

Developing collaborative lab experiments across disciplines through the identification of bacteria

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Outline of today's session

- Development of our collaborative lab activity
- Review the background that students learn before being given the assignment.
- Work through the lab experiment
- Discussion:
 - Student impressions of the lab.
 - What you liked / think can be improved.
 - How you can begin to design a collaborative lesson.
 - Tips on writing an teaching grant.

SJU offers four non-science major, lab-based courses

<p>Bio 165: Exploring the Living World</p> <ul style="list-style-type: none"> - Cells - Genetics - Evolution & Biodiversity - Physiology - Ecology 	<p>Env Sci 106: Exploring the Earth</p> <ul style="list-style-type: none"> - Global Climate Change - Evolution & Ecology - Natural Resources (air, water, soil) - Sustainability - Energy
<p>Phy 115: Investigations in Astronomy</p>	<p>Chem 115: Chemistry in Daily Life</p>

Our basis for collaboration:
having students knowledgeable in one topic teach others

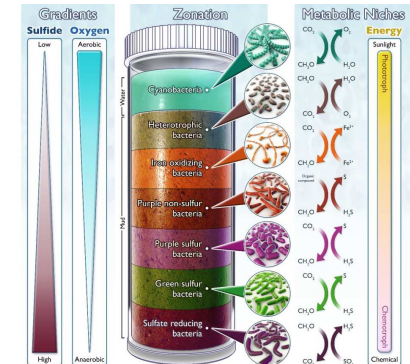
“Tell me, I’ll forget
Show me, I’ll remember
Involve me, I’ll understand”

Collaboration in a non-science majors laboratory: Winogradsky Column



Observation → Question
What causes the change?

Procedure:
Test for the presence of bacteria.

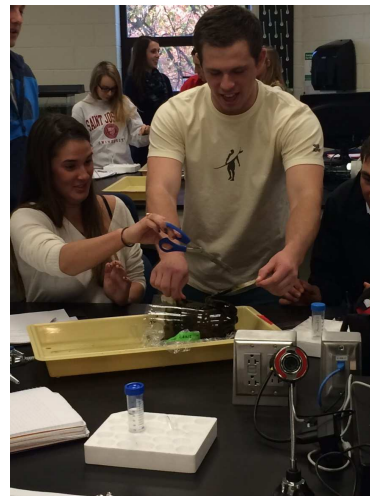


Identify what bacteria are present:

Non-majors: Identify the pigments.

Majors: Sequence the DNA

Grouping: Having majors work with non-majors

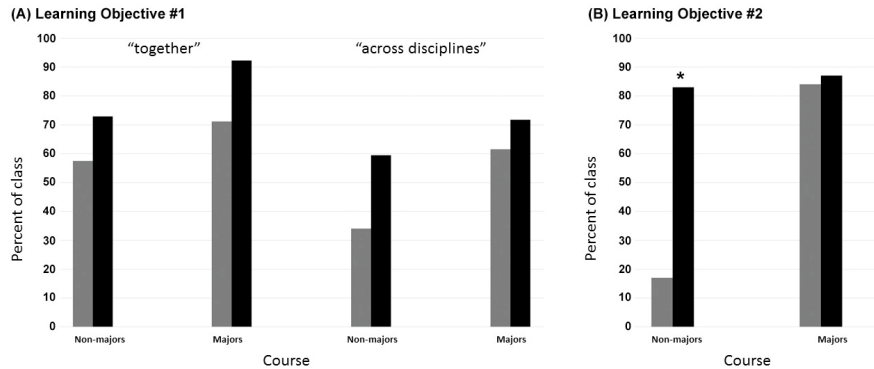


Grouping: Having majors work with non-majors

Collaborative Wiki Site



Grouping: Having majors work with non-majors



1. define the term collaboration as it applies to the nature of science;
2. state two different techniques of identifying microorganisms;

Ramirez et al. 2015
Journal Microbiology & Biology Education

Collaboration in a non-science majors laboratory: Having non-majors work with non-majors in a related, but different class

Students taking ENV 106: Learn about water quality and how to detect the presence of organisms in water.

