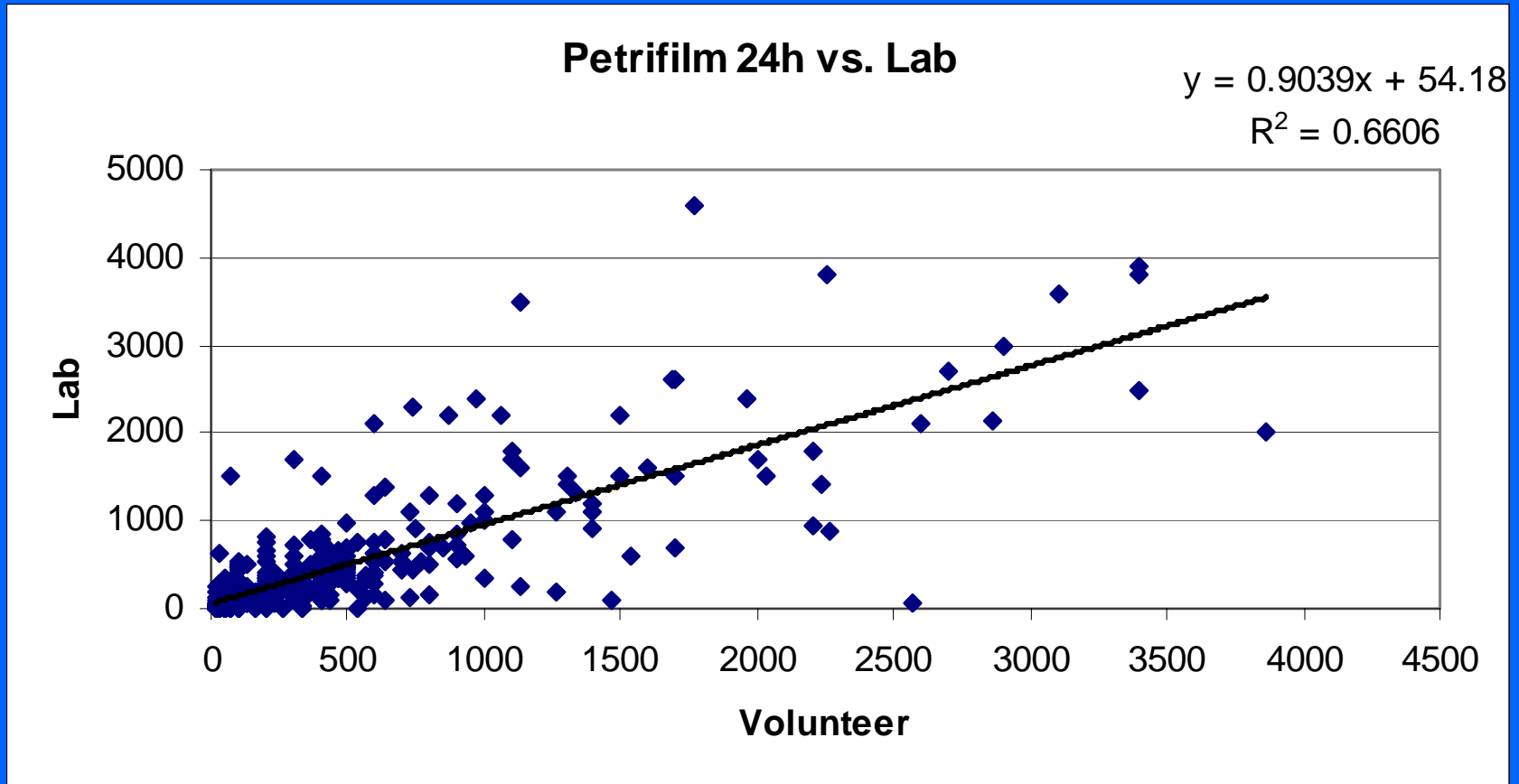
Petrifilm @24 hrs, 2006

Percent of samples with test kit and lab values **both** either above or below the 1000 cfu value

93.2 % agreement

lab kit \	<1000 cfu	>1000 cfu
<1000 cfu	89.8 %	4.1
>1000 cfu	2.7	3.4

Petrifilm vs lab results, all data



Volunteers ...

