

**Microbiology 2730
Study Guide #3
Winter Semester 2008
E. Hoffman**

LABORATORY SECTION

THE SIMPLE STAIN

Textbook reference for this set of questions is chapter 3
You should also read the introductory material presented in your laboratory manual

1. What is the major purpose behind conducting a “simple stain” on a smear of bacteria?
2. The procedure known as a simple stain, involves what action being carried out on a smear of bacteria?
3. Biological stains can be classified as being basic, acidic, or neutral. Define each of these stain types based on the nature of their chromophore group. What is a chromophore group?
4. In conducting the simple stain, you “heat fixed” your smear before conducting the actual staining procedure. What is the major reason for carrying out the heat-fixing step?
5. Why should you start flaming a loop from the back to the front, as opposed to the opposite direction? What does the flaming procedure accomplish?
6. What are the 3 major cell shapes that bacterial cells usually assume?
7. Chains of cocci are referred to as a ___ cell arrangement.
8. Irregular clusters of cocci (grape like) are referred to as a ___ cell arrangement.
9. A group of cocci, arranged to form a packet or cube of 8 cells is referred to as a ___ cell arrangement.

THE GRAM STAIN

Textbook reference for this set of questions is chapter 3

You should also read the introductory material presented in your laboratory manual

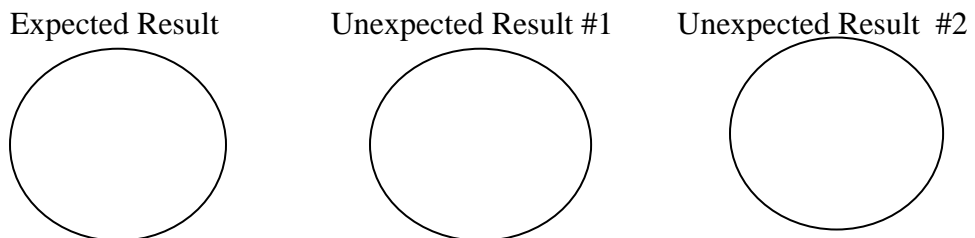
1. The Gram stain is considered to be an example of a differential stain. What does this imply?

2. In the case of the Gram stain, you should be able to identify the following reagents:
 - a. The Primary Stain
 - b. The Mordant
 - c. The Decolorizing agent
 - d. The Counter stain

3. If a Gram stain is correctly carried out, a Gram (+) bacterium will appear ___ in color, while a Gram (-) bacterium, will appear ___ in color.

4. For best results, the Gram stain should be conducted, only on “young” cultures. Why?

5. Given the indicated normal Gram stain, explain the “unexpected” results shown below. Note: The bacteria in all fields are of the same species.
 * = Purple in color o = Pink in color



6. Table 4.1 on page 88 of your textbook lists numerous differences between Gram (+) and (-) bacteria. You should be able to cite 3 of these differences.

LABORATORY CALCULATIONS

Here is the “recipe” for the construction of the Gram’s Iodine solution that you used in carrying out the Gram Stain. This information comes from the eighth edition of Microbiological Applications by Benson. Dissolve 2.0 gm of potassium iodide in 300 ml of distilled water and then add 1.0 gm of iodine crystals. The question is, how would you make 3.5 liters of this reagent. Note, I have not lectured on this, just see what you can do yourself. If you have questions, please feel free to ask.

THE FUNGI

The index of your textbook should be consulted for page references.

1. During the course of the laboratory, you had the opportunity to view several different species of fungi. One of those fungi has been given the scientific name of *Penicillium chrysogenum*. This particular fungus has proven to be very important to medical science for what reason?
2. The other fungal organism that you had the opportunity to view was that of *Rhizopus stolonifer*. This particular fungus is identified as being one of the common black molds that grow on ____, a common household item.
3. The slide materials associated with the fungal demonstrations allowed you to view cells and structures that are associated with the process of ____ (sexual or asexual) reproduction in these molds.
4. In my very brief comments on fungal anatomy I noted two terms, mycelium and hypha. You should be able to relate these to the molds that you had the opportunity to view on demonstration.