BIO 165: Learn how to identify and classify organisms.

What happens when the two are put together?

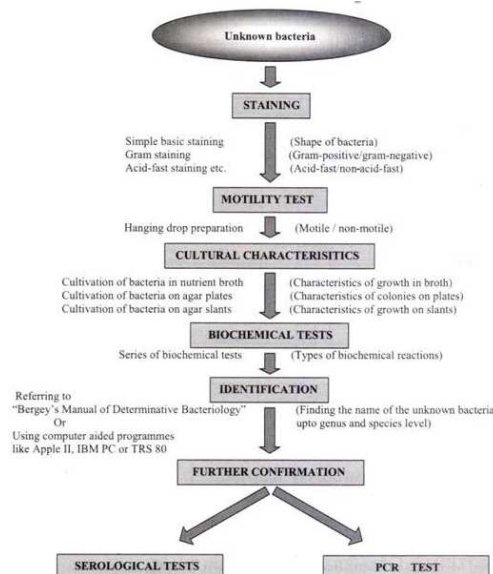


Is there any bacteria present in the water?



If so, what?

The ID of unknown bacteria is a traditional microbiology experiment

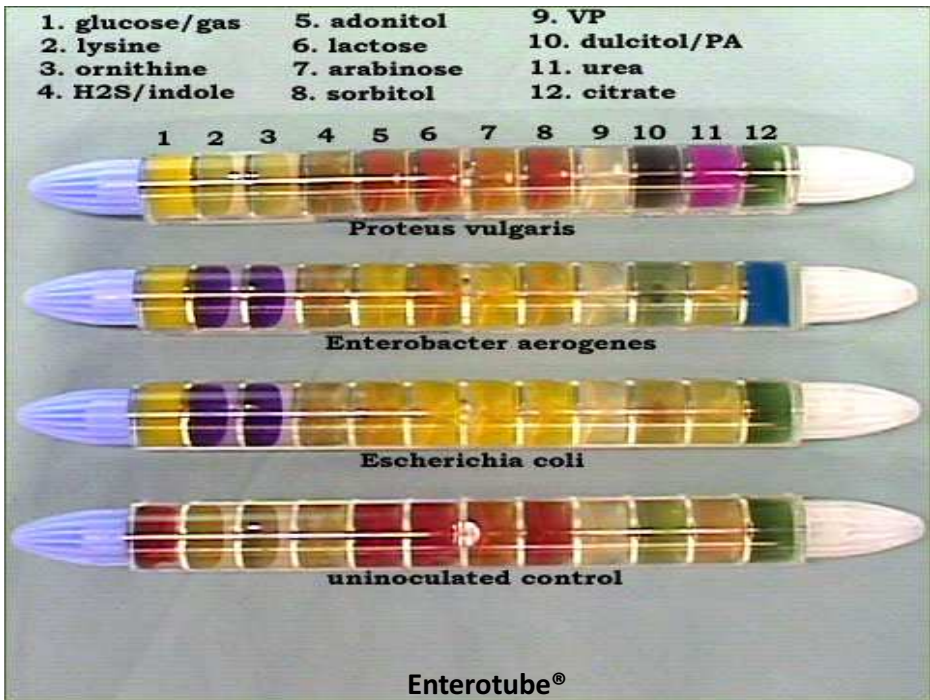


Saritha Pujari
<http://www.yourarticlelibrary.com/>

Bacteria Identification can be colorful!

