### Georgia Adopt-A-Stream

# BACTERIAL MONITORING WORKSHOP



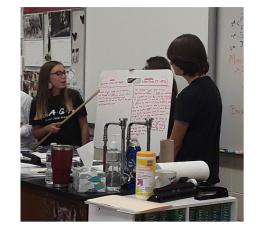


### Georgia Adopt-A-Stream

A citizen science water quality monitoring program encouraging all Georgians to get familiar with their watersheds, monitor impacts, improve streams, rivers, wetlands, lakes, and estuaries, and inform others about their effect on water quality.



## Awareness



Increase public

awareness of nonpoint
source pollution & water
quality issues

#### D Data



Collect baseline water quality <u>data</u> according to Adopt-A-Stream protocols

## Observations



Take <u>observations</u> of sites to note water quality conditions

## Partnerships



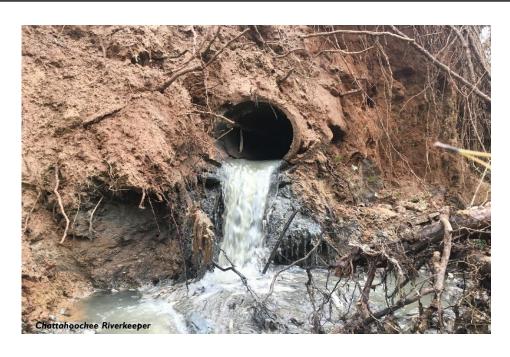
Seek <u>partnerships</u> with local gov'ts, nonprofits, & other organizations to share results & resources

## Tools & Training



Utilize tools & training provided by staff & local coordinators

#### TYPES OF POLLUTION



#### **POINT SOURCE POLLUTION**

- Easily identifiable pollutant source
- Regulated by GA EPD through NPDES permitting process



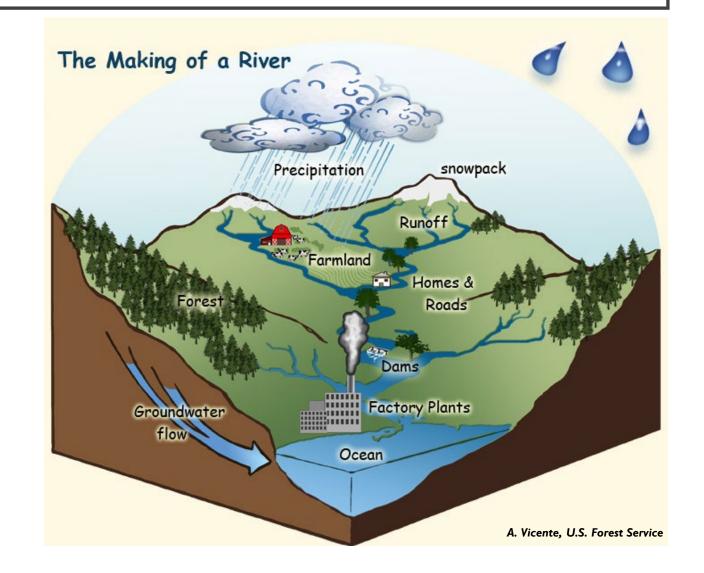
#### NONPOINT SOURCE POLLUTION

- Sources not easily distinguished/identified
- Everyone contributes
- Main cause of water quality problems in GA

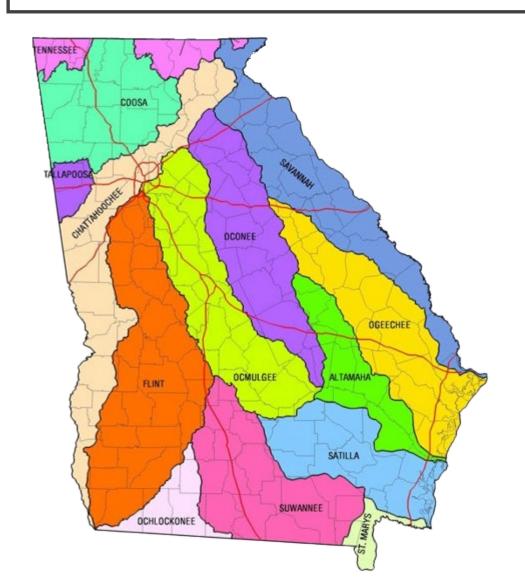
#### WHAT IS A WATERSHED?

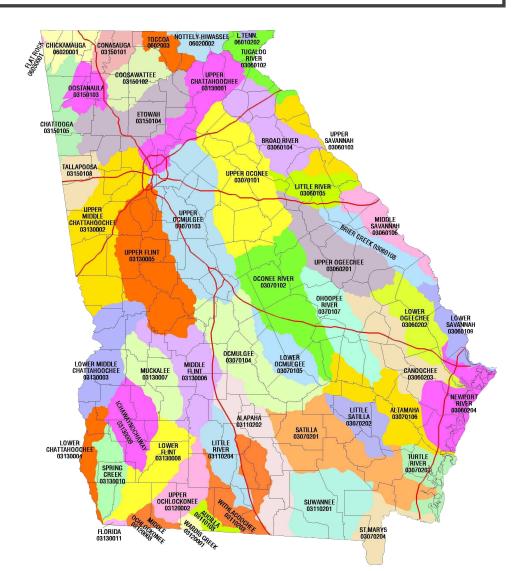
- A land area from which water, sediment, and dissolved materials drain to a common point along a stream, wetland, lake, or river.
- Its boundaries are defined by the highest points of land around the waterbody.

There is an unbreakable link between human health and wellbeing and ecosystems. -Walter Reid

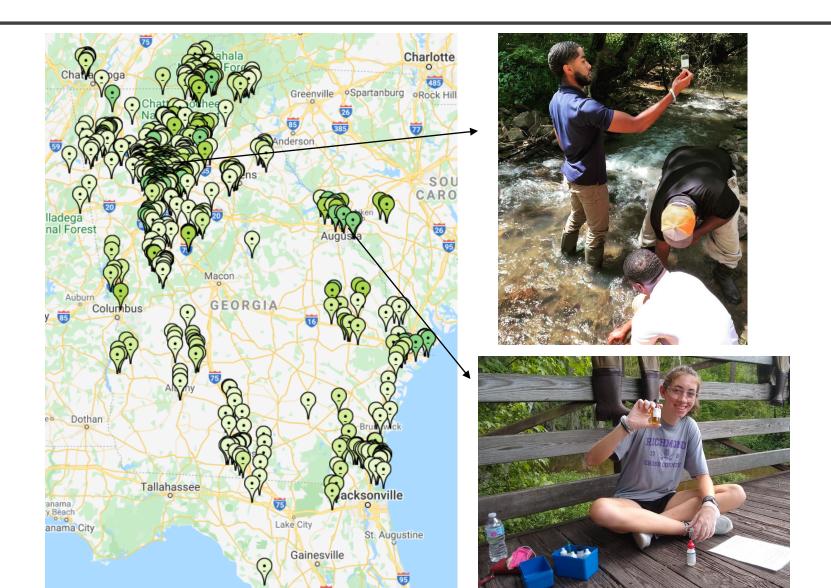


#### WHERE IS YOUR WATERSHED?





#### **VOLUNTEER NETWORK AND SUPPORT**



#### **VOLUNTEER NETWORK AND SUPPORT**

State Staff 2 to 4

Board Members 20

Local Coordinators 70

Volunteers 3,000 +

#### AAS VOLUNTEERS USE STANDARDIZED PROTOCOLS

- EPA Approved Quality Assurance Project Plan (QAPP)
- Quality Assurance/Quality Control (QA/QC)
  - Required to attend workshop(s) and pass certification test(s) to become certified
  - Only individuals are certified
  - Set monitoring protocol ensures all volunteers are collecting baseline data using standard methods
  - Only certified volunteers can <u>enter</u> data, but anyone can access the 20+ years of data in the online AAS database



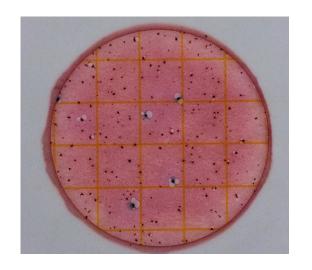
#### EARNING YOUR QA/QC BACTERIAL CERTIFICATION





FIELD & LAB:

Volunteers must demonstrate how to properly collect and plate a sample





#### **WRITTENTEST:**

Volunteers must pass a written evaluation with a score of at least 80% and must correctly identify E. coli colonies and calculate E. coli levels of example plates with accuracy of at least 90%

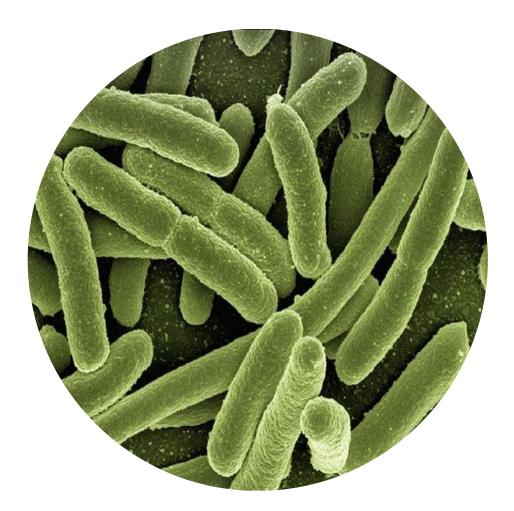
#### WHY IS BACTERIAL MONITORING IMPORTANT?