The Textbook Reference for this set of questions is chapter 2

1. In your brief introduction to chemistry, it was noted that the world of chemical substances that we see about us can be divided into 2 basic types:
 - a. Elements
 1. These are approximately ____ known elements, of which ____ are naturally found on the earth

2. Analysis of materials taken from a variety of living organisms indicates that 4 of these elements make up about 99% of living matter. What are these elements?
3. You should be able to define the term, element.

b. Chemical Compounds

1. While the number of different elements is really quite small, what did I say about the # of known chemical compounds?
 2. You should be able to define the term, chemical compound.
 3. The world of chemical compounds is usually divided into 2 major subgroups?
 4. Define each of the above-mentioned subgroups in terms of C to C or C to H bonding.
2. When discussing chemical elements and compounds, 3 types of particles normally are mentioned. What are these? Briefly define each.
 3. Chapter 2 includes a brief discussion of chemical bonds:
 - a. These bonds result from differences in how ___ are shared between atoms.
 - b. What are the three classes of chemical bonds that I mentioned in lecture?
 - c. Which of those bonds is an outgrowth of an actual electron transfer between atoms?
 - d. Which of these bonds is the result of a relatively equal sharing of an electron pair between two atoms?
 - e. Which of these bonds is the result of markedly unequal sharing of an electron pair between two atoms?
 4. A very important "bond" in biologically important molecules is that known as the hydrogen bond. This bond can be described as an attraction between areas of ___ electrical charge. Hydrogen bonding can be found between individual ___ and ___ the same molecule (if its chemical makeup permits).
 5. You were introduced to two types of chemical formulas.
 - a. $\text{C}_2\text{H}_5\text{O}_2\text{N}$ is the ___ formula for the amino acid, glycine.
 H H O
 - b. H-N-C-C-OH is the ___ formula for the same amino acid.
 H

These formulas, while cumbersome to write, do provide what kind of information concerning the molecule under discussion

6. You were introduced to the pH scale.
 - a. If you were to determine the pH of Lake St. Claire the pH reading that you would get would fall some where between ___ and ___.
 - b. What number on the pH scale represents neutrality? Readings which are acidic in nature fall in what are of the pH scale? Readings which are basic in nature fall in which area of the pH scale?
 - c. It was noted in class that the numbers associated are quite different that what we usually make use of in our daily lives. If the pH of a person's blood changed from a pH of 4 to 2, what could you say happened to the level of acidity of the individual's blood?

7. If you were to put a sodium atom and a chlorine atom very close together, a chemist would predict that the sodium atom would be oxidized and the chlorine atom would be reduced. In term electrons, what would happen?

8. Some time was spent discussing the so-called four "molecules of life". What were the 4 groups of organic molecules included in this group?

9. Carbohydrates:
 - a. As noted in class, carbohydrates are a rather large and diverse group of chemical materials. Many of the commonly encountered ones contain 3 elements, ___, ___, and ___ and exhibit a 2 to 1 ratio between ___ and ___.
 - b. Carbohydrates are routinely placed into 1 of 3 major classes based on the results of their total hydrolysis.
 1. What occurs in the process of hydrolysis?
 2. What are the 3 classes mentioned above? Cite an example for each of the above-mentioned classes.
 3. Carbohydrates have several major cellular functions. You should be able to cite 2 such functions.
 - c. In the discussion of carbohydrates, you were introduced to the term, polymer. You should be able to define this term.

10. Proteins:
 - a. Proteins are normally described as being polymers of ___. At the present time, chemical analysis indicates that there some ___ different kinds of these subunits routinely found in proteins.
 - b. All proteins contain 4 elements, what are they? Note, some proteins may contain other additional elements.
 - c. Proteins have numerous functions in living cells. You should be able to cite 4 such functions.

11 Nucleic acids:

- a. Nucleic acids are usually described as being polymers of _____. You should be able to describe one of these structures.
- b. The world of nucleic acids comes in two major forms or groups. What are these groups (you should be able to spell the names out as well as knowing their acronyms)
- c. Probably the most famous organic molecule in the world is DNA. The structure of this molecule is usually presented as a _____ helix. This structure was worked out in 1953 by the team of _____ and _____ and won for them the Nobel Prize in the year _____ (see your textbook)
- d. If presented with the type of diagram shown below, you should be able to identify the parts of the DNA molecule indicated by an asterisk (*)
- e. Nucleic acids have several functions within living cells, however all of these functions seem to revolve around their role as serving as the _____ information of cells.

12. Lipids:

- a. Lipids encompass a very large and diverse group of chemical substances. For the purposes of this course, we will consider only two of these groups, the triglycerides and the phospholipids. Both of these groups contain 3 different elements. What are these elements? How does the hydrogen to oxygen ratios compare to that of the common carbohydrates?
- b. The complete hydrolysis of a triglyceride results in the formation of 1 molecule of _____ and 3 molecules of _____.
- c. Phospholipids have a structure somewhat similar to the triglyceride except that one of the fatty acids has undergone a substitution such that end result is a molecule, which has a portion, which is soluble in water, and another portion, which is not. This property of phospholipids allows them to function as the structural backbone of what very important structure in cells?
- d. You should be able to cite 2 functions for lipids in living microbial cells.