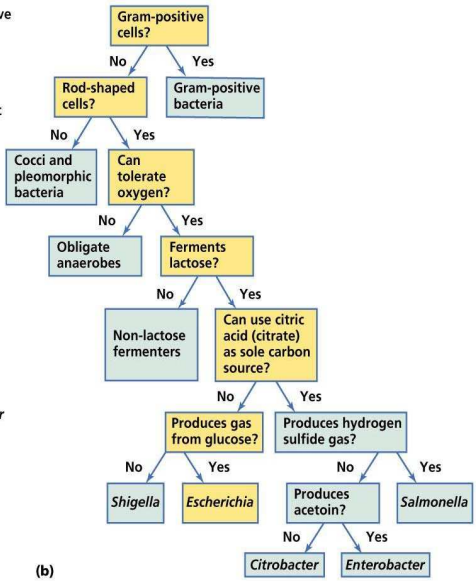


API®-20E



One skill we enforce in our classes is the use of a dichotomous key.

- 1a. Gram-positive cells..... Gram-positive bacteria
- 1b. Gram-negative cells..... 2
- 2a. Rod-shaped cells..... 3
- 2b. Non-rod-shaped cells..... Cocci and pleomorphic bacteria
- 3a. Can tolerate oxygen..... 4
- 3b. Cannot tolerate oxygen..... Obligate anaerobes
- 4a. Ferments lactose..... 5
- 4b. Cannot ferment lactose..... Non-lactose fermenters
- 5a. Can use citric acid as a sole carbon source..... 6
- 5b. Cannot use citric acid alone..... 8
- 6a. Produces hydrogen sulfide gas..... *Salmonella*
- 6b. Does not produce hydrogen sulfide gas..... 7
- 7a. Produces acetoin..... *Enterobacter*
- 7b. Does not produce acetoin..... *Citrobacter*
- 8a. Produces gas from glucose..... *Escherichia*
- 8b. Does not produce gas from glucose..... *Shigella*



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"It is an old maxim of mine that when you have excluded the impossible, whatever remains, however improbable, must be the truth."

-Sherlock Holmes
The Adventures of Sherlock Holmes

What ENV science students learn before being assigned the activity.

Basics of bacterial growth in the laboratory

Bacteria are microorganisms that can be grown in a wide variety of ways:

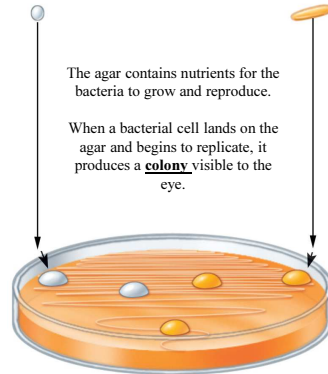
They can be grown in liquid suspension.



Before growth After growth

When the liquid is turbid, there is $> 10^7$ bacteria present!

They can be grown on semi-solid plates (nutrient agar plates).



Different bacteria produce different types of colonies (color, size, shape)

Detection of bacteria in water

The presence of microorganisms, particularly disease causing bacteria, in water samples is often difficult to detect, due to the low concentration of these cells in such environments.

It is also hard to look for every type of microorganism!

So, in testing water quality, we look for the presence of an **indicator organism**

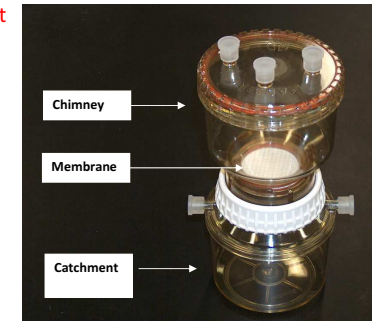
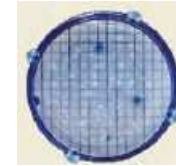
- Example: *Escherichia coli*

E. coli is a fecal coliform bacteria – it can ferment lactose and grow at 42C.

We can use a **membrane-filtration device** and “catch” any bacteria onto a membrane.

The membrane is then placed on semi-solid agar with nutrients to allow the bacteria to grow into **colonies**

Report any BLUE colonies

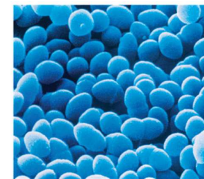


Membrane-filtration device

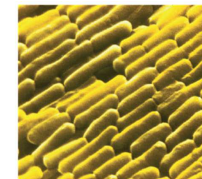
[VIDEO](#)

What BIO science students learn before being assigned the activity.