- Monitor bacterial contamination of surface waters to assess if pathogens are present
  - Human health is at risk when in contact with waters that contain harmful bacteria
  - Gaining a snapshot of surface water contamination (E. coli colonies), not long-term trends

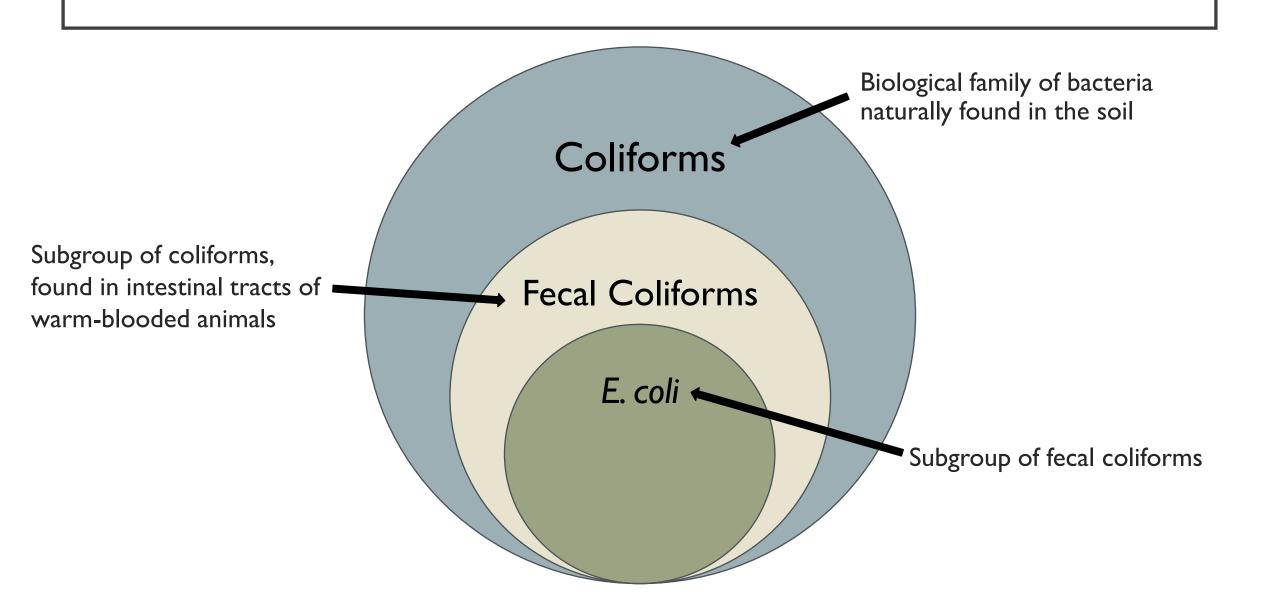


#### ABOUT BACTERIA

- Single-celled, living microscopic organisms
- Pros:
  - Decomposition
  - Digestion
  - Nutrient Cycling
  - Pollution Control
- Risks:
  - Release Toxins
  - Cause Disease (Pathogens)



#### WHAT IS E. COLI?



#### WHY MONITOR FOR E. COLI?

- High levels indicate the possible presence of pathogens
- Sources of *E. coli* in waterways:
  - Wildlife
  - Livestock
  - Urban storm runoff
  - Sewage
    - Leaking sewage pipes
    - Combined sewer overflow
    - Wastewater treatment plants
    - Failing septic systems



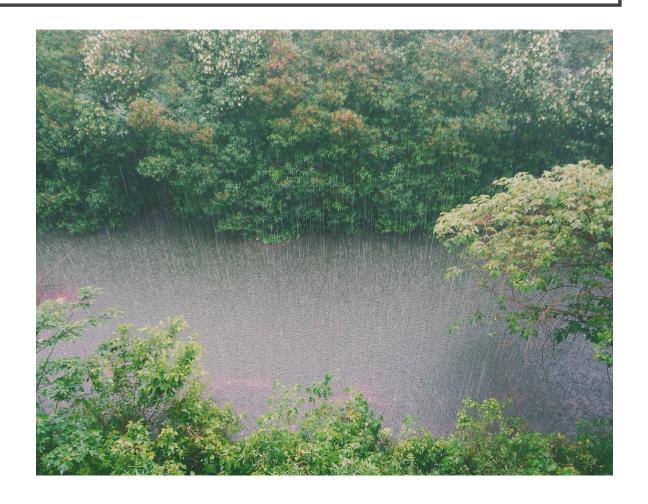
#### WHERE, WHEN, AND HOW OFTEN?

- Where to monitor:
  - Well mixed, flowing area of water
  - Same site location
- When to monitor:
  - Normal flow conditions
  - Same time of day
- How often to monitor:
  - Once a month



#### FACTORS INFLUENCING E. COLI COUNTS

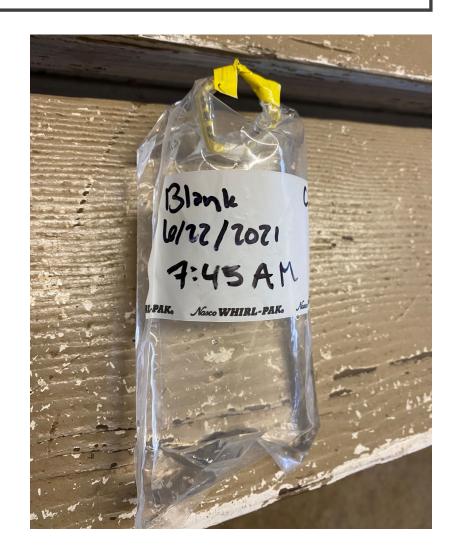
- Weather:
  - Higher levels following a rainstorm or heavy runoff event
  - Avoid sampling during high flow
- Season/Temperature:
  - Warmer water temperature = higher *E. coli* replication rates
  - Colder water temperature = lower E. coli replication rates



#### HOW TO MONITOR FOR E. COLI

#### STEP I: PREPARE THE BLANK/CONTROL

- Proves no contamination occurred during sample collection, transport, or plating
- How to prepare a blank:
  - Label Whirl-pak® bag as a blank
  - Put gloves on and remove perforated seal
  - Use small white tabs to open the bag
  - Fill the bag 2/3 up with DISTILLED WATER
  - Whirl!
  - Place in sanitized cooler with ice



#### STEP 2: COLLECTING A SAMPLE

- Label a Whirl-pak® bag with the location, date, and time
- Use same procedure as blank to open
- Fill Whirl-pak® 2/3 with stream water (collected upstream) and whirl!
- Place sample in cooler immediately after collection
  - Ice prevents E. coli from replicating
  - Lid prevents UV rays killing existing bacteria
- Plate within 24 hrs of collection



#### STEP 3: PLATING YOUR SAMPLE

- Clean area with disinfectant
- Invert Whirl-pak® to mix sample
- Label 3M Petrifilm plates
  - Date and time
  - Site name
  - Blank, Plate 1, Plate 2, Plate 3
- Use pipette to take one 1 mL sample from the blank, plus three 1 mL samples from the stream water (4 plates total)





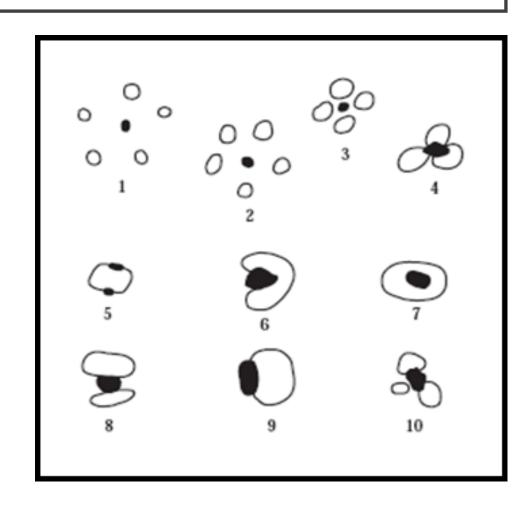
#### STEP 4: INCUBATING YOUR SAMPLE

- 35°C ± 1°C for 24 hours ± 1 hour
- Check minimum and maximum temperatures after incubating
- Use AAS or EPA approved incubator



#### STEP 5: READING YOUR RESULTS

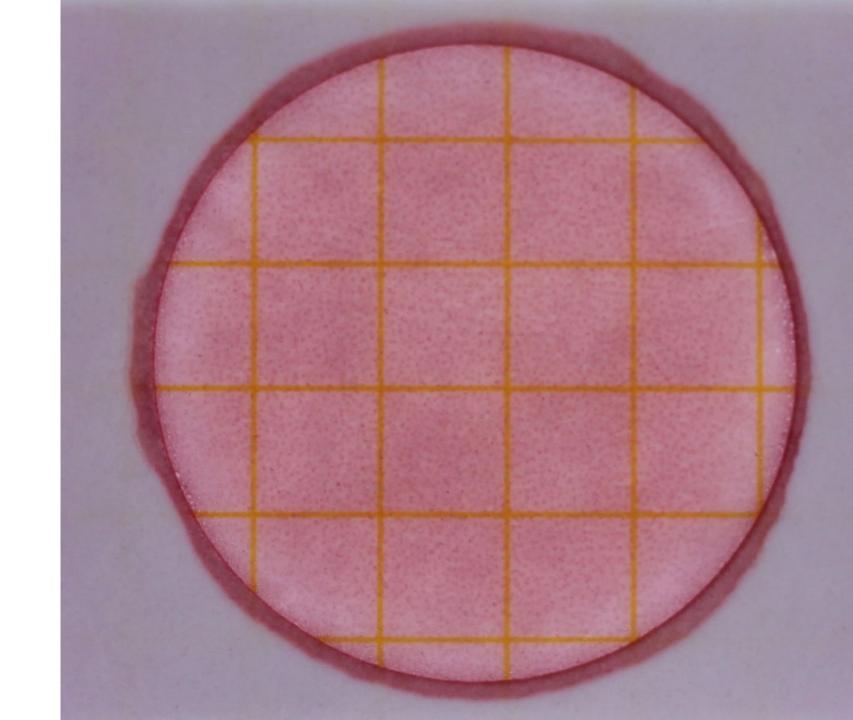
- Only count blue colonies with gas bubbles
- Do not count colonies growing more than halfway off the medium
- Units for bacteria: Colony Forming Units (CFU)/100 mL



Possible gas bubble patterns

### BLANK/ CONTROL

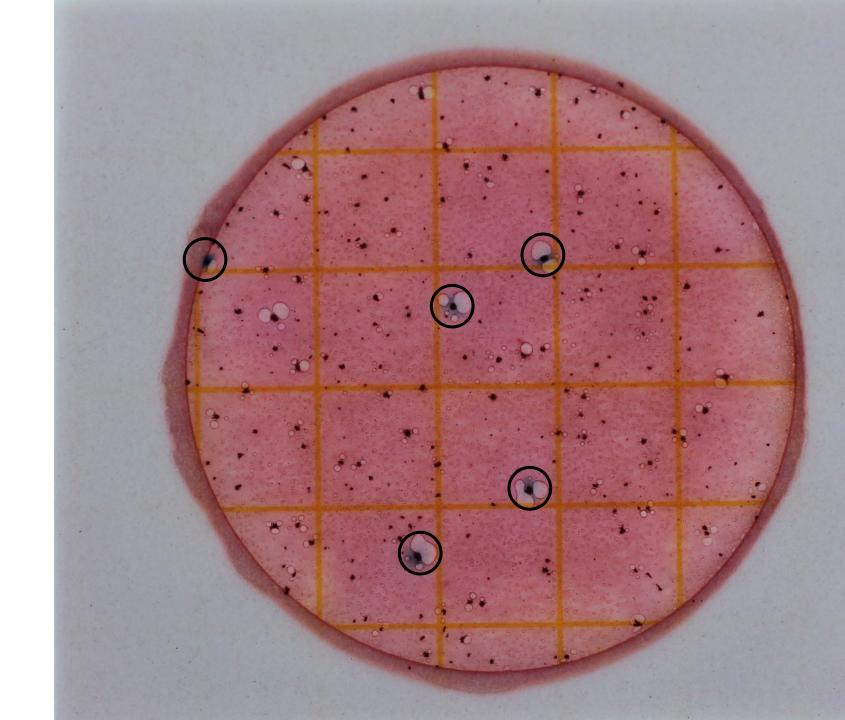
There should not be any colonies (E. coli or general coliform) on the blank!



