MICROBIOLOGY STUDY GUIDE

Sierra College Biological Sciences Department
Bio. Sci. 4

The questions included in this study guide are representative of those found on quizzes and exams given in the microbiology course provided by Harriet Wilson. For your convenience, the sample lecture questions (from quizzes and exams) have been divided into sets according to the topics indicated on your lecture schedule. Four sample laboratory exams (two for lab exam #1 and two for lab exam #2) are also included. Most duplicate questions have been eliminated; however, multiple questions containing different wording but requiring the same answers are included. Remember to read questions carefully and completely before attempting to answer them, especially when taking quizzes and exams.

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**History of Microbiology:**

(5) 1. Define:

   Microbiology

   Abiogenesis

   Koch's postulates

   Etiological (etiologic)

   "Magic bullet"

(1) 2. Microorganisms were being used by humans long before they were observed or recognized to exist. What were they used for?

(2) 3. ________________________________ may be defined as the science or study of living organisms too small to be observed with the naked eye. What types of organisms fall into this category, i.e., what groups of organisms would be identified as microbes? ________________

(2) 4. The science or study of organisms too small to be observed with the naked eye, i.e., those requiring magnification for observation, is called ________________________________, and was first popularized by the Dutch naturalist ____________________________________. This individual made his own microscopes, observed living microorganisms, and documented his findings with a well-established scientific organization, the Royal Society of London.

(2) 5. ________________________________ is sometimes credited with being the founder of microbiology because he was the first to observe living microbes and because he documented his findings. However, microbiology could not become a true science until the concept of ________________________________ was shown to be invalid.

(2) 6. Anton Van Leeuwenhoek is sometimes referred to as the “father of microbiology”. What did he do to receive this recognition?

(2) 7. The concept of ________________________________ or the spontaneous generation of living organisms was shown to be invalid at the macroscopic level by Francesco Redi (working with flies on meat). This long accepted presumption was disproved at the microscopic level by several investigators including Lazzaro Spallanzani and ________________________________.

(2) 8. Two individuals given credit for discrediting the concept of abiogenesis at the microscopic level are ________________________________ and ________________________________.
9. While working as a chemist for the French wine industry, Louis Pasteur discovered that microorganisms were responsible for the ___________ process (converting grape juice into ethanol or other substances). Pasteur also developed a heat treatment that could be used to control the populations of microbes within juice without destroying the flavor. This treatment is called ___________________________ and is still used to control microorganisms in food materials today.

10. ___________________________________ was an English surgeon recognized for developing and using aseptic or antiseptic techniques in his surgical procedures. What methods did he use?

11. What was the significance of Koch’s postulates at the time they were developed?

12. If an investigator were using Koch’s postulates to identify an etiological agent, and the microbe had been isolated and grown in pure form, what would be the next step in the investigation?

13. What important contribution did Richard Petri make to the study of microbiology?

14. Louis Pasteur is credited with being the first microbiologist to use attenuated microbial cultures to prevent disease in humans. Pasteur used the term ___________________________ to identify such cultures, thus crediting Edward Jenner for his work with cow pox.

15. By the late 1800s it was recognized that microorganisms were responsible for causing a variety of diseases. The work of Jenner and Pasteur indicated that microorganisms could also be used to ______________________________ disease, and later investigations conducted by Alexander Fleming showed that the products of microorganisms could be used to __________________ disease.

16. What contribution did Fanny Hess make to the science of microbiology?

17. ___________________________________ coined the term “magic bullet” in reference to chemicals that could be taken internally to kill disease causing microorganisms without doing damage to the body.

18. ___________________________________ was a Scottish bacteriologist and medical practitioner recognized for his discovery of __________________________, one of the first antibiotics obtained from fungi in the genus Penicillium.

19. Multiple topics included in microbiology texts are currently recognized as separate disciplines in the biomedical sciences including: __________________________, the study of immune function and disease prevention, __________________________, the study of chemicals used to control microorganisms inside the body (and to treat other types of diseases/disorders), __________________________, the quantitative study of disease and factors influencing disease frequency and distribution within populations, and __________________________, the study of disease etiology, pathogenesis and the anatomical and physiological changes occurring within diseased individuals.
Characteristics of Life and Biochemistry:

(5) 1. Define:

Assimilation

Protoplasm

Electrolyte

Polysaccharide

Triglyceride

(2) 2. Two characteristics that all living organisms have in common are the ability to  
_________________________________ and ____________________________________.