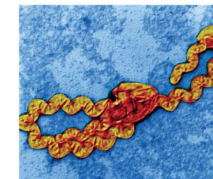
Bacterial Shapes



(a) spheres (**cocci**)



(b) rods (**bacilli**)



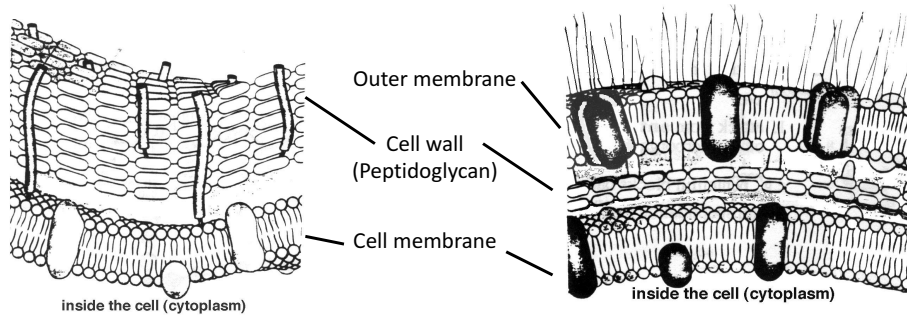
(c) spirals (**spirilla**)



(d) curved rods (**vibrios**)

Bacteria are classified according to their cell envelopes

The "cell envelope" in bacteria is everything outside and including the cell membrane.



