LAB WORK 3.

Subject: Viewing Specimens

Session Purpose: Learn of how to make a wet mount slide of a specimens. To view microorganisms learning to operate a microscope.

Objectives:

1. Learn of how to make a wet mount slide «The crushed drop» from liquid microbic culture.
2. Learn of how to make a wet mount slide «The crushed drop» from agar microbial culture.
3. Performexercises 1,2.

«The crushed drop» from liquid and agar microbial culture.

The preparation «the crushed drop» quickly prepares and allows to establish the form of cells, their sizes, character of congestions, mobility, but is short-lived.

Lab Exercise 1.b«The crushed drop» from liquid microbial culture.

Methodical instructions: Prepare a specimen of liquid microbial culture. Observe the cells of algae. Observe the following and describe what you see:

a) Chlorella vulgaris;
b) Spirulinaplatensis.

Procedure:

1. Obtain a clean microscope slide.
2. Using the dropper, place a drop of liquid microbial culture onto the center of a clean, dry slide.

3. Hold the side edges of the coverslip and place the bottom edge on the slide near the drop of specimen.

4. Slowly lower the coverslip into place. The water should spread out beneath the coverslip without leaving any air bubbles. If air bubbles are present, you can press gently on the coverslip to move the air bubbles to the sides.

5. Set up the microscope.
6. Remove the dust cover from the microscope. Plug in the microscope. Turn on the microscope's light source. Place the condenser at its highest position and close the iris diaphragm somewhat to prevent overbrightness. If the microscope has two ocular lenses, adjust the width between the lenses to your eyes. You will know the width is correct when you see one bright circle of light rather than two.
7. View the specimen with the **low-power objective.** Turn the nosepiece until the low-power objective locks into place.Place the slide on the stage and center it over the

condenser.Looking from the side, turn the coarse adjustment knob until the low-power objective is in its lowest possible position. Looking through the ocular lens, slowly turn the coarse adjustment knob in the other direction. This raises the low-power objective away from the slide. Continue until a clear image appears. Slowly turn the fine adjustment knob until the object comes into sharp focus. Adjust the light for maximum contrast using the iris diaphragm. Move the slide around on the stage using your fingers or the control knobs until you find a microorganism.
8. Sketch a picture of the microorganism.
9. Sign the pictureand specify Total Magnification (TM).
10. View the microorganism with the **high-power objective.** Center the microorganism exactly in the center of the field of view. Turn the nosepiece until the high-power objective locks into place. At this point, the microorganism should still be roughly in focus. Turn the fine adjustment knob slowly to focus the image. (Never use the coarse adjustment when the high power objective is in place!). Open the iris diaphragm until optimum contrast is achieved.
11. Sketch a picture of the microorganism. Sign the pictureand specify Total Magnification (TM).

Lab Exercise 2.«The crushed drop» from agar microbial culture.
Methodical instructions: Prepare a specimen of agar microbial culture. Observe the cells of Yestsand describe what you see.

Procedure:
1. Obtain a clean microscope slide.
2. Place one drop of water on the slide.

3. Sterilize the transfer loop in the in the burner flame (NOTE: Never pick up the loop to use it or put it down without sterilizing it first!)
4. Allow loop to cool.
5. Using your sterilized inoculation loop, obtain a small sample of microbial culturefrom thesource tube.

Note:
1) If obtaining microbe sample from slant tubes:
- Never pick up test tube by the cap;
- DO NOT set cap down on lab bench;
- Flam neck of the test tube before and after obtaining sample.

2) Be gentile! The most common error is transferring **too much** inoculum (bacteria) when making smears from solid media. The microbial colonies are found growing on the surface of the agar medium. DO NOT remove agar with your sample!

3. After mixing the inoculum in the water, flame the loop to sterilize it before setting it down.
4. Hold the side edges of the coverslip and place the bottom edge on the slide near the drop of specimen.

5. Slowly lower the coverslip into place.

6. View the microorganism with the low-power objective (x10) and high-power (x40) objective.
7. Sketch a picture of the microorganism.
8. Sign the pictureand specify Total Magnification (TM).

Proper Storage of Your Microscope

To practice proper care of your microscope, make certain to clean it and put it away properly at the end of this exercise.

1. Make certain the slide is removed from the stage.
2. Clean all lens with Lens Paper. Obtain a clean sheet of lens paper. Rub oculars to clean.
3. Pull the body tube away from the stage (i.e. lower the stage as far as possible)
4. Turn off the microscope. Turn the nosepiece of the microscope until the low-power objective locks into place. Carefully lower the objective to its lowest position by turning the coarse adjustment knob.
5. Turn off the light source.
6.Remove your slide. Clean the slide and coverslip with water.
7. Unplug the microscope and store it under a dustcloth.
8. Wrap the cord.
9. Return the proper storage location in the cabinet.

Equipment:

- Microscope
- Slide
- Coverslip
- Dropper
- Dropper bottle of water
- Disinfectant tray
- Liquid culture of Chlorella vulgaris
- Liquid culture of Spirulinaplatensis
- Culture of Yeasts in slant tubes
- Inoculation loop
- Burnerflame