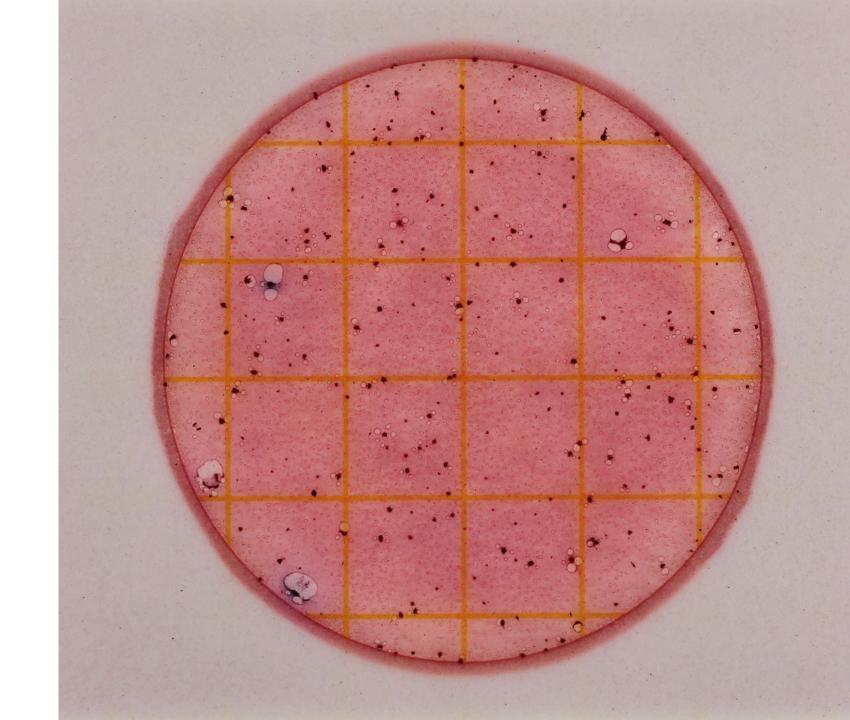
EXAMPLE #1



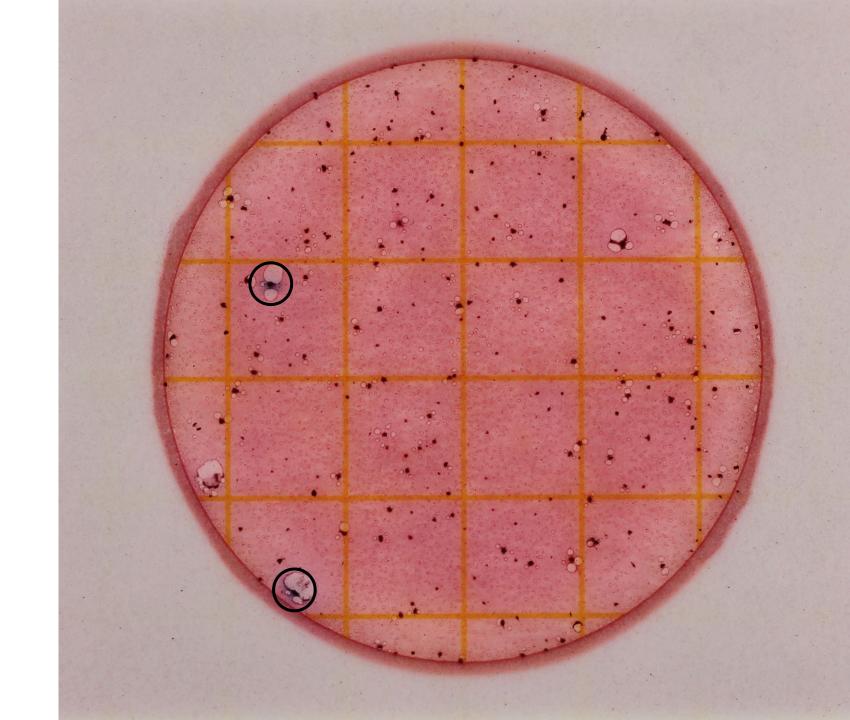
### EXAMPLE #1 ANSWER



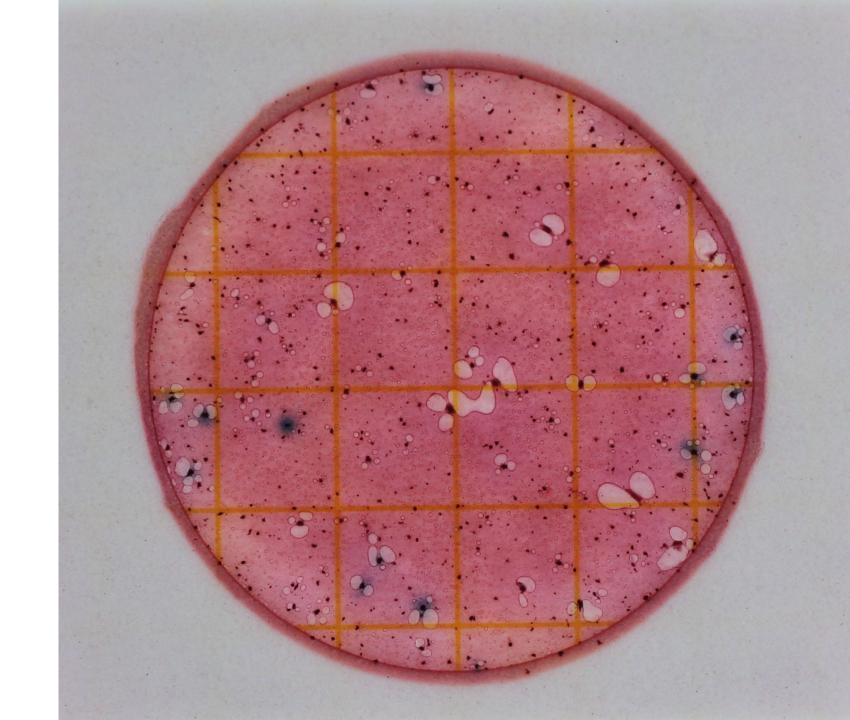
# EXAMPLE #2



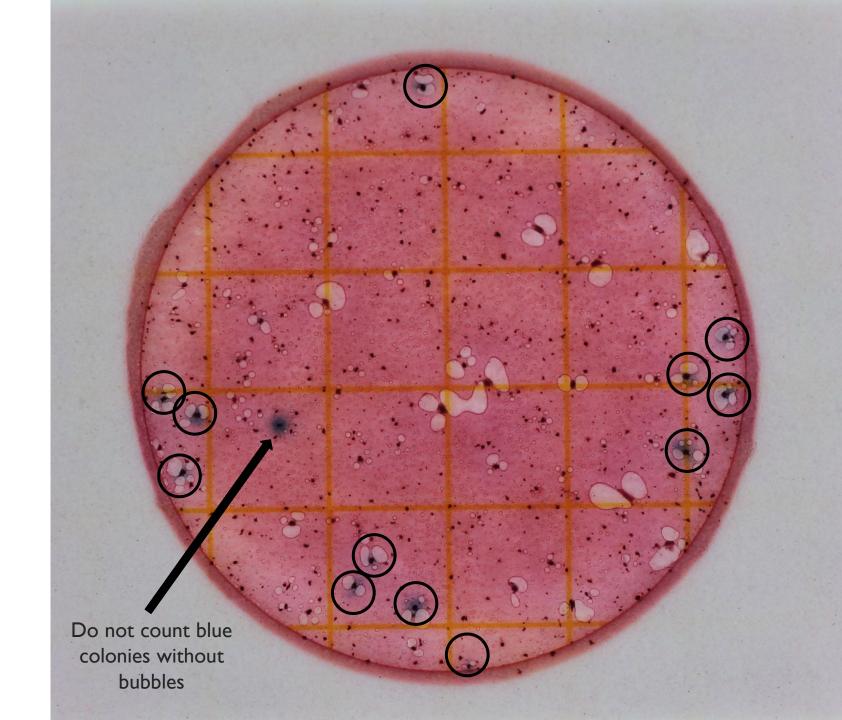
### EXAMPLE #2 ANSWER



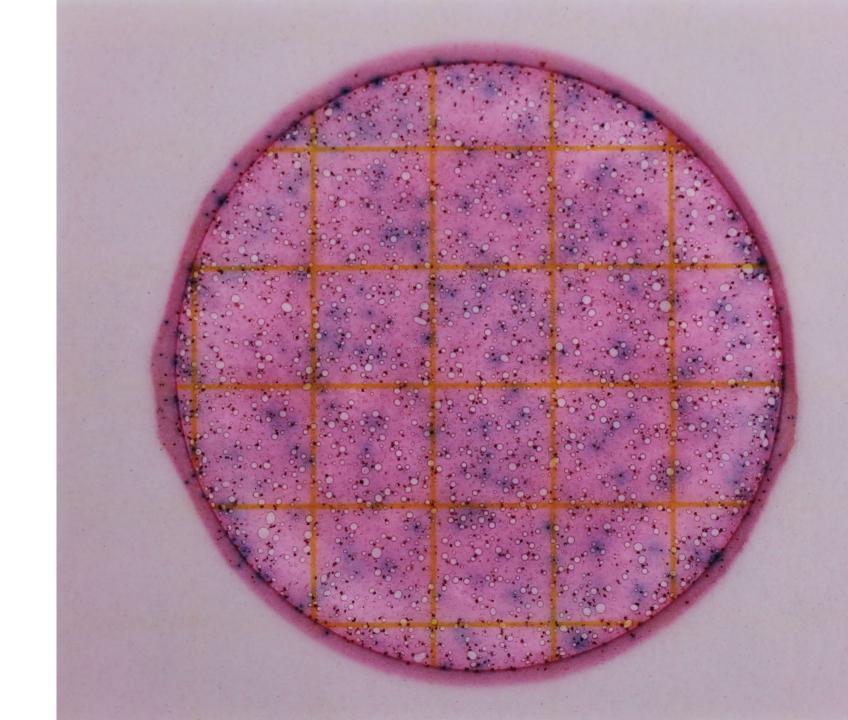
# EXAMPLE #3



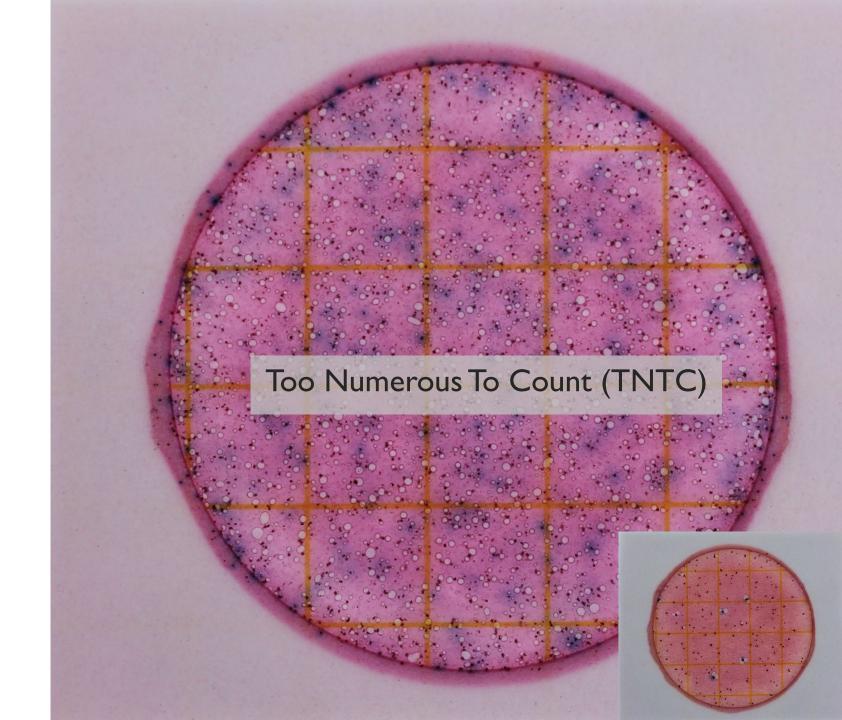
### EXAMPLE #3 ANSWER



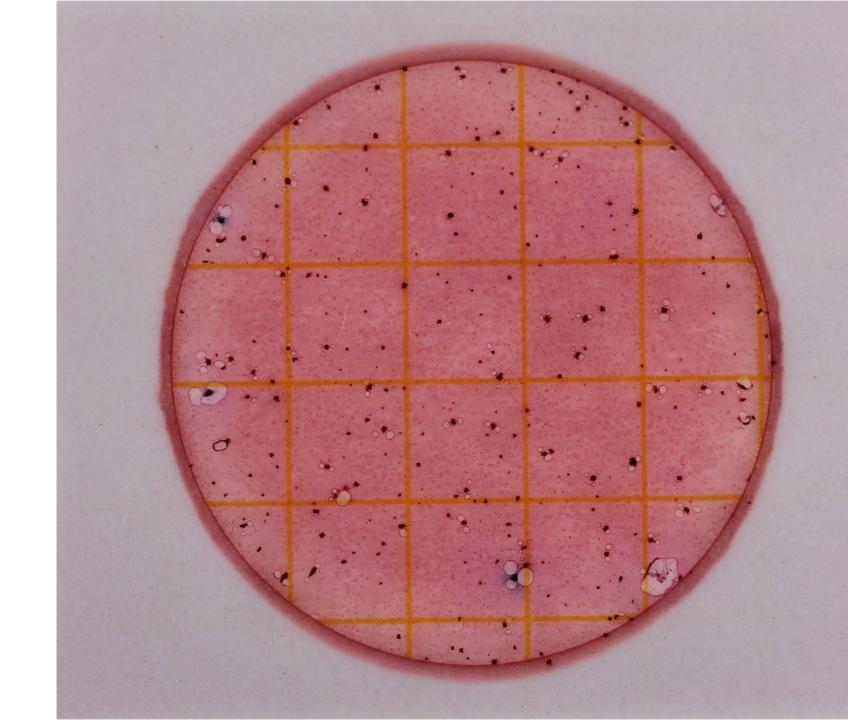
### EXAMPLE #4



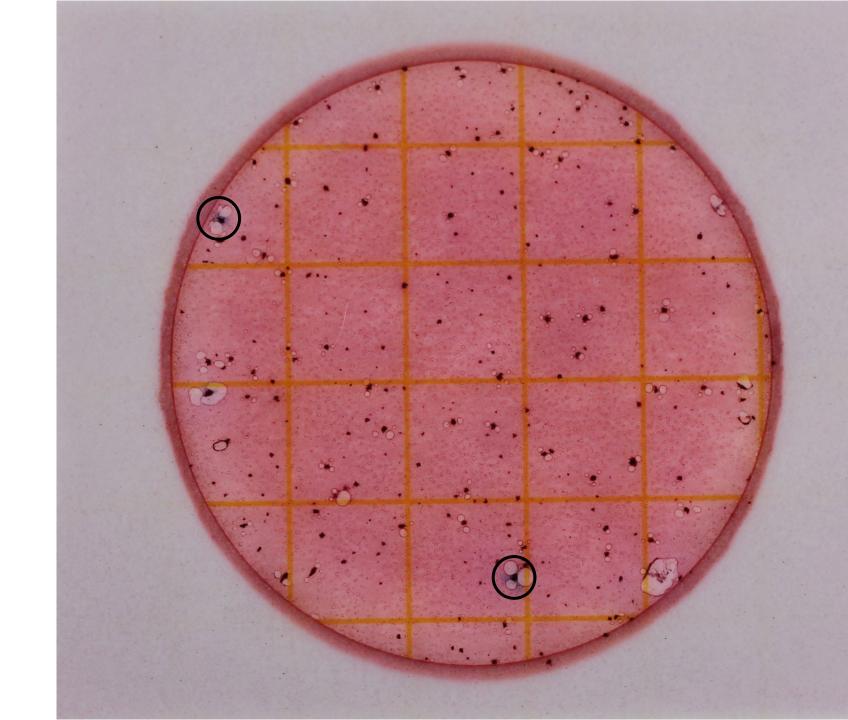
### EXAMPLE #4 ANSWER



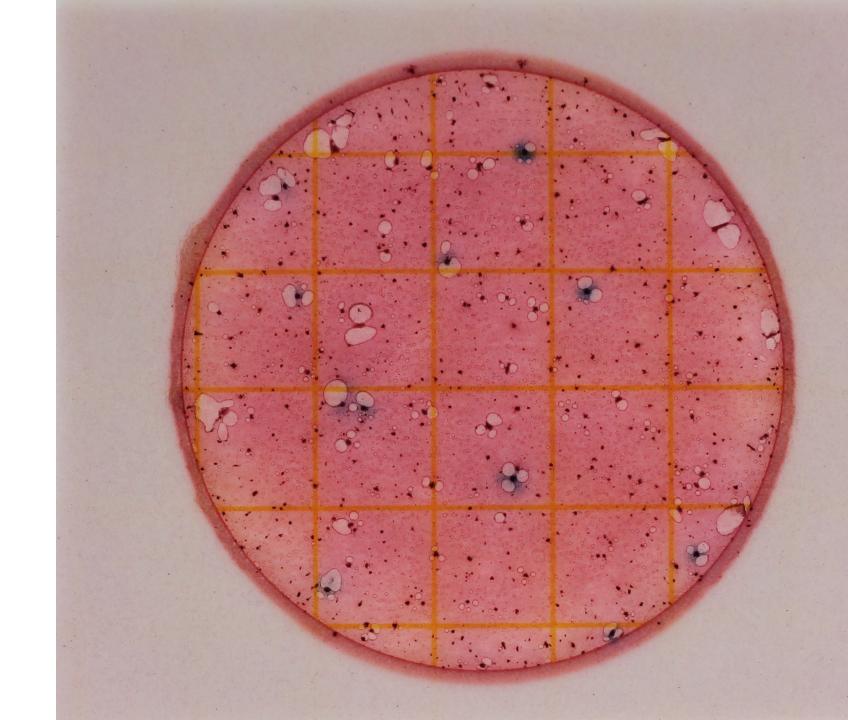
# EXAMPLE #5



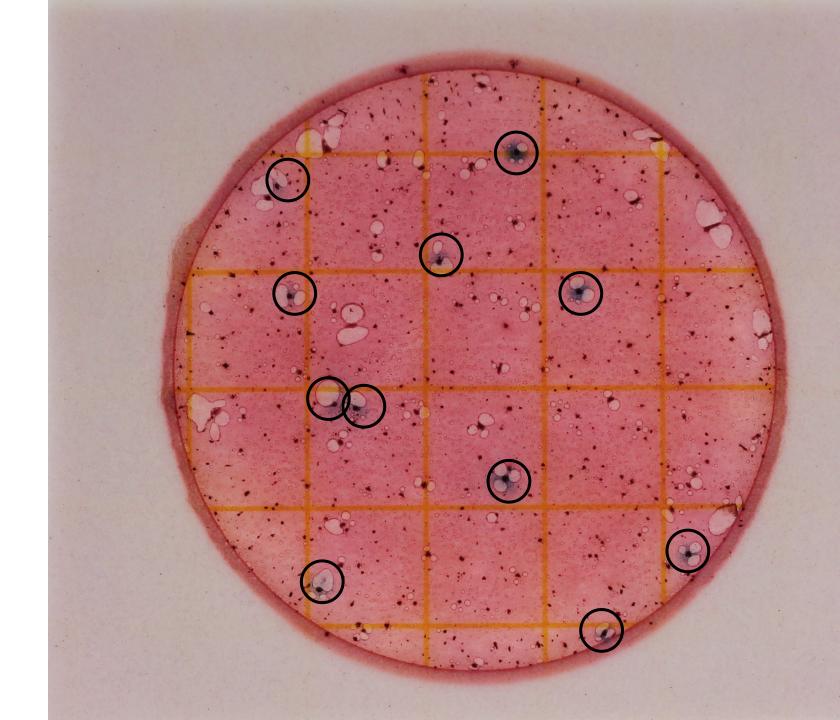
### EXAMPLE #5 ANSWER



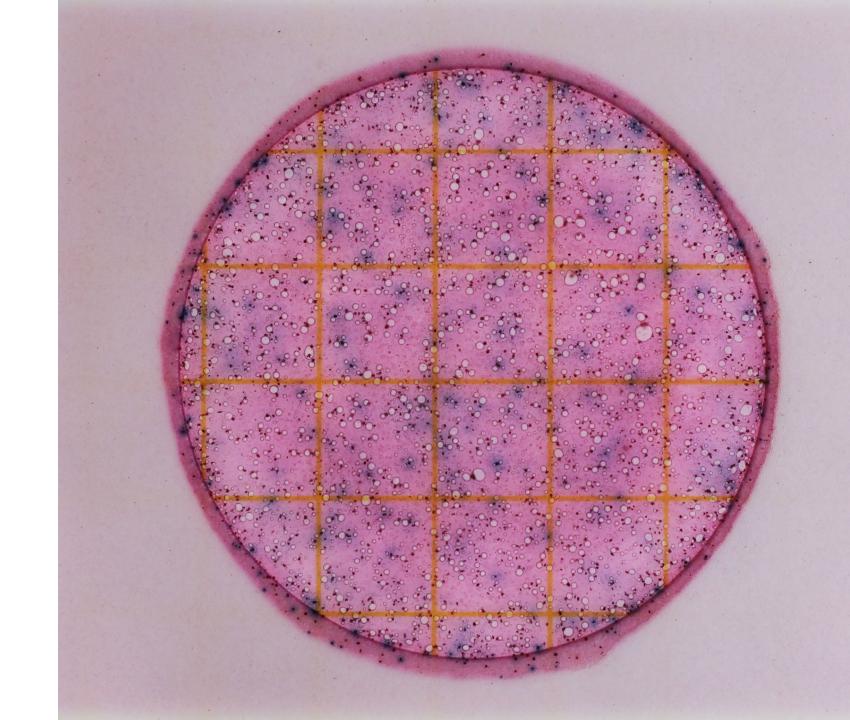
### EXAMPLE #6



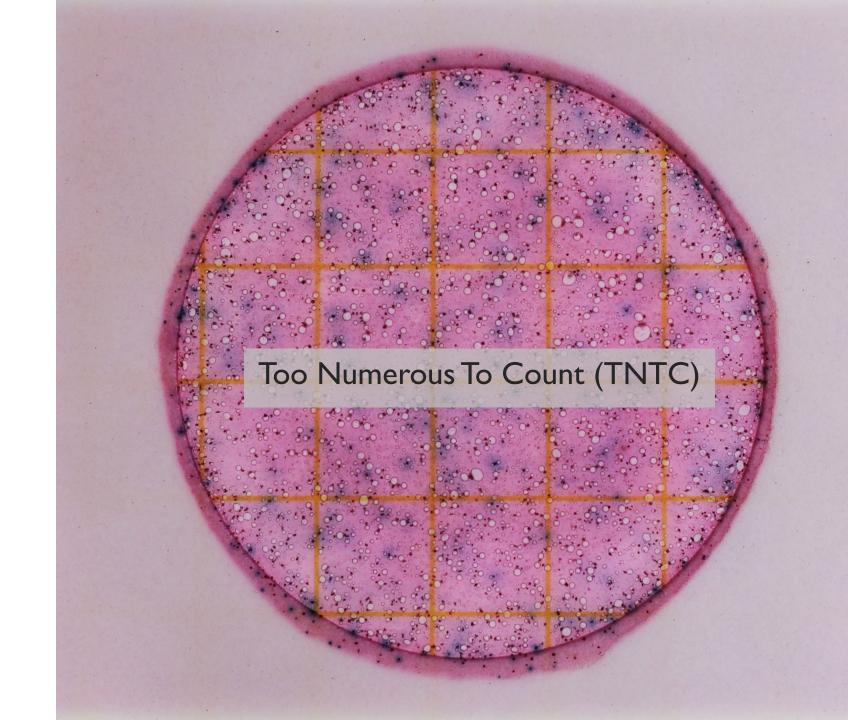
### EXAMPLE #6 ANSWER



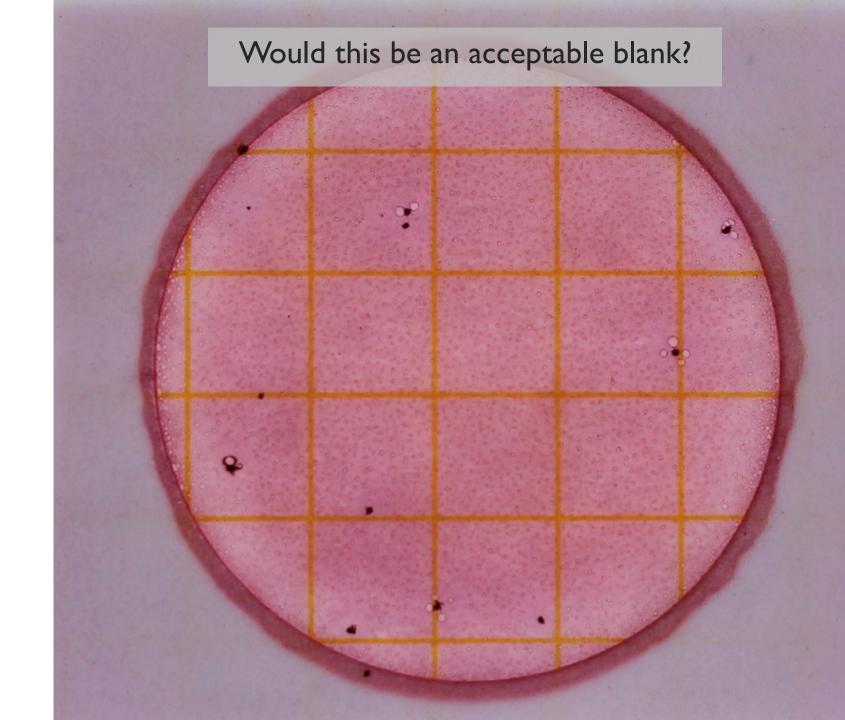
