Exercise 19

Fungi: Molds and Yeasts F10

Or The Rotten World Around Us

INTRODUCTION:

Student Learning Objectives: After completing this exercise students will:

- a. Define the terms Saprophyte, Mycosis, hyphae, mycelium, spores, ascus, and budding.
- b. Identify the differences in cultural characteristics of selected fungi.
- c. Investigate the presence of fungi in environmental and household samples.
- d. Demonstrate the presence of fungi on different locations on the human body by growing them in cultures.
- e. Demonstrate the fermentation process of yeasts, and identify the importance of fungi in food production, ecology, as well as biotechnology.
- f. Describe the morphology of selected fungi, showing the different fungal structures.
- g. Identify and list 5 pathogenic fungi and the diseases they cause.

Lab Activities:

- Inoculation of Sabouraud's Dextrose Agar plates with *Saccharomyces cerevisiae* and *Candida albicans.*
- Inoculation of SDA plates with Penicillium spp. And Rhyzopus spp. Using the single dot inoculation method.
- Isolating and culturing mold from room air, AC filters, vacuum cleaners, cheese, refrigerators, footwear, and skin.
- Setting up a fermentation system.
- Observation of microscope slides of selected species of molds and yeasts.

Materials

Sabouraud Dextrose Agar Plates 8 per	Clean glass slides	
table	Lactophenol Cotton Blue	
Broth cultures of Saccharomyces	Cover slips	
cerevisiae and Candida albicans.	Clear Tape (to seal plates)	
Slant cultures of <i>Penicillium sp.</i> and	Prepared slides of selected fungi	
Rhizopus sp.	Litmus paper	
Grape juice	Forceps	
Small balloons	Lab Atlas	
Large screwcap test tubes		
Sterile cotton swabs		

Introduction: Lab Mycology Powerpoints and discussion on Fungi

1. Environmental sampling

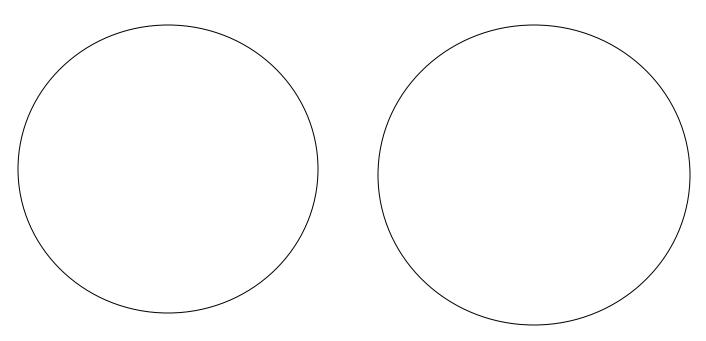
This part of the exercise will be given to you one week ahead of lab to give the fungi enough time to grow.

- 1. Each student will receive an assigned site to sample for fungi using a SDA plate.
- 2. Using a swab, touch the tip and roll on the surface being tested.
- 3. Inoculate the agar by rolling the swab on the surface.
- 4. Place the swab in a plastic bag provided and bring back to the lab.
- 5. Place a piece of clear tape (Scotch tape) to seal the agar plate.
- 6. Incubate the plate at room temperature right side up and bring to lab as soon as possible to continue incubation.
- 7. After several days of incubation, observe for growth.
- 8. Describe and draw the fungal growth. Do not open the agar plates in lab.

Discuss the results of your findings with the class. Draw your plate below.

2. Dot inoculation method to observe colony morphology

- 1. Using the inoculating needle and slant cultures of *Penicillium sp.* and *Rhizopus sp.*, inoculate SDA plates by touching the center (make a dot) of the agar surface.
- 2. Place a piece of clear tape to seal the agar plates.
- 3. Incubate at room temperature for several days and observe for growth.
- 4. Draw the growth seen on the top and the bottom side of the agar plate.
- 5. Do not open the plate in the room.



Top view of the plate

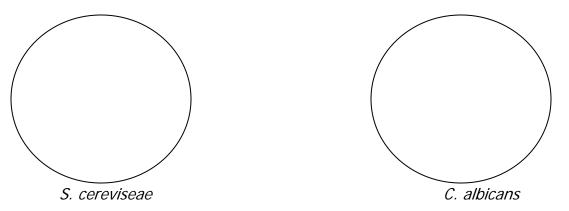
Bottom view of the plate

Are there any differences between the bottom and top growth? Describe.

What are the fuzzy structures (some might be black in color) you see? Explain.

3. Inoculation of *S. cereviseae* and *C. albicans*

- 1. Using the loop and the broth cultures of *S. cereviseae* and *C. albicans*, inoculate two SDA plates (streak for isolation).
- 2. Incubate at 30 C for 48 hours and observe for growth.
- 3. Make a wet mount of the colonies.
- 4. Draw the cells that you observe.



Do you see hyphae? Budding? Explain.

Name two pathogenic yeasts and the diseases they cause.

4. Fermentation of a substrate (sugar) by yeasts

- 1. Transfer 12 ml of grape juice into a large screw cap tube.
- 2. Inoculate with a loopful of *S. cereviseae* and place a deflated small balloon around the mouth of the tube.
- 3. Do the same for *C. albicans*.
- 4. Incubate at 30 C for 24 48 hours and observe for growth and gas production when the ballon is inflated.
- 5. Remove the balloon and smell the liquid for alcoholic odor. Record your observations.
- 6. Remove a small amount of the liquid and place on Litmus paper to measure the pH.
- 7. Record the pH (acidity) of both species.

Species	CO ₂ Production	Alcohol Production	PH (Acidity)
S. cereviseae			
C. albicans			

5. Prepared slides

Examine the slides and draw the structures you see in the space provided below. Label your drawings. You may attach additional pieces of paper.

6. Questions

1. Name three pathogenic mold species and the diseases they cause.

2. Name two medications used in the treatment of mycotic infections.

3. Why is it more difficult to treat mycotic infections compared to bacterial infections?

4. Define the terms:

- Saprophyte:
- Mycosis:
- Hyphae:
- Mycelium:
- Spores:
- Ascus:
- Budding:

5. How are yeast cells used in biotechnology? Give specific examples.