- **Preferred Petrifilm (71%)**
 - Ease of use, interpretation
 - Limitation of just 1 ml
- **Contributed (on average):**
 - Time - 35 hours
 - Direct expenses - \$15.25
 - Mileage – 200 miles



Volunteers shared information

- 64% shared with neighbors and friends
- 30% with Lake or River Associations
- 24% with local resource managers
- 30% with elected officials
- 11% with state agencies
- 3 used the data to secure grant funding
- Helps target resources more effectively

What can the data be used for?



Photo by Wayne Goeken, 2006

- Classroom education
- Volunteer knowledge
- Public awareness
- Local decision-making
- Targeting resources
- Assessing water quality
- Impaired waters - TMDLs

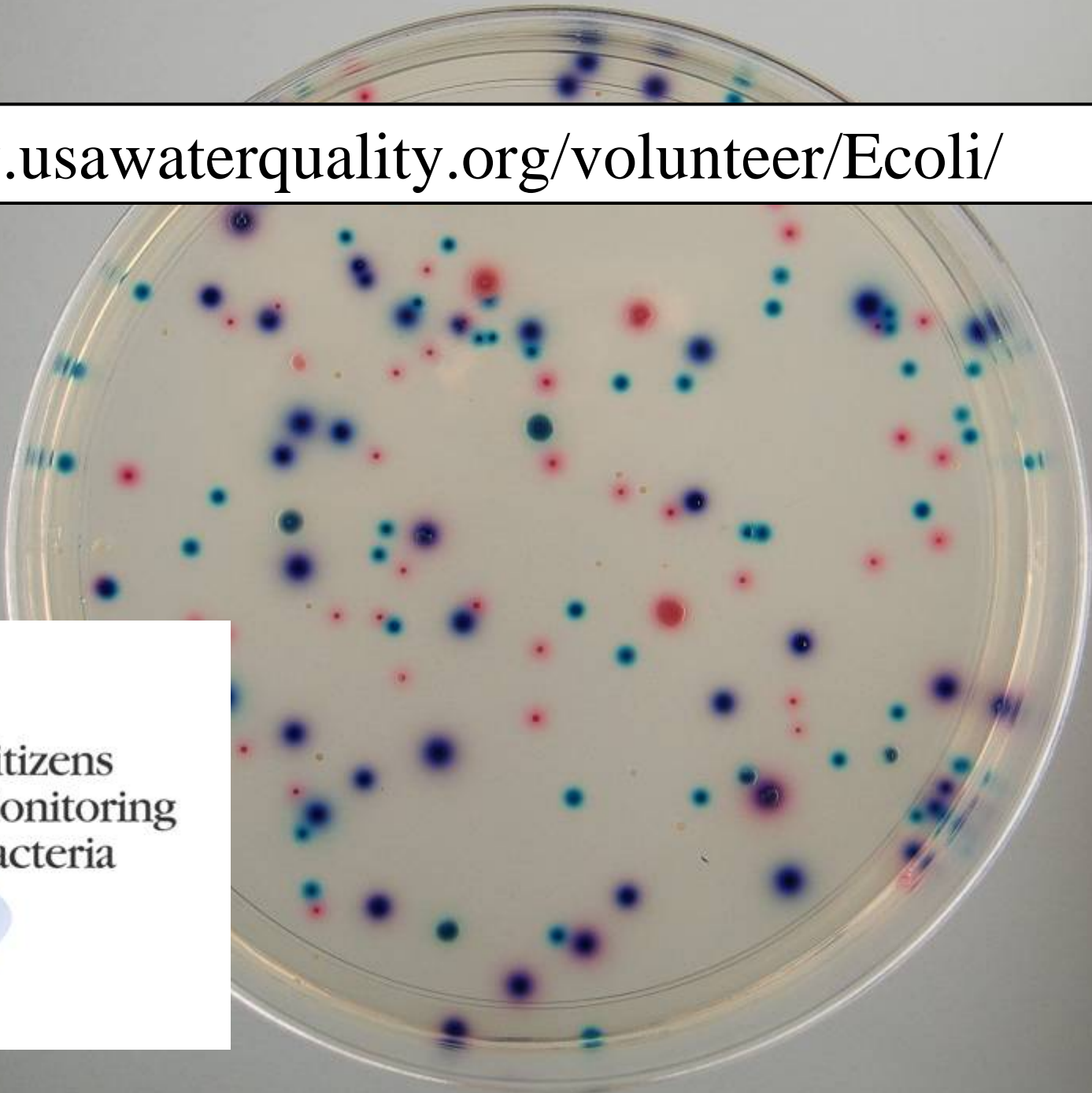


Conclusions

- Kits compared fairly well with lab analysis
- Kits are good for screening & targeting resources
- As much variability between labs as between test kits and labs
- Petrifilm and IDEXX essentially equal in performance
- Volunteers preferred Petrifilm & lower cost



www.usawaterquality.org/volunteer/Ecoli/



Bacteria 101 - Scope

- Bacteria as indicators
- Sources of fecal bacteria
- Health risks
- Standards for bacteria



Quick Review: Rules for Fecal Indicator Bacteria

- Bacteria from feces of warm-blooded animals
- Present in higher number than pathogens
- Nonpathogenic



- Do not persist in the environment



E. coli are used as indicators because they:

- Indicate fecal contamination
- Suggest the presence of pathogens
- Are easy to collect and analyze
- Are relatively safe to handle and generally harmless