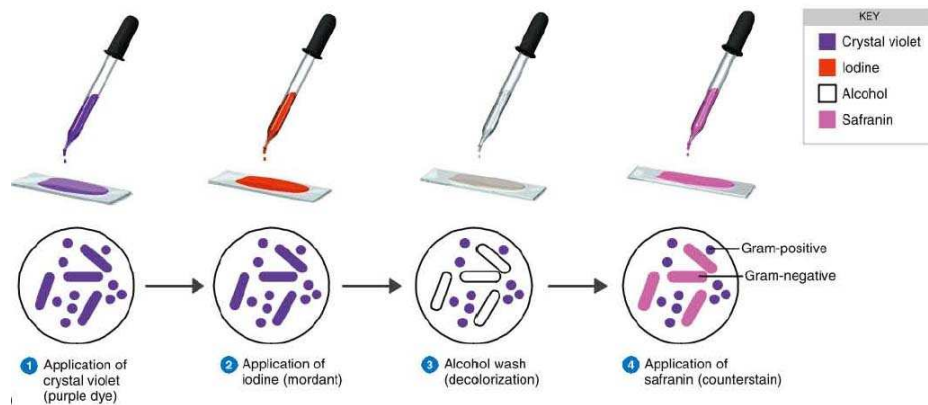
Gram Positive

Gram Negative

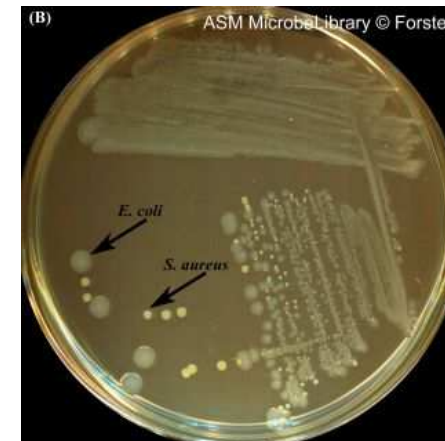
The Gram Stain Procedure



The Gram Stain Procedure

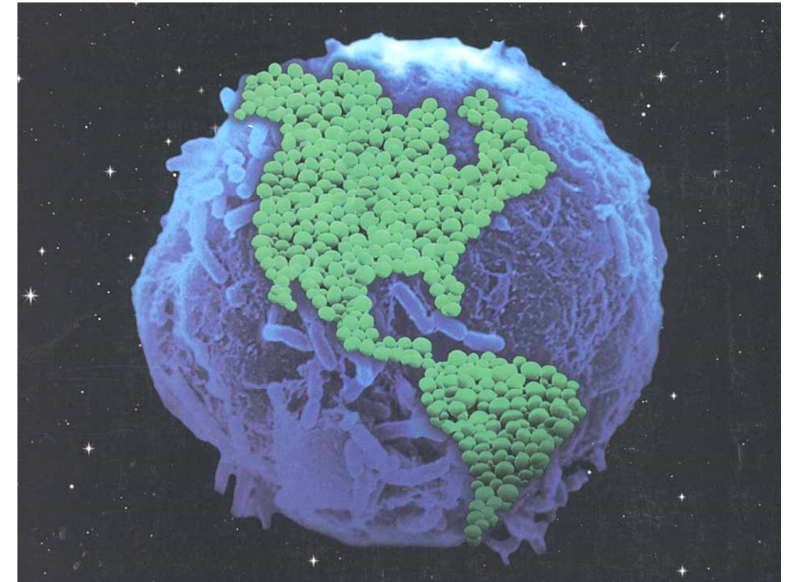


Different Bacteria can appear as different colonies on agar plates

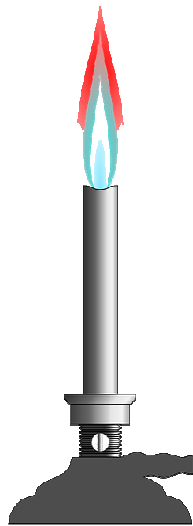


What both students learn before being assigned the activity.

Bacteria are everywhere!

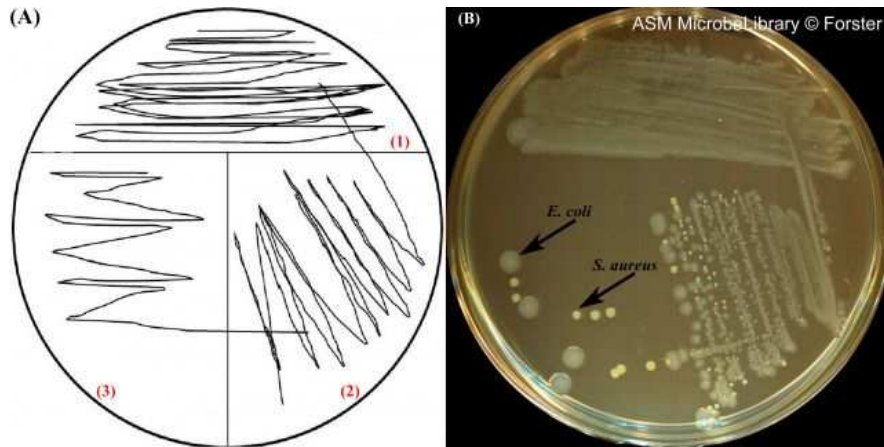


Aseptic Technique



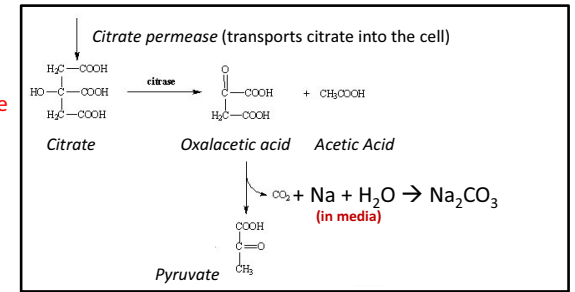
What both students learn while working through the assigned activity.