EXAMPLE #7



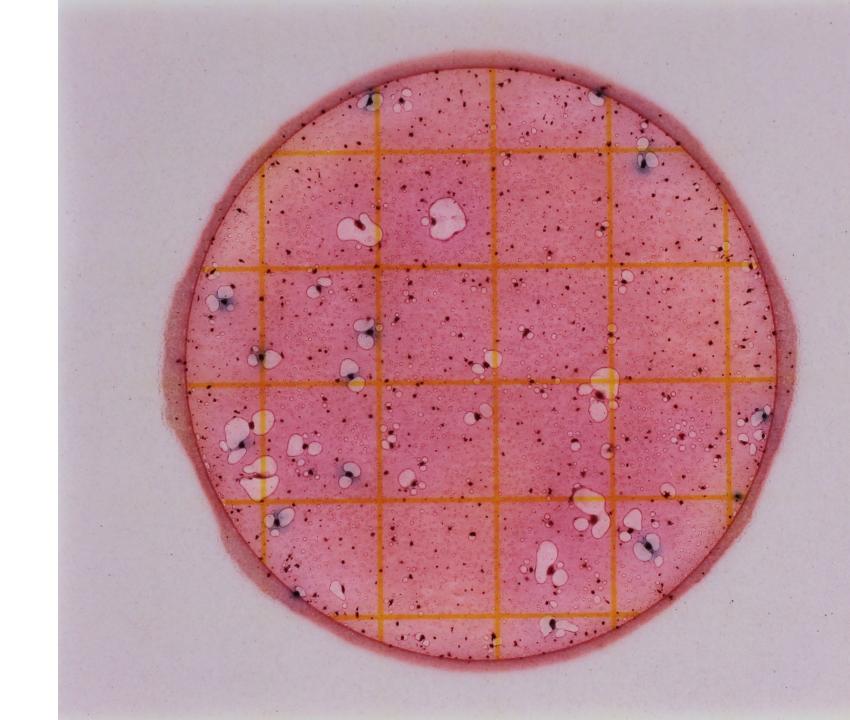
### EXAMPLE #7 ANSWER



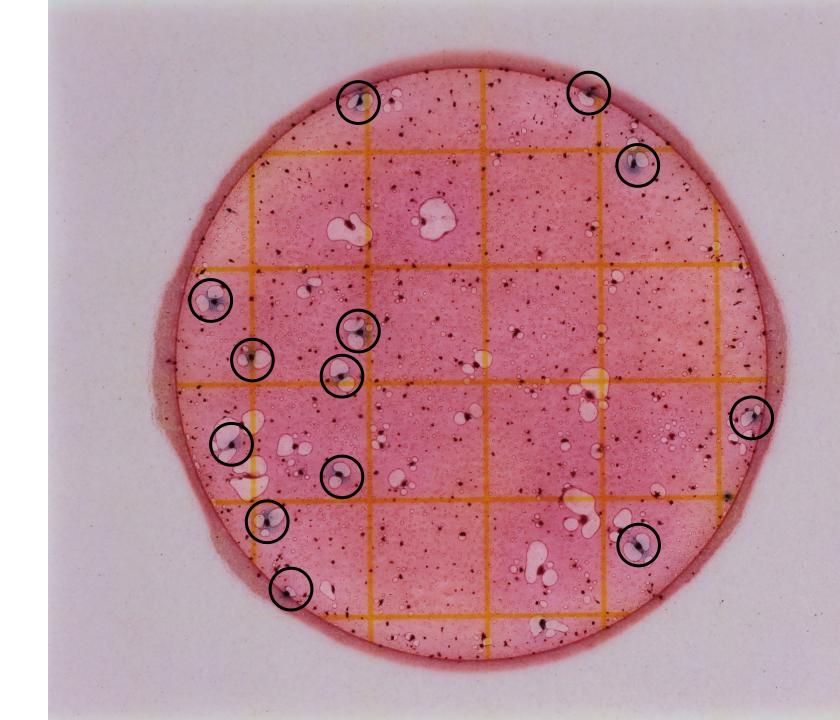
EXAMPLE #8



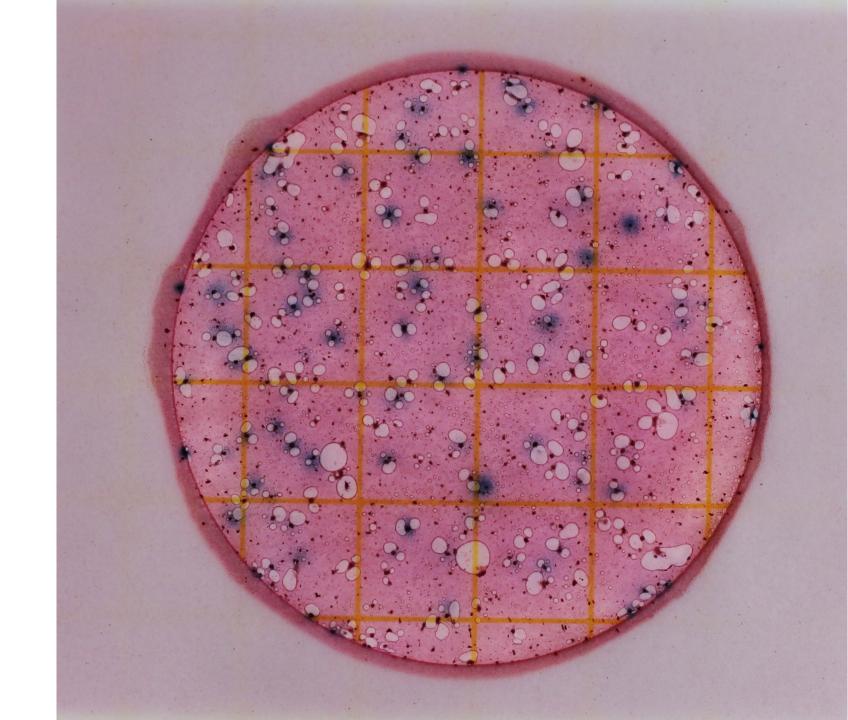
# EXAMPLE #9



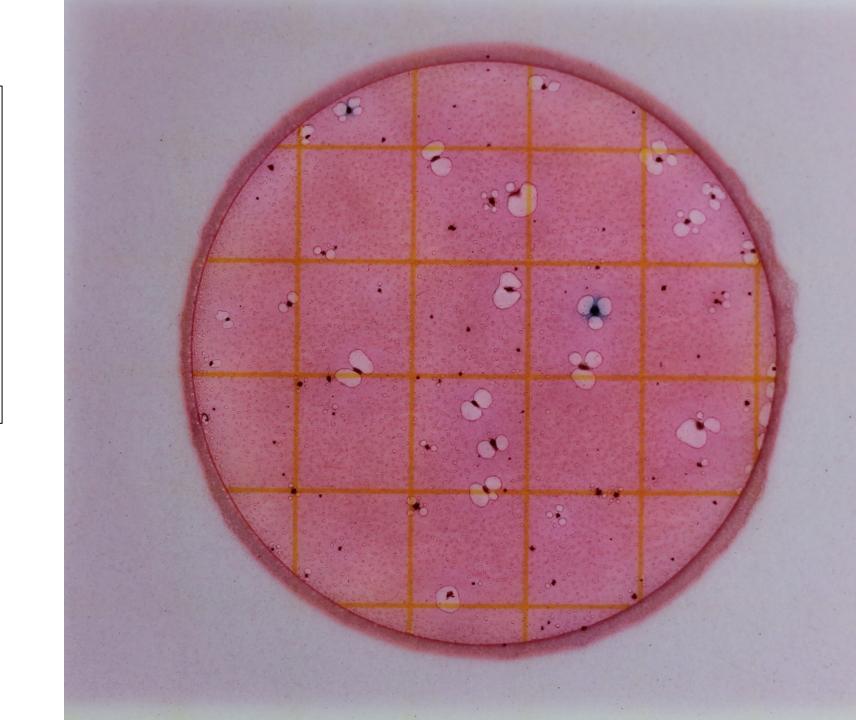
# EXAMPLE #9 ANSWER



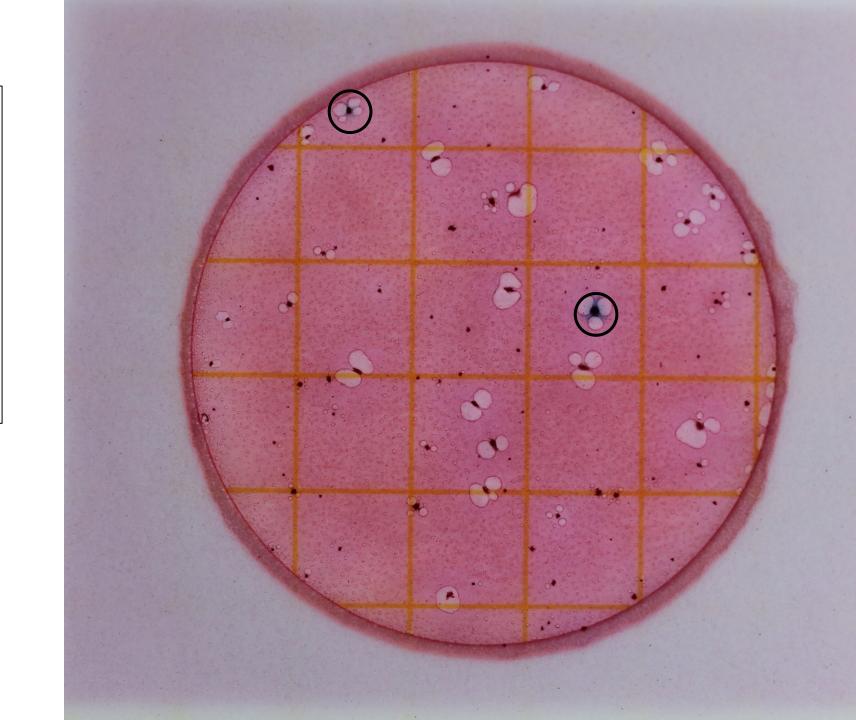
# EXAMPLE #10



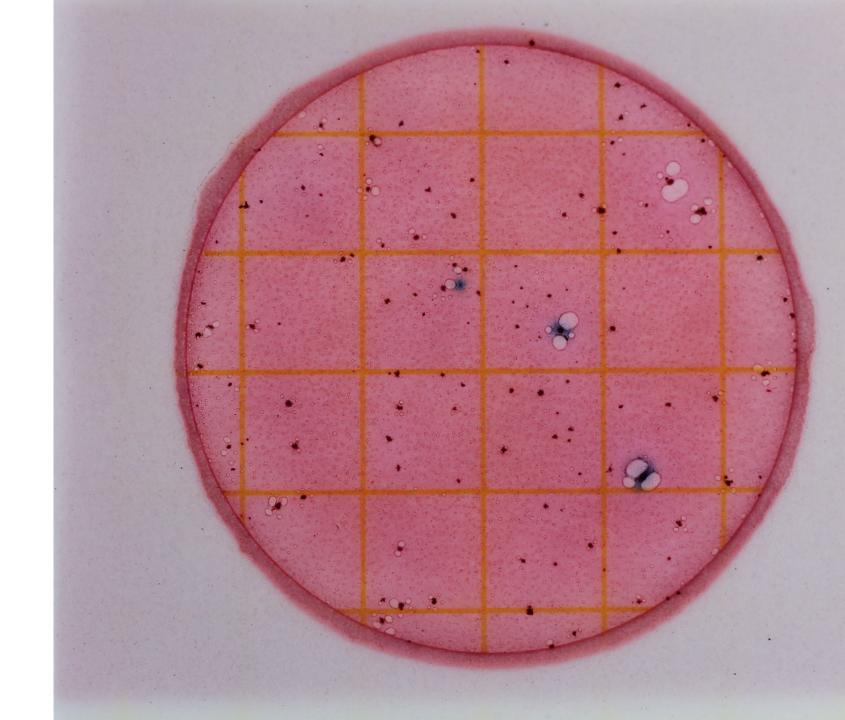
# EXAMPLE #11



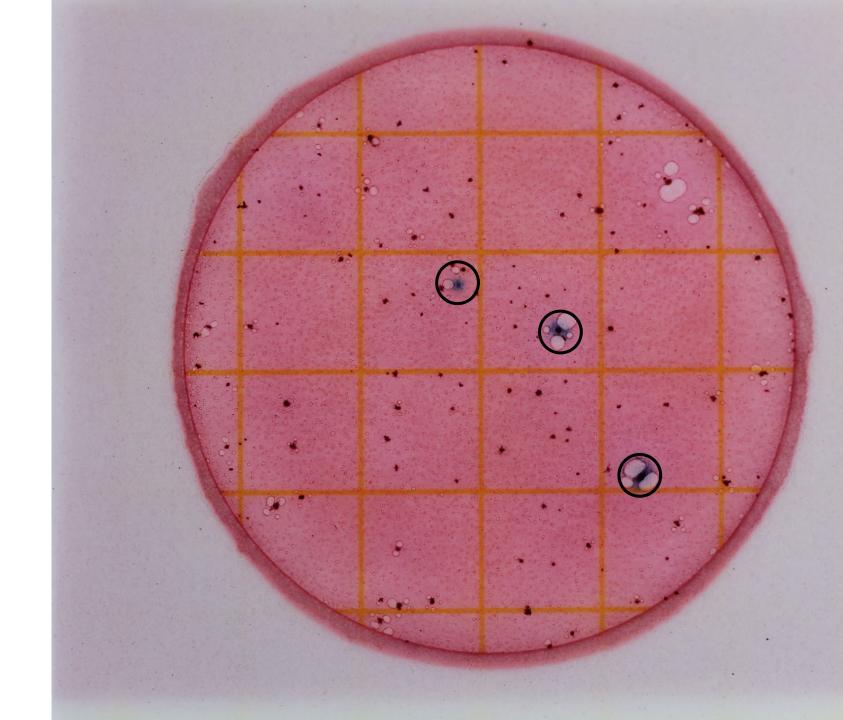
# EXAMPLE #11 ANSWER

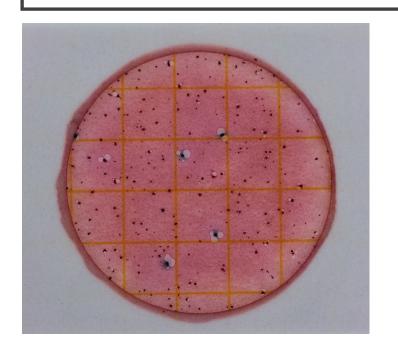


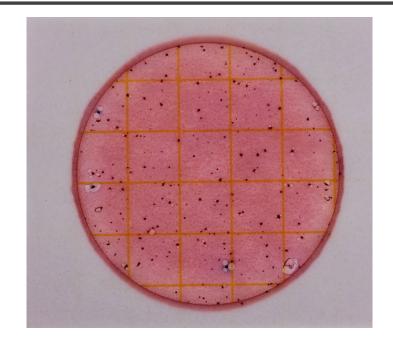
# EXAMPLE #12



# EXAMPLE #12 ANSWER







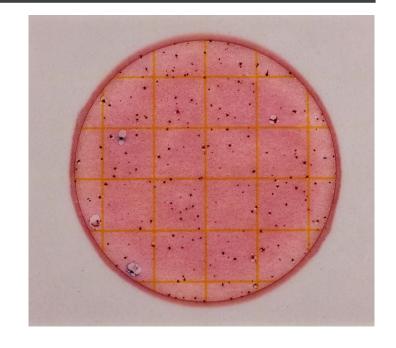
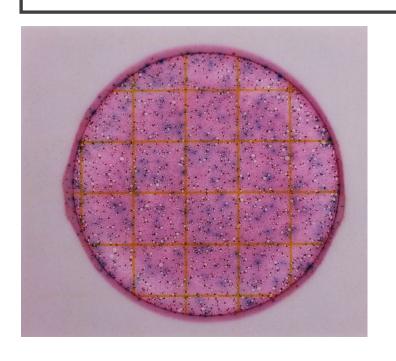


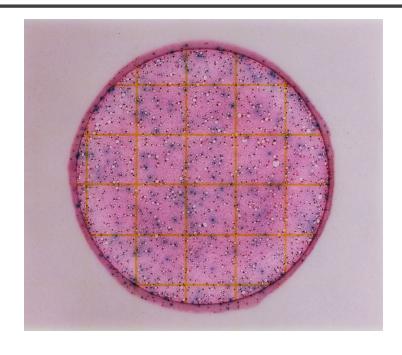
Plate I Plate 2 Plate 3

	Plate I	Plate 2	Plate 3
E. coli colonies	5	2	2