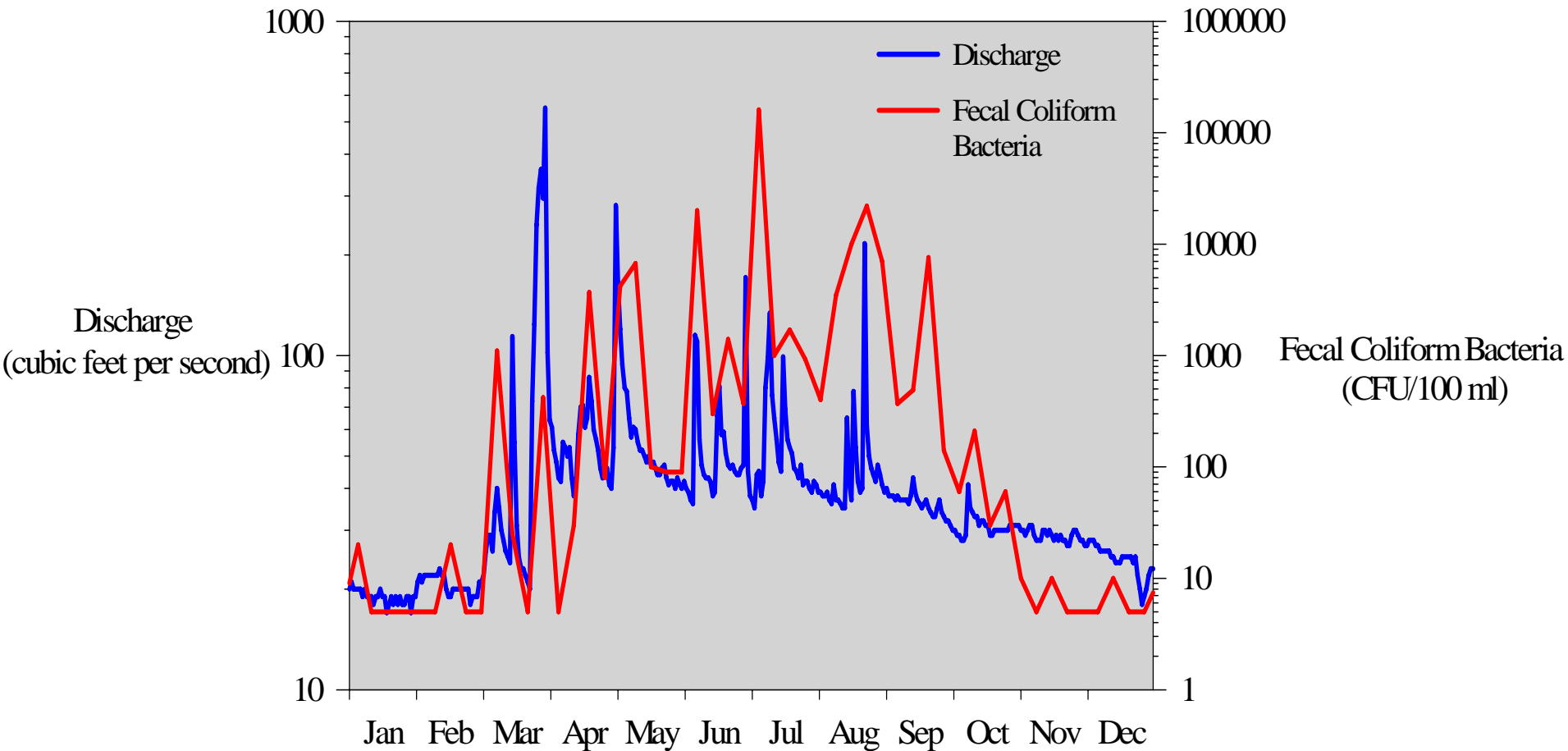


Indicator bacteria survival in environment

- Sunlight (UV radiation and white light) – *can cause die off*
- Temperature – *freezing destroys cells, but can survive at cold temps below the ice*
- In sediment – *may survive and thrive in bottom or bank sediments or at water interface in beach sand*
- In algal mats – *Cladophora, sun-dried, stored at 4°C 6 months*
- Water body conditions that enhance survival – *low light penetration, high turbidity, low salinity, presence of elevated nutrients and organic matter*

Bacteria levels can be related to flow: *More runoff = Higher bacteria counts*

Discharge and Fecal Coliform Bacteria
Bloody Run Creek, Clayton Co., IA



1993

Persistence in the environment (Academy Creek–Brunswick, GA)

Condition	Enterococci Most Probable Number
Colony-forming units g ⁻¹ of dried sediment	
Moist sediment	3,160
Dried 2 days and rewet 24 h after rewet	16,980 23,440
Dried 30 days and rewet 24 h after rewet	510 16,980
Dried 60 days and rewet 24 h after rewet	1,200 28,840



**Provided by Peter Hartel*

Bacteria levels are affected by:

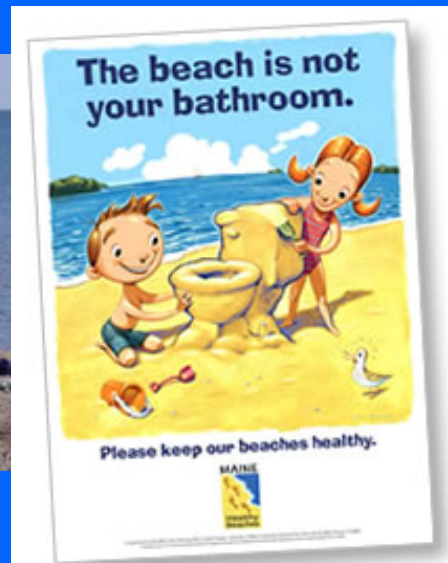


- Source and amount of loading
- Air and water temperature
- Rainfall and runoff

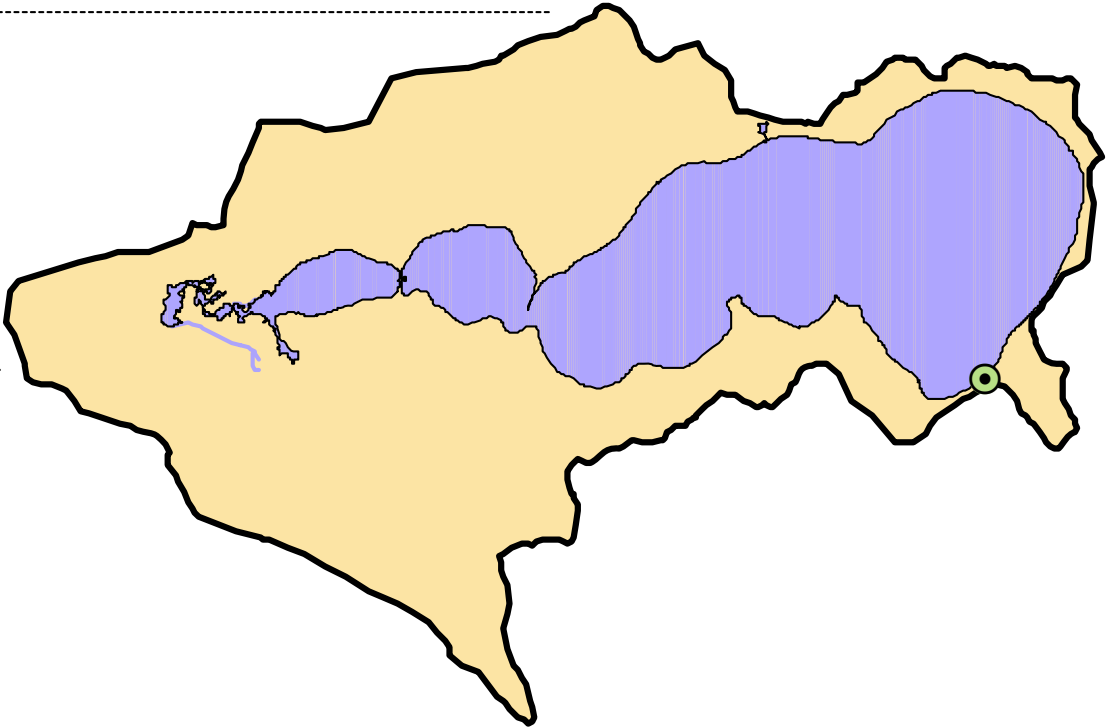
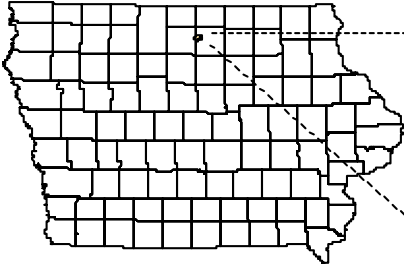


Sources of fecal bacteria

- Human sources – anytime fecal matter reaches water there will be bacteria
 - Wastewater treatment – inadequate or leaky septic systems or discharge from municipal systems
 - Swimming “*accidents*”, diapers
 - Boat dumping, fish derbies, water recreation