Streak plates can be used to separate strains in a mixed culture



Carbon Sources: Citrate Utilization

Tests the ability of some bacteria
To use citrate as sole carbon source



Procedure:

Day 1:
Inoculate citrate agar slant (urea + bromthymol blue)
Incubate at 37°C overnight

Day 2:
Observe results

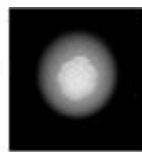
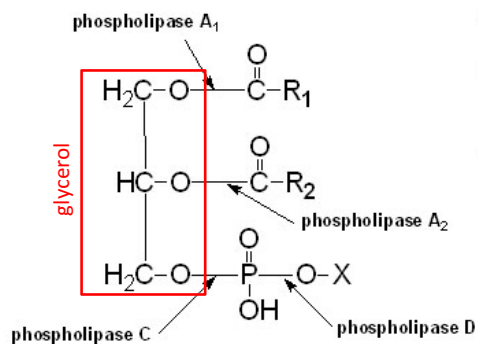
pH Indicator



If can use citrate:
sodium carbonate generated...
pH will increase...
turn bromthymol blue dark blue

If can not use citrate:
sodium carbonate not generated...
pH does not increase...
bromthymol blue remains green

Lipases: Lipid Hydrolysis

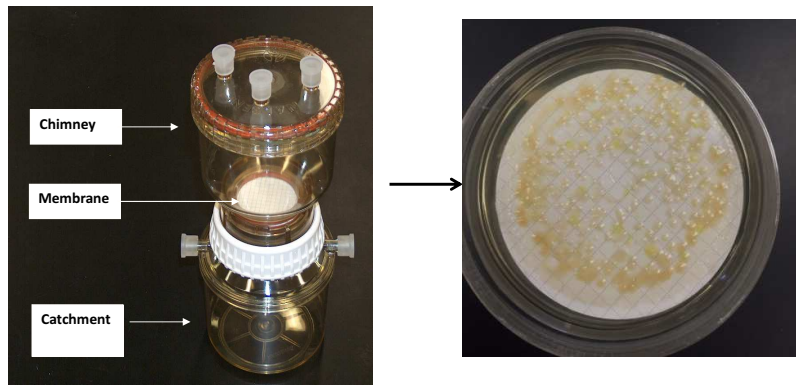


Potential Bacteria in Water Samples

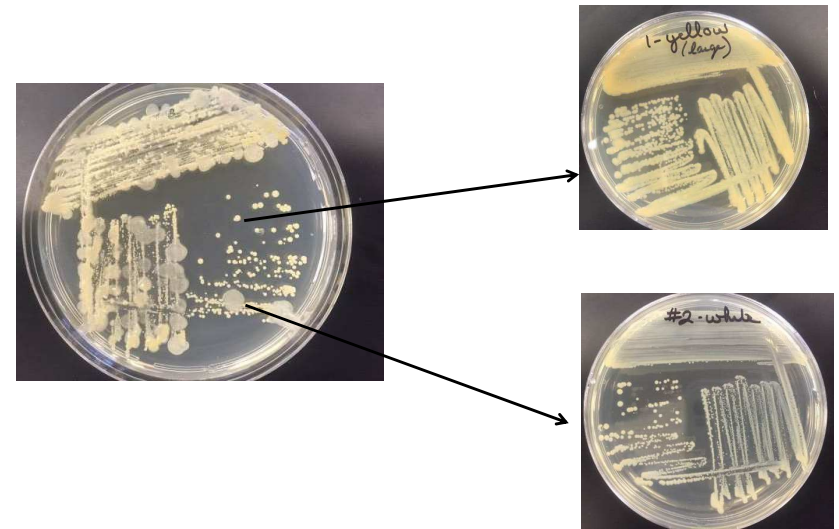
Bacterium	Gram-Reaction	Shape	Citrate utilization	Lipid Hydrolysis
<i>Escherichia coli</i>	Negative	Rod	Negative	Negative
<i>Enterobacter aerogenes</i>	Negative	Rod	Positive	Negative
<i>Staphylococcus aureus</i>	Positive	Coccus	Negative	Negative
<i>Bacillus cereus</i>	Positive	Rod	Negative	Positive
<i>Bacillus subtilis</i>	Positive	Rod	Negative	Negative

- Because Gram-positive bacteria are being used, you **cannot** use the mFC or mENDO agar.
- Use general growth media (Luria-bertani) with dark filter.

