Exercise 7

(Ed. Fall 2010)

Bacterial Growth and Colony Morphology: The Cultural Characteristics of Bacteria

INTRODUCTION:

Student Learning Objectives: After completing this exercise students will:

a. Demonstrate the ability to recognize morphological differences between different bacterial colonies.
b. Describe and define the morphology of bacterial colonies growing on agar plates and broth cultures.
c. Describe the differences in size and pattern of growth between different bacterial species.
d. Recognize some typical shapes and growth patterns of specific bacterial species

Activities:

- Observing colony morphology of prepared cultures.
- Streaking plates for observation of colony growth.
- Inoculating broth media to determine growth patters.
- Observing the results of inoculation after 24 hour incubation.
- Observation of pigment production by *Serratia marcescens* and *Pseudomonas aeruginosa*.
- Observing photographs of several species, and completing a table of characteristics.

Materials

Work in groups of 4 per table. This is a group activity. Each group needs the following:

- Photographic Atlas By Alexander and Streete
- 48 hour old streak plates of *Proteus vulgaris, E. coli, Serratia marcescens, Pseudomonas aeruginosa,* and *Micrococcus luteus*.
- Slant cultures of *E. coli, B. subtilis, P. vulgaris, P. aeruginosa, S. aureus,* and *S. pyogenes*
- Broth cultures of *S. marcescens* and *P. aeruginosa*
- Nutrient broth tubes (5 tubes)
- Nutrient agar 1.5% plates containing NaCl (2 plates)
- TSA Plates (6 plates)
- Dissecting scope or magnifying glass

Introduction:
Many bacterial species have distinct growth patterns on agar as well as in broth, which helps in their preliminary identification. You will be using the Microbiology Atlas pages 61 – 68 to help you describe the growth characteristics using the correct terms. You will also observe prepared cultures of several bacteria showing their characteristic growth morphology. You will observe these growth characteristics on agar (slant and plate) and broth. Remember to look at the colony size, shape, margin, and elevation. As you make your observations, you will also notice that some species will produce particular pigmentation, as well as characteristic odors typical of that species. Make note of these as well.

1. Observing colony morphology on prepared agar plates

- Obtain the 48 hour old culture streak plates and observe the growth characteristics of the colonies. Remember, these are pure cultures, and unless there is contamination, all colonies should look the same for each species.
- Draw a colony of each species and note the colony size, shape, margin, and elevation.
- Fill in the appropriate tables.

2. Streaking plates to observe colony growth characteristics

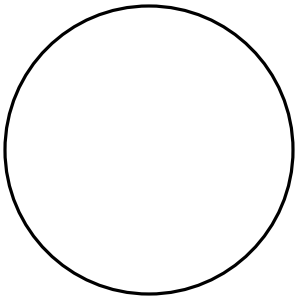
- Streak (for isolation) two NA 1.5% plates with *B.subtilis* and *P. vulgaris*.
- Streak (for isolation) 6 TSA plates with *E. coli, P vulgaris, S. marcescens, P. aeruginosa, S. aureus,* and *S. pyogenes*.
- Incubate the *S. marcescens, P. aeruginosa, P. vulgaris (NA 1.5%),* and *B.subtilis* plates at 30 C for 24 – 48 hours.
- Incubate the *E. coli, S. aureus, P. vulgaris (TSA plate)* and *S. pyogenes* plates at 37 C for 24 – 48 hours.
- Observe the growth characteristics of the colonies.
- Draw a colony of each species and note the colony size, shape, margin, and elevation.
- Fill in the appropriate tables.

3. Observing Growth Characteristics in Broth Cultures

- Using the loop and the slant cultures of *E. coli, B. subtilis, P. aeruginosa, S. aureus,* and *S. pyogenes*, inoculate 5 nutrient broth tubes. Remember your aseptic technique.
- Incubate the *B. subtilis* broth tube at 30 C, while all other broth tubes at 37 C.
- Do NOT shake or agitate the cultures when you remove them from the incubator.
- Observe for growth characteristics (turbidity, sediment, pellicle formation)
- Draw your observations and note your findings.

4. Complete the table of Cultural characteristics of selected Bacteria.

1. Prepared cultures – colony morphology



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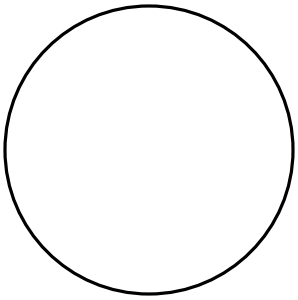

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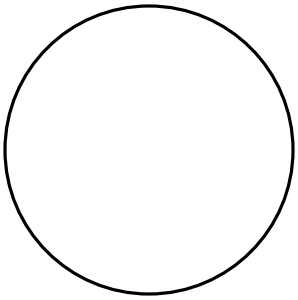
2. Student-streaked cultures – colony morphology



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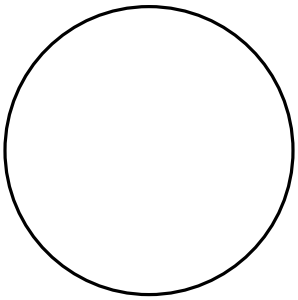

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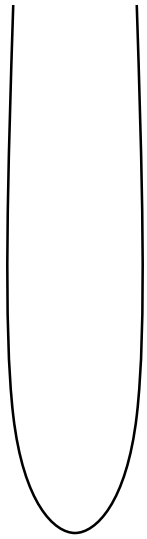
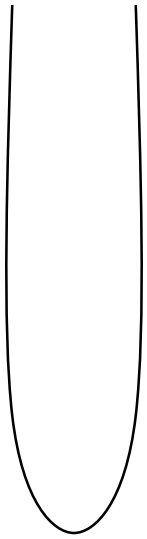
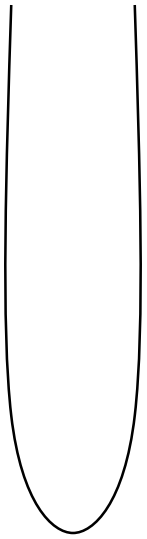
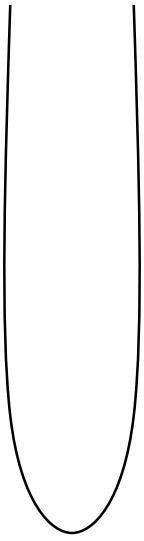
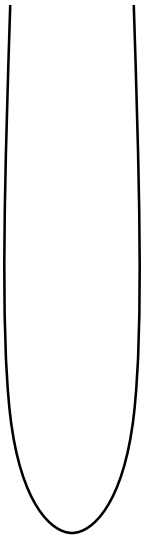
3. Growth in broth tubes

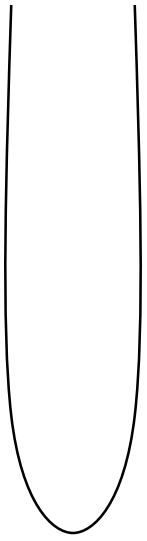












4. Cultural characteristics of colonies of selected Bacteria

Bacterial Species	Size (mm)	Shape	Margin	Elevation	Pigment
B. subtilis					
E. coli					
P. vulgaris (NA 1.5%)					
P. vulgaris					
P. aeruginosa					
S. marcescens					
S. aureus					
S. pyogenes					

Questions:
1. Which of these species has a characteristic "sweet corn tortilla" odor?

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2. If a broth shows the majority of turbidity is located at the bottom of the tube, then one can conclude that?

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3. Which of these has characteristic "swarming" growth on NA 1.5% agar at 30 C?

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