Clear Lake, Iowa



Beach



Rivers



Lake



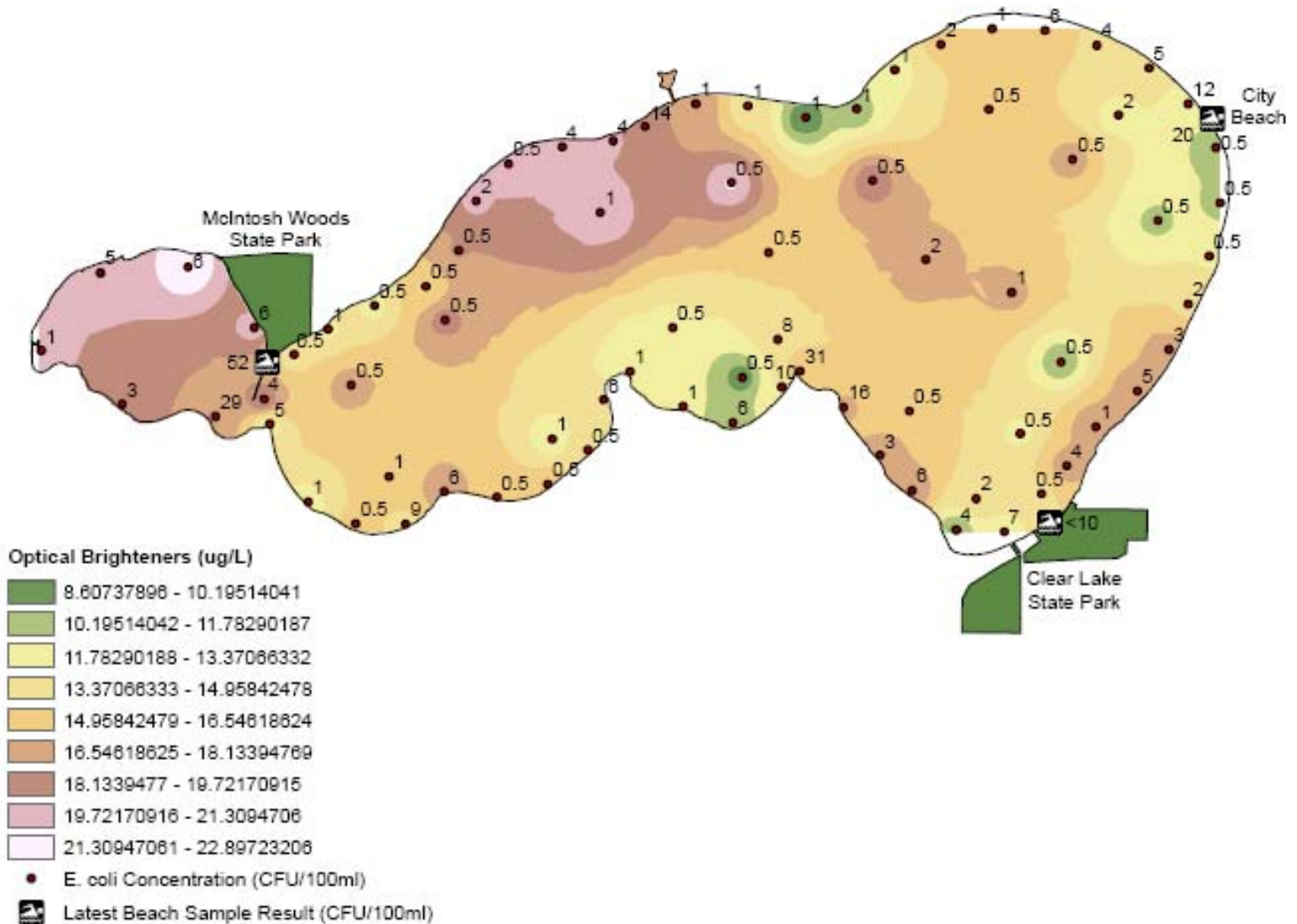
Watershed

0

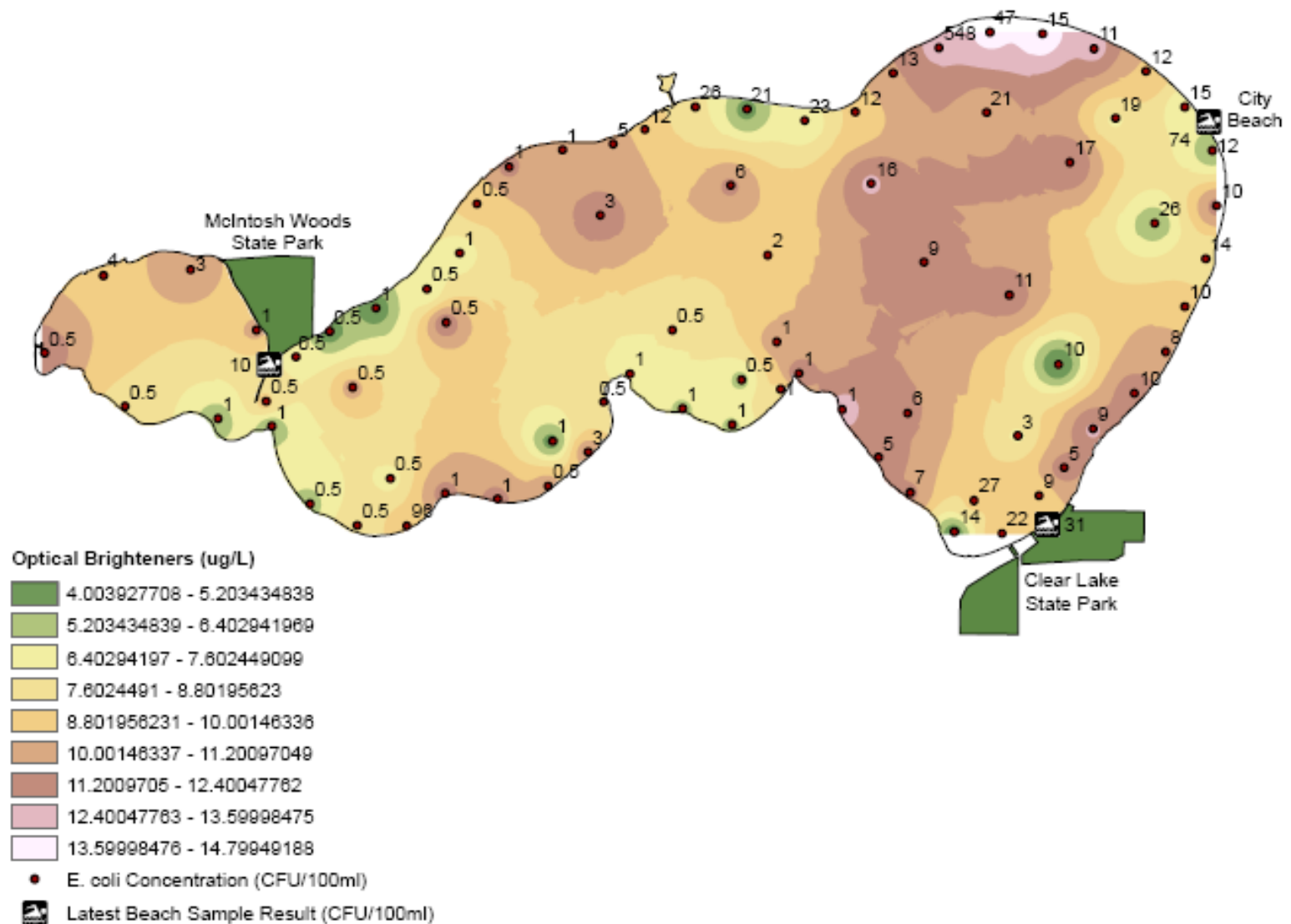
3 Miles



Clear Lake Fluorometry - July 5, 2007



Clear Lake Fluorometry - September 17, 2007



More bacteria sources

- Animal sources