(2) 3. All living organisms are able to reproduce, grow by means of a process called  
_________________________________, carry out metabolic processes, respond to stimuli,  
_________________________________ and maintain a high degree of organization.

(2) 4. Evolution, a biological phenomenon involving changes in populations of organisms over time  
requires _________________________, i.e., changes in nucleotide sequences within DNA or RNA molecules, and _________________________, which may be rapid or slow. Ultimately populations change because there is variation among the organisms present, and the environment exerts selective pressure, i.e., only some of the organisms survive and reproduce.

(1) 5. __________________________________ is the name given to the living dynamic material  
that all cells are made of. This material is sometimes referred to as the chemical and physical basis for life.

(1) 6. The thirteen elements that make up about 99% of any living organism (by weight) are:  
_________________________________________ (Chemical symbols are adequate here.)

(2) 7. Positively charged particles of matter (may be atoms or molecules) are known as  
___________, while negatively charged particles of matter (may be atoms or molecules) are  
known as __________.

(1) 8. An inorganic compound that aids transport, helps maintain cell size and shape, resists  
temperature change and serves as an important solvent within living cells is ________________.
9. Chemical reactions that involve the splitting of water and the splitting of organic compounds may be referred to as __________________________ reactions. When small organic monomers are joined together by removing water molecules, the reactions involved may be called ________________________________ reactions.

10. Organic compounds referred to as ___________________ contain primarily carbon, hydrogen and oxygen and serve as an important source of energy and carbon for living organisms. If these compounds contain six carbons, are sweet to the taste and soluble in water they are called __________________________ if they are macromolecules containing many sugar units, do not taste sweet and are insoluble in water they are referred to as __________________________.

11. Carbohydrates such as arabinose, ribose and deoxyribose contain five carbon atoms and are referred to as __________________________.

12. Sucrose, lactose and maltose are examples of carbohydrates known as ____________________, while starch, glycogen, cellulose and agar are examples of carbohydrates known as __________________________ because they are made up of many sugar units.

13. The primary structure of a protein is determined by the sequence of __________________________ present in that protein. If a protein contains more than one polypeptide chain, it is said to have ________________________________ structure.

14. The formation of a protein molecule involves the removal of __________________________ molecules (condensation or dehydration synthesis) and the formation of covalent bonds called __________________________ bonds between adjacent amino acids. A long chain of amino acids may also be called a polypeptide.

15. Lipids that contain three fatty acid chains attached to a glycerol "backbone" are called ________________________________, and serve an essential role in the structure of cellular membranes.

16. Lipids that contain the maximum number of hydrogen atoms possible are referred to as __________________________ fats and tend to be solid at room temperature. Those that contain one or more __________________________ have fewer hydrogen atoms and tend to be liquid at room temperature.

17. Lipids that contain two fatty acid chains and a phosphate group attached to a glycerol “backbone” are referred to as _______________________________. These molecules have both polar (hydrophilic) and non-polar (hydrophobic) parts in their structure, and so are said to be ________________________________.

18. Ring-form lipids called sterols are essential to the physiology of eukaryotic organisms, and are often incorporated into cellular membranes. For example, __________________________, an important sterole, forms approximately 10% of the lipid content within animal cell membranes. The cell membranes of bacteria typically lack such compounds.
(2) 19. Nucleotides are small organic compounds that serve as the "building blocks" for long chain molecules known as ________________________________. Explain two other functions of nucleotides. __________________________________________________________

(3) 20. The nucleic acids, DNA and RNA are long chain molecules made up of smaller units called ________________________________. These small units are also important to cell function in that they serve as chemical messengers and as ________________________ essential to metabolism. They may also take on extra phosphate groups to become high energy compounds, e.g., __________.