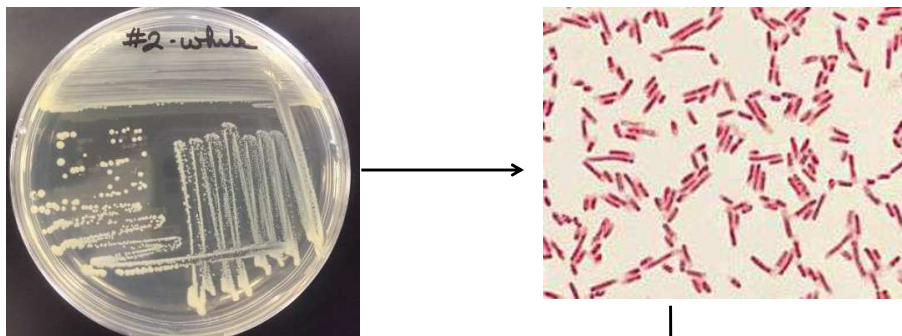
Detection of bacteria in water



Detection of bacteria in water

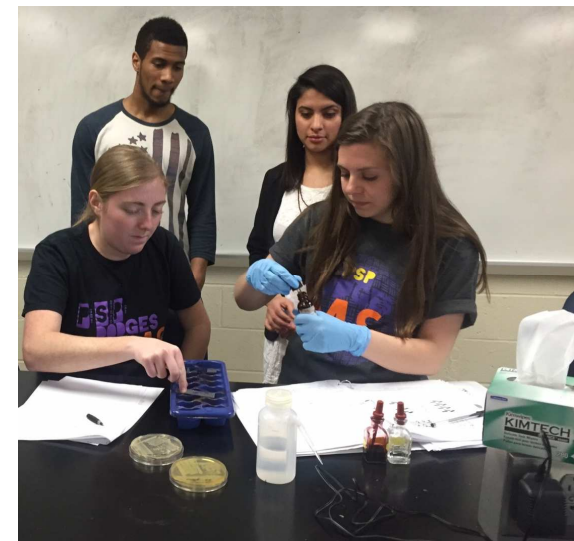


Detection of bacteria in water

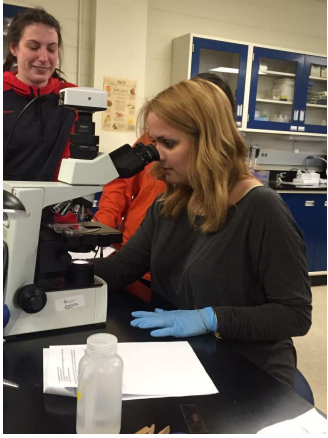


Use table to determine which test
(citrate uptake or lipid hydrolysis) to use.

Biology students showing the Environment students how to perform the Gram-stain



Gram-staining and inoculating egg yolk agar plates



Lab Budget to ensure critical thinking of lab results



Rich Uncle Pennybags

4 weeks in one afternoon...



4 weeks in one afternoon...

You have a sample of water.

You will filter the water through the membrane and plate onto LB agar
(*non-selective*).

You will then receive your plate after “one week” of incubation

- try to determine how many different types of bacteria you have.
- Pick one of each and re-streak onto a fresh LB agar plate.

You will then receive “your” re-streaked plates.