 - Livestock – in streams, manure applied to fields, manure pits or lagoons
 - Wildlife – geese, ducks, deer, etc.
 - Pets





Waterborne Illnesses

- Pathogens are disease causing micro-organisms
- Three families cause illnesses (bacteria, viruses, and protozoans)
- Symptoms may be mild and confused with other diseases, so people may not realize that water made them sick



Keep in mind ...



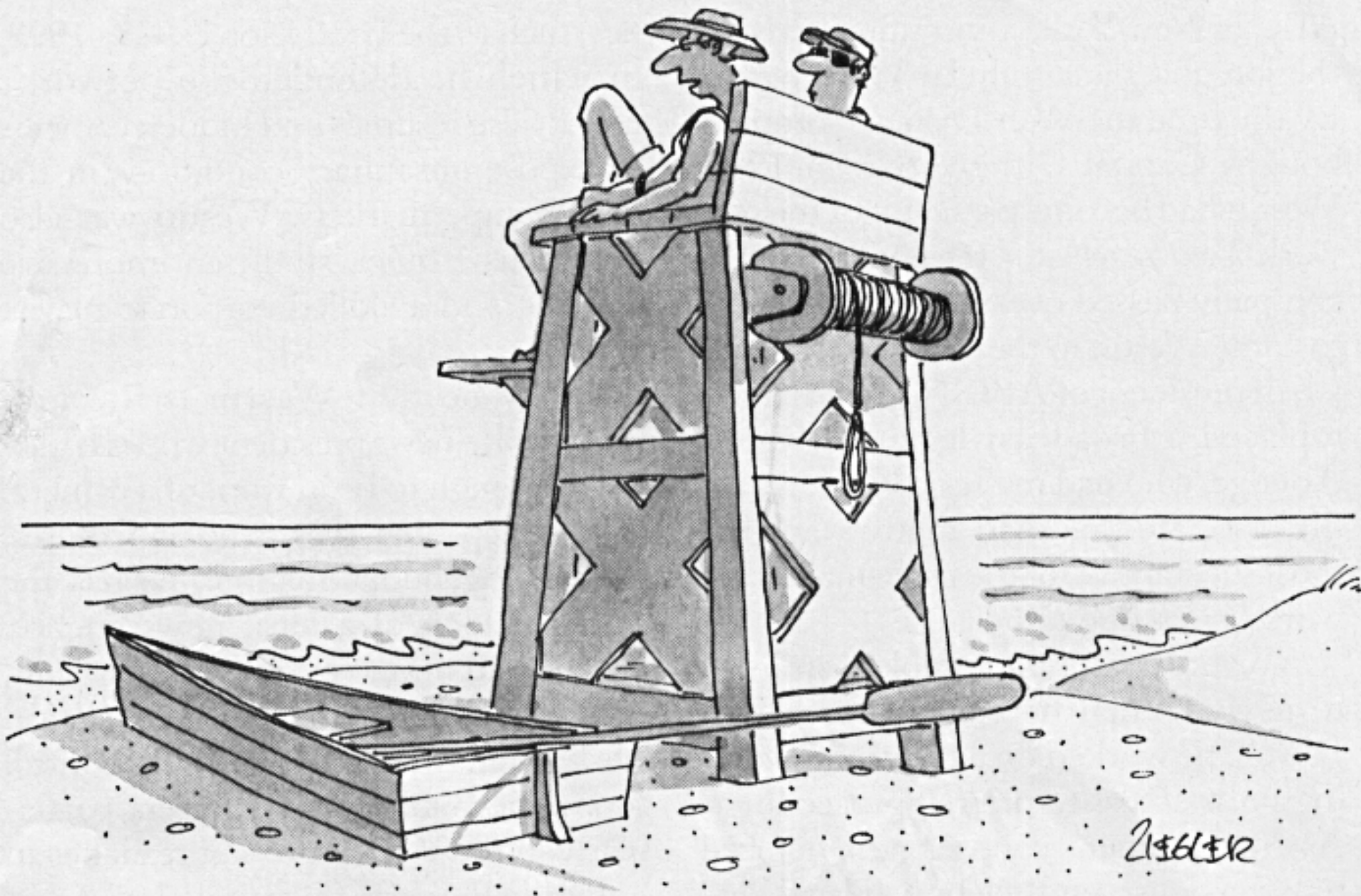
- Not all bacteria present a health risk
- Most won't make you sick, but some may
- Low infectivity rates