	Plate I	Plate 2	Plate 3
E. coli colonies	5	2	2

Step 1: 
$$\frac{(5 + 2 + 2)}{3} = 3.00 \text{ CFU/I mL}$$
Step 2: 
$$3.00 \text{ CFU/I mL} * 100 = 300 \text{ CFU/I00 mL}$$





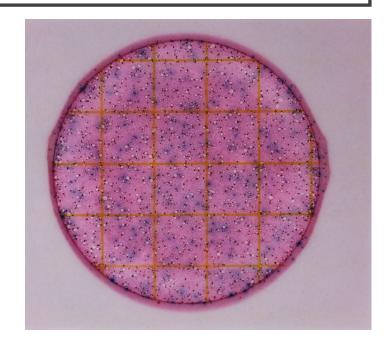


Plate I Plate 2 Plate 3

	Plate I	Plate 2	Plate 3
E. coli colonies	TNTC	TNTC	TNTC

	Plate I	Plate 2	Plate 3
E. coli colonies	TNTC	TNTC	TNTC

$$\frac{(\mathsf{TNTC} + \mathsf{TNTC} + \mathsf{TNTC})}{3} = \mathsf{TNTC}$$

### STEP 7: DISPOSAL AND CLEAN-UP

- Spray plates with disinfectant, seal in bag/used Whirl-Pak, and throw away
- Wipe down incubator & surrounding surfaces with disinfectant
- Wash hands!



# HOW TO STORE PETRIFILM

- If using within one month, keep in the fridge
- If not, store in the freezer and thaw before use