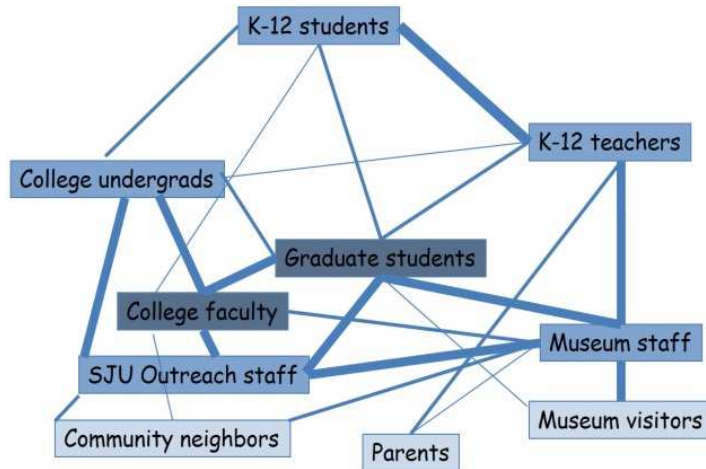
- Perform the Gram-stain
- Visualize your bacteria
- Decide what tests you want to choose next and inoculate.

You will then receive the results of “your” test.

- Determine what bacteria you have (if you are able to...)

Community of Practice

SJU views outreach and teaching philosophy as part of a larger **science education community of practice** designed to promote engagement and mutual learning for all participants

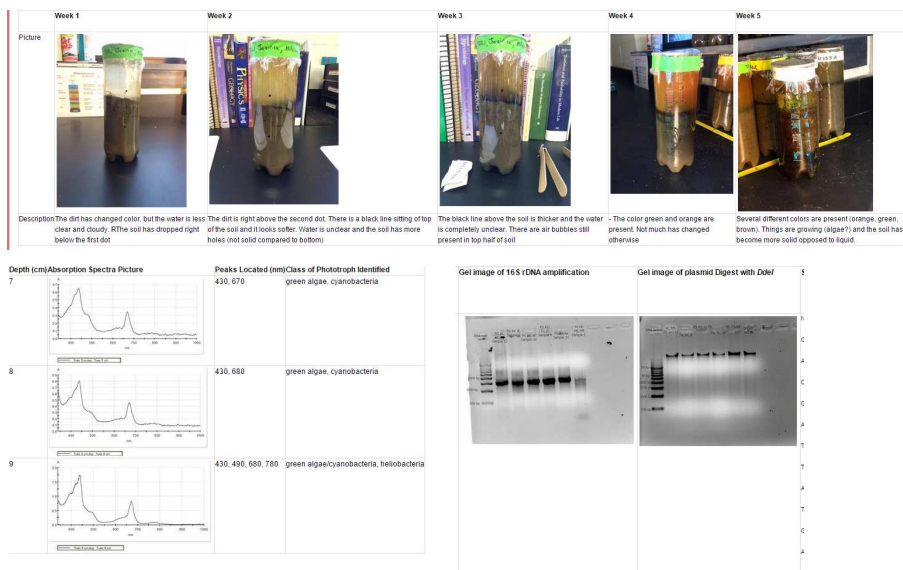


Logistics and Housekeeping

- How to get the classes together?
- How many times to get the classes together?
- Where to meet (is room big enough?)
- What about the rest of the curriculum/material we need to get to?

Meet together and work together online

Collaborative Wiki Site



Beginning to develop a collaboration (hypothetical)

On an index card, write down a **topic / lab technique** that you think your class would benefit from using collaboration.

- For that topic, does any **other class** cover a topic related to it?
- What type of students (intro, upper-class, non-majors?)
 - If so, do they use the same approach/technique?
 - What benefit would your class get from this?
 - What benefit would the other class get from this?

How do you address **logistic issues**?

- Getting together
- How many times
- Where to meet
- Timing

Writing teaching grants

- Same as writing a research grant – just apply it to your class

Research	Teaching
Question Posed	What do you want students to learn?
Sample Data	Sample procedure
What if experiment doesn't work	What if the students are having difficulty completing the activity or obtaining the learning objective?
\$ for research equipment	\$ for teaching supplies

- Ask questions while writing (especially to determine what equipment the grant will support).
- Need to be sustainable