Why not sample for pathogens?

- Few laboratories have the capacity
- It's expensive
- Takes a long time for analysis
- Requires a large volume of water
- Most tests identify only one pathogen
- Most polluted waters have few pathogenic organisms - they are difficult to isolate and identify





"I adore the beauty and tranquillity of these raw-sewage days."

Current Monitoring Approach Leads to Errors



Courtesy Richard Whitman - USGS

Body contact standard



- Indicator of potential health risks from body contact
- Varies by state – check YOUR state's standards
- EPA one time standard is 235 cfu per 100 ml for swimming beach advisories



Water Quality Guidelines-1986

US EPA recommended guidelines

Recommended Guideline = $\frac{\text{Fecal Coliform Std} \times \text{Geo. Mean of New Indicator}}{\text{Geo. Mean of Fecal Coliforms}}$

Example for enterococci in fresh waters:

$$\begin{aligned} \text{Recommended Guideline} &= \frac{200 \text{ FC}/100 \text{ ml} \times 19 \text{ ENT}/100 \text{ ml}}{115 \text{ FC}/100 \text{ ml}} \\ &= \frac{3800 \text{ ENT}/100 \text{ ml}}{115} \\ &= 33 \text{ ENT}/100 \text{ m} \end{aligned}$$

where: FC = Fecal Coliforms
ENT = enterococci

- Geometric Mean Shall not exceed 126 *E. coli* / 100ml
- Geometric Mean shall not exceed 33 *enterococci* / 100ml
- Geometric Mean shall not exceed 35 *enterococci* / 100ml

Where did the numbers come from?

Pre-1968	Most States	1000 Coliforms
1968	NTAC recommendation	200 fecal coliforms – same as 1000 coliforms
1986	EPA recommendation	FW - 126 <i>E.coli</i> 33 enterococci
		MW 35 enterococci
		All equivalent to 200 fecal coliforms

How far have we come with water quality guidelines?

Geometric Mean

Method recommended by EPA. Based on 5 samples collected over a 30-day period. Minimizes influence of a one-time high result.

Example: Sunshine Lake with bacteria readings of 5, 10, 120, 20, 2700

Average
would be

$$= \frac{5 + 10 + 120 + 20 + 2700}{5} = 571$$

GM

$$= \sqrt[5]{5 * 10 * 120 * 20 * 2700} = 50$$