#### REGULATORY STANDARDS

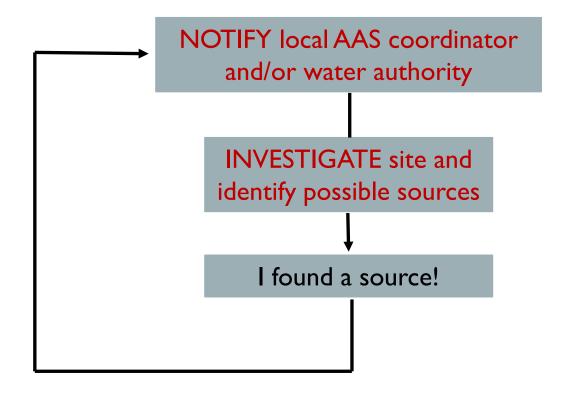
- State of GA regulatory data transitioned from fecal coliforms to E. coli
  - State standards reflect established EPA guidelines
  - Includes enterococci to indicate presence of pathogens in coastal waters
- Statistical Threshold Value (STV) <10% of samples should exceed given level</li>
  - Risk level of 36/1,000 people getting sick from primary contact activities
  - Sample again if sample > STV
  - Report if samples are consistently > STV (2+ months out of the year)

	E. coli STV	Enterococci STV
EPA recommended level (CFU/100 mL)	<410	<130

### HIGH E. COLI COUNTS

#### AAS Action Value: >1000 CFU/100 mL

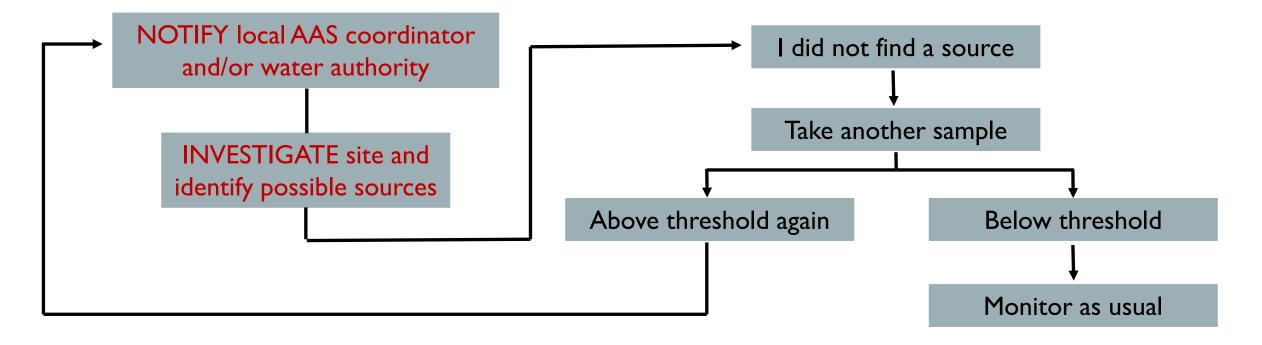
If your count exceeds this value and you did find a source:



