HELMINTHS


In the adult stage, helminths are definitely macroscopic. However, they are usually diagnosed in a clinical specimen by the appearance of their ova (eggs) or larval forms. If the doctor suspects a helminthic or protozoal infection, an "O and P" will be ordered. O stands for ova or eggs and P represents parasites or protozoa.

**Demo slides:** Eggs: Clonorchis sinensis, Taenia sp., Ascaris lumbricoides, Enterobius vermicularis, Schistosoma Larva: Strongyloides stercoralis Scolex: Taenia sp.

You should be able to identify each helminth in its egg or larval stage as well as name the disease, symptoms of the patient, type of clinical specimen that would be taken and the source of the organism. It may be helpful in taking the practical to remember which objective is used for viewing the different helminths. Most are viewed at 40x but some are viewed at 10x or 4x.

*Clonorchis sinensis*

40x
*Strongyloides stercoralis*
Larval form
10x

*Ascaris lumbricoides*
ova in feces
40x

*Enterobius vermicularis*
ova in feces
40x
Decrease the light to increase your success of finding this one.
*Taenia* sp.

scolex (head of worm)

4x

You'll never forget this one!

*Taenia solium*

ova

40x

These are almost transparent so you will need to decrease the light.
Protozoa

These are unicellular eukaryotic organisms. Most of the protozoa exist in two forms--the trophozoite and the cyst. Some only have a trophozoite stage.

Depending upon the differential diagnosis, the clinical specimen will either be blood or stool. History taking on the part of the physician or nurse is vital in helping to identify possible exposure to these organisms.

Demo slides: Cysts: Entamoeba histolytica, Giardia Trophozoites: Giardia, Plasmodium sp., Trichomonas, Trypanosoma Toxoplasma gondii (there is no slide for this organism)

For each of these, you should be able to recognize the trophozoites and/or cysts, know the name of the disease caused, symptoms, transmission and clinical specimen.

**Trypanosoma sp.**

Trophozoite in blood outside red blood cells

40x

**Entamoeba histolytica**

Cyst in feces

40x
**Giardia lamblia**

Trophozoite or cyst in feces

40x

The troph looks like a tear drop and the cyst usually appears oval with a dark line in it.

Pictures by Gary Moo Young, Science Specialist, PBCC

**Plasmodium sp.**

Various forms inside red blood cells

40x
Trichomonas vaginalis

40x
With the increasing number of immunocompromised patients, the isolation and identification of fungi from clinical specimens have become more important. Yeasts can be distinguished from molds both microscopically as well as macroscopically. Yeast is slimy like bacterium, but you can notice filamentous growth like fungus under the microscope.

Demo slides and plates: Yeast: *Candida*, Mold/fungi: *Aspergillus*, *Penicillium*


![Yeast growth, 40x](image1.png)  ![Yeast growth, SAB (*Candida albicans*)](image2.png)

![Penicillium sp., 40x](image3.png)  ![Penicillium on SAB](image4.png)
Aspergillus

Aspergillus on SAB
UBIQUITY OF MICROORGANISMS

Frequency of handwashing would certainly increase if we could only see the microbes that are around us---on doors, in the air, on our clothes, under our fingernails, etc. Microbes are indeed ubiquitous---they're everywhere. Students sampled different environments for this lab---here are just a few of the results. Samples were inoculated onto TSA (Tryptic Soy Agar) plates, a general all purpose medium.

Link:  https://en.wikipedia.org/wiki/Trypticase_soy_agar

Materials: 4 nutrient agar (TSA) plates, swabs and saline tubes

A student allowed the sleeve of her lab coat to touch the agar. Two different molds grew. Now do you see why you need to cover the plate as much as possible when you are streaking it?

A student dragged her fingers across the agar resulting in multiple kinds of bacteria. These are normal flora but could be harmful to an immunocompromised patient or one with burns or open wounds.
Two curious male students swabbed the inside door of the men's bathroom. You can see several different kinds of bacteria growing. Whether or not they are harmful depends on many factors. Most surfaces would yield similar results.

This growth resulted from a nasal swab. The nose is a wonderful place for microbes!

This microbial community was cultured from the sole of a student's shoe. Evidently we cannot control microorganisms by stepping on them!
STAINING TECHNIQUES

Link:  https://courses.lumenlearning.com/microbiology/chapter/staining-microscopic-specimens/

Simple Stains pages 100 - 102 Use procedure on page 101 for making a smear of either *Escherichia coli* or *Staphylococcus epidermidis*. Once the smear is completed, follow instructions for Gram stain in Experiment 3-6, page 109. 3-8

Video for Procedure (slightly modified):  
https://www.youtube.com/watch?v=sxa46xKfIOY

Gram Stain pages 105 - 109 Use procedure on page 101 for making a smear of either *Escherichia coli* or *Staphylococcus aureus*. Once the smear is completed, follow instructions for Gram stain in Experiment 3-6, page 109. Each student does a Gram stain on the smear they made in 3-5.

Link:  https://en.wikipedia.org/wiki/Gram_stain

Video:  https://www.youtube.com/watch?v=qdZrT5yB-kg

Acid Fast Stain pages 110 - 114 Demo slide of *Mycobacterium tuberculosis* 3-9

Link:  https://en.wikipedia.org/wiki/Ziehl%E2%80%93Neelsen_stain

Video:  https://www.youtube.com/watch?v=YzTgHU-aCqo

Capsule Stain pages 115 - 116 Demo slide of *Klebsiella pneumoniae* 3-10 Endospore Stain pages 117 – 120

Link:  https://en.wikipedia.org/wiki/Bacterial_capsule

Video:  https://www.youtube.com/watch?v=-Yv-Ye3aNt4
ASEPTIC TRANSFER

Video for procedure: https://www.youtube.com/watch?v=bRadiLXkqoU

1-3 Aseptic Transfer of Microorganisms pages 17 - 24. Organisms: Staph aureus, Escherichia coli (slants) Each student transfers one organism (either B. subtilis or E. coli) into a broth (liquid medium) and a slant (semi-solid medium, slanted). Read the Basics, pages 17-24. Follow instructions on page 21 for removing a sample from a slant; page 22, fish-tail inoculation; page 22, inoculation of broth tubes. A practice run will be done by each student using blank tubes.

ISOLATION OF A SINGLE COLONY OF BACTERIA

STREAK-PLATE METHOD

Video for procedure: https://www.youtube.com/watch?v=0heifCiMbfY

1-4 Streak Plate Method of Isolation pages 25 - 28. Your instructor will demonstrate a correct streaking technique. Each student will do a practice run on an empty Petri dish and then streak the following mixture onto an agar plate. Organisms: mixture of Serratia, Enterobacter.

SPREAD-PLATE METHOD

(Know the procedure, we did not do in lab)

Video: https://www.youtube.com/watch?v=FwW_2ii1Zoo