### HIGH E. COLI COUNTS

#### AAS Action Value: > 1000 CFU/100 mL

If your count exceeds this value and you did not find a source:



#### SAFETY

- Try not to sample alone- take a monitoring buddy!
- Do not sample during high flows or after a heavy rain event
- Obtain permission if sampling on private property
- Wear PPE when sampling and plating
- Disinfect thoroughly before and after plating and counting



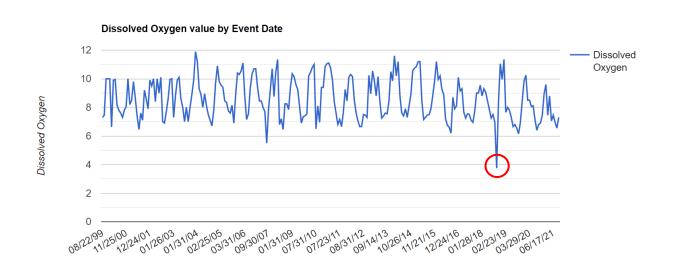
# ONCE YOU'RE CERTIFIED

- You get an account to our online database!
- Only certified volunteers can submit data
- Certification is valid for one year
- Volunteers must attend an annual recertification workshop



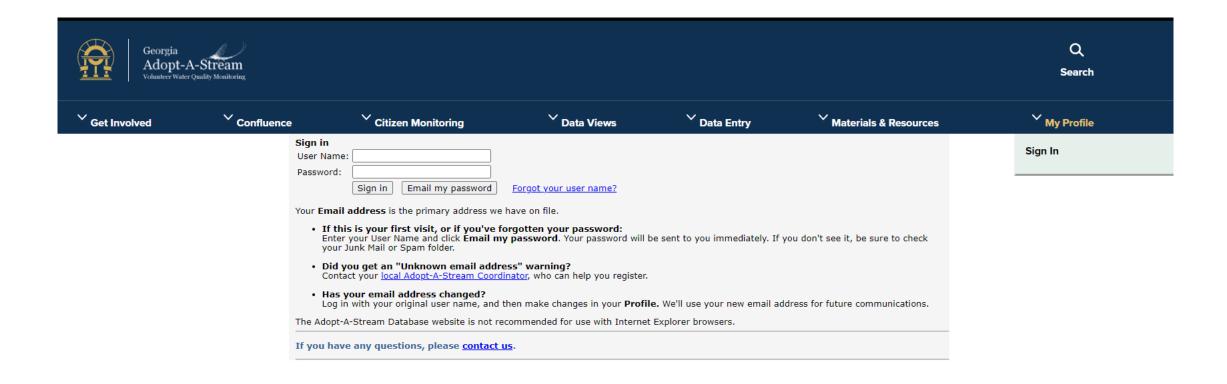
### HOW ARE YOUR DATA USED?

- Establish baseline conditions for waterbodies across the state
- Discover and report water quality issues
- Educate your community
- Help inform status of streams for 303d/305b list



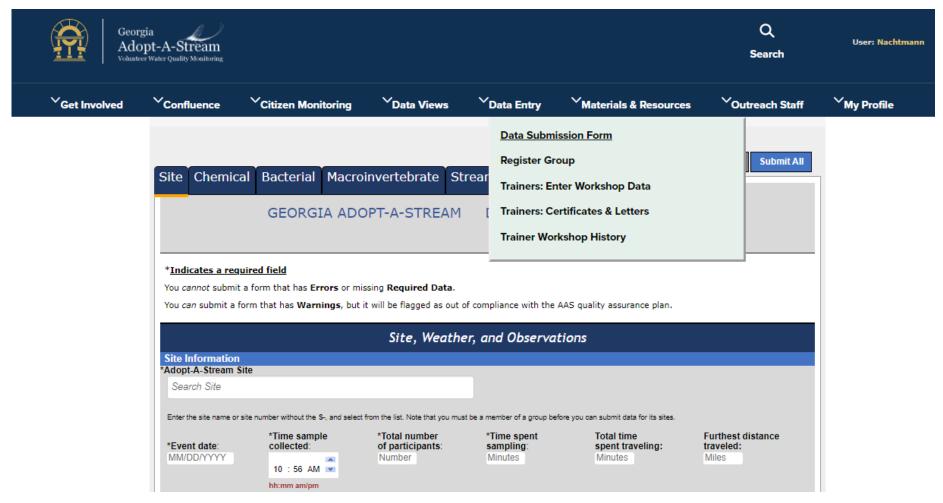


#### DATABASE LOGIN



From the AAS website's homepage, hover over the My Profile tab and click Sign In

#### DATA SUBMISSION FORM



From the AAS website's homepage, hover over the Data Entry tab and click Data Submission Form

# SITE, WEATHER, AND OBSERVATIONS

#### **GEORGIA ADOPT-A-STREAM: Bacterial Form**

To be conducted every month

NO.	Group Name:	Event Date:		(MMDDYYYY)	
SITE INFORMATION	Group ID: G Site ID: S	Time Sample Collected:		(HHMM am/pm)	
	Stream Name:	Time Spent Sampling:		_ (Min)	
Ž	Monitor(s):	Total Time S	Spent Traveling (optional):	(Min)	
SITE	Number of Participants:	Furthest Dis	tance Traveled (optional):	(Miles)	
WEATHER	Present conditions (check all that apply)  Heavy Rain Steady Rain Interm Overcast Partly Cloudy Clear/S		Amount of rain, if know Amount in Inches : In Last Hours/Days: *Refer to wunderg		
	Flow/Water Level: Dry Stagnant/Still Low Normal High Flow (over banks)				
	Water Clarity: Clear/Transparent Cloudy/Somewhat Turbid Dpaque/Turbid				
S	Water Color: No Color Brown/Muddy Green Milky/White Tannic Other:				
2	Water Surface: ☐ Clear ☐ Oily Sheen: does it break when disturbed? Yes/No (circle one) ☐ Algae ☐ Foam ☐ Greater than 3" high ☐ It is white				
×					
OBSERVATIONS	Water Odor: Natural/None Gasolin	ne 🗌	Sewage	Rotten Egg	
OB	☐ Fishy ☐ Chlorin	e	Other:	_	
	Photos: Please take images to document your observations and changes in water quality conditions.				
	Photo point directions can be found in the manuals. Send photos to AAS@gaepd.org.				
	Trash: ☐ None ☐ Yes, I did a cleanup ☐ This site needs an organized cleanup				

# BACTERIAL DATA

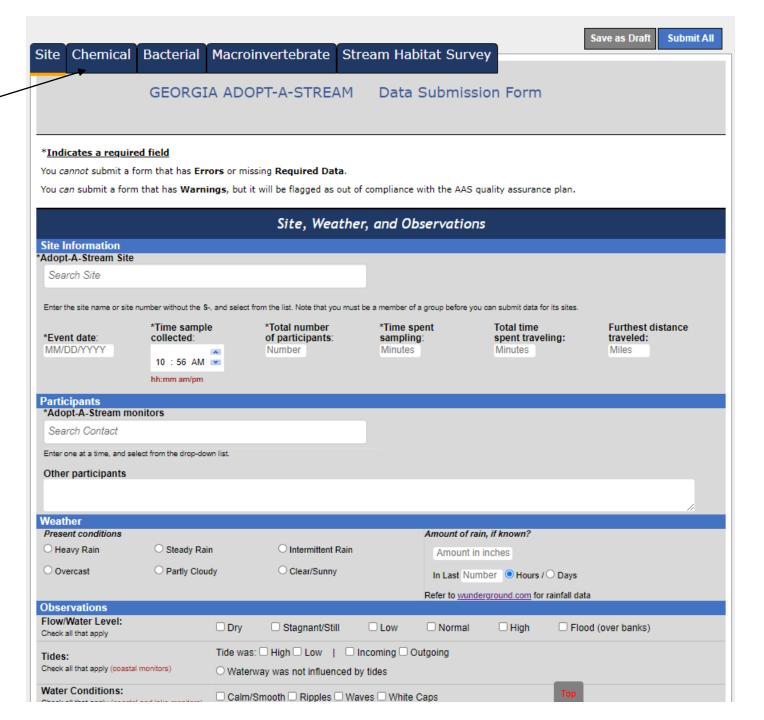
	3M Petrifilm Method: Escherichia coli				
	Plate	Colonies	re, plus one (1) blank plate. Process within 6-24hrs, incubate at 35°C ±1° and reals  Find AVG of Number of Colonies		
BACTERIAL	Blank		(total # colonies/total # of plates (do not include blank)	cfu/100mL	
ĒR	1		( / ) x 100 =		
\C	2	Sa	ample Holding Time (HH):		
B/	3	Da	ate START(MMDDYYYY): Date END (M	Date END (MMDDYYYY):	
	Total # Colonies	Ti	me START (HHMM): Time END (I	Time END (HHMM):	
		M	IN Temp ( <sup>0</sup> C): MAX Temp (	MAX Temp ( <sup>0</sup> C):	
	Any changes since you last sampled at this site? If yes, please describe.				
TS					
COMMENTS					
M					
ဗ					

Please submit data to our online database at AdoptAStream.Georgia.gov

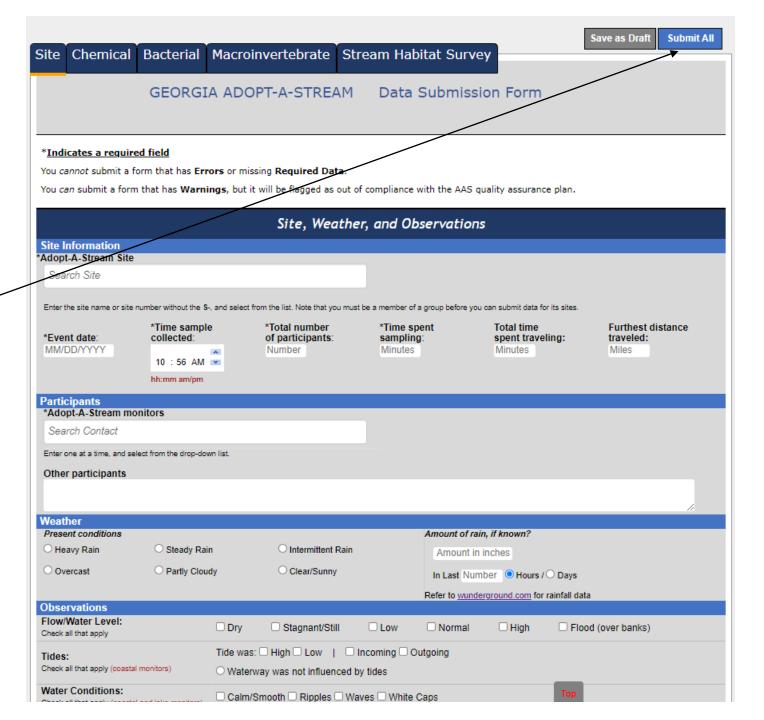
#### Submit data ASAP to online database

Access database via AdoptAStream.Georgia.gov

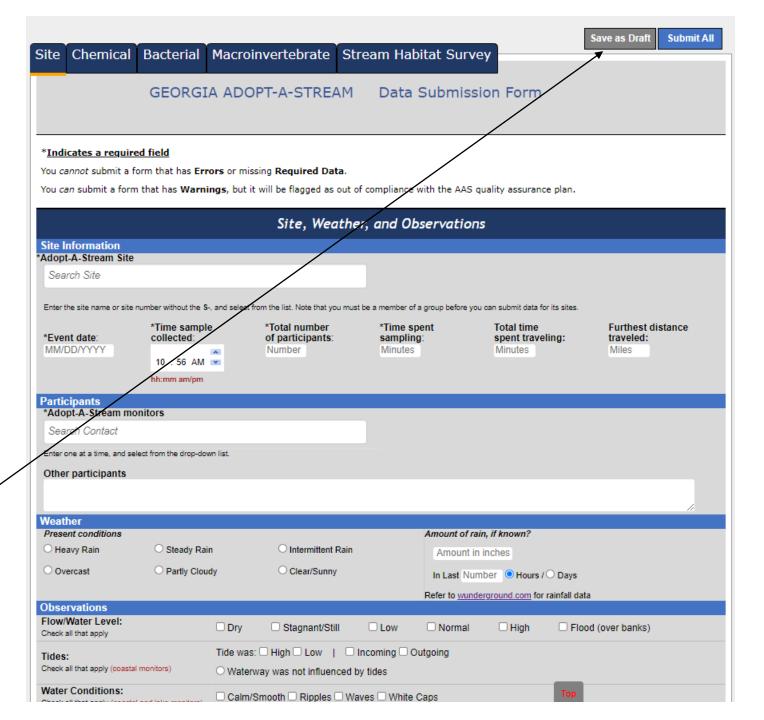
Fill out site data first, then navigate to the chemical tab to continue entering data



After entering all of your data, click "Submit All" to submit your data to the database



Use "Save as Draft" to finish submitting data at a later time. Data must be submitted within 7 days of saving as a draft.



#### FOLLOW AAS AND STAY CONNECTED

- AAS@dnr.ga.gov
- AdoptAStream.Georgia.gov
- facebook.com/georgiaadoptastream
- @georgiaadoptastream
- 2 Martin Luther King Jr. Drive Suite 1462, East Tower Atlanta, Georgia 30334
- 470-938-3341 and 470-524-5791

# **TEST REVIEW**