(5) 21. Matching - Important compounds: Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

| _____ Monosaccharides | A. Used to make high energy compounds, coenzymes and chemical messengers. |
| _____ Nucleic acids   | B. Composed of three polysaccharide chains attached to a glycerol "backbone". |
| _____ Proteins        | C. Amphipathic (amphiphilic) molecules associated with cell membranes. |
| _____ Polysaccharides | D. Two or more polypeptide chains coming together to form a single protein. |
| _____ Disaccharides   | E. Glucose, fructose, galactose, ribose, arabinose, and deoxyribose. |
| _____ Nucleotides     | F. Composed of three fatty acid chains attached to a glycerol "backbone". |
| _____ Phospholipids   | G. Long chain molecules made up of amino acids connected by peptide bonds. |
| _____ Anions          | H. Positively charged particle of matter. |
| _____ Quaternary structure | I. Lactose, sucrose and maltose |
| _____ Triglycerides   | J. Negatively charged particle of matter. |
|                     | K. Glycogen, cellulose, chitin and starch. |
|                     | L. Made up of many nucleotides joined together by phosphodiester bonds. |
**Cell Membrane Structure and Function:**

(5) 1. Define:

- Plasma membrane
- Amphipathic (amphiphilic)
- Osmosis
- Phototaxis
- Quorum sensing

(1) 2. The living dynamic layer that surrounds and limits a cell is called the ________________, and is so thin that it is not visible with a light microscope.

(2) 3. According to the Singer-Nicholson model, the chemical composition of eukaryotic cellular membranes is about 50% _____________________ and 50% _____________________. The membranes of bacteria are similar, but contain these components in a 40:60 ratio.

(3) 4. Proteins that extend through the cell membrane (contact the surface on both sides) are called ____________________ proteins and may serve a variety of functions within the membrane. Some of these proteins pump ions such as _____________________ across the membrane while others serve as carrier molecules in a passive transport process called _____________________.

(2) 5. About 65% of the lipids associated with eukaryotic cell membranes are amphiphilic/amphipathic molecules called _____________________. The polar phosphate groups associated with these molecules contact the watery environments outside and inside the cell while the non-polar _____________________ “tails” form the lipid bilayer surrounding the cytoplasm.

(2) 6. Small molecules such as oxygen and carbon dioxide can move into or out of cells by means of a passive transport process called _____________________. This process involves the movement of materials "down" their _______________________ gradients.

(2) 7. Water can move into or out of cells by means a passive transport process called _____________________. If cells are placed into an environment that has a high solute concentration (5% NaCl), water will move out of the cells. Such an environment is said to be _____________________.


8. Cells placed into a hypotonic environment would tend to ______________ water via a passive transport process called osmosis.

9. Active transport differs from facilitated diffusion in that it requires energy and can be used to move particles (molecules and/or ions) against ___________________________ gradients.

10. If two different types of material are being transported across a cell membrane in different directions by the same protein complex) they are said to be moving in _________________.

11. Explain briefly how facilitated diffusion differs from active transport.

12. Eukaryotic cells such as protozoa are able to take in large particles (macromolecules and other cells) via an active transport process known as ___________________________ or “cell eating” (please be specific). Cells can also take in smaller particles (not visible with a light microscope) by means of a process called ___________________________ or “cell drinking”.

13. The directed movement of a protozoan toward a chemical (food) source is technically referred to as ___________________________ (be specific).

14. Bacteria that move away from a light source would be said to display ___________________________ (be specific).

15. Bacteria respond to chemical signals released by other bacteria through a regulatory mechanism called ___________________________. When membrane receptors bind with chemical signals, they trigger the activation of specific ___________________________ resulting in cellular responses, e.g., bioluminescence, the release of digestive enzymes, and increased virulence.

16. The cell membranes of typical bacteria differ from eukaryotic cell membranes in that they contain little or no ___________________________ and are involved in the synthesis of materials such as wall components and ___________________________. Note - the cytoplasmic membranes of archaea contain branched, isophrenoid, ether-linked lipids rather than fatty-acid, ester-linked lipids, so are unlike the membranes of other cells.
Eukaryotic and Prokaryotic cells:

(5) 1. Define:

   Organelle
   Ribosome
   Histone
   Glycocalyx
   Sporulation

(1) 2. ________________________________ may be defined as the science or study of cells.

(2) 3. Eukaryotic cells typically contain a large number of membrane bound structures called _______________________________ that carry out specific functions and are supported by a fluid-like material called the ________________________________.

(2) 4. A complex membranous organelle that serves as a transport system, a site for storage and for lipid synthesis is called the ________________________________. Sometimes this structure is made "rough" by the presence of small granular bodies called ribosomes. These serve as the site for ____________________________ synthesis within the cell.

(3) 5. A membranous organelle known as the ________________________________ is involved in storage, the synthesis of polysaccharides, and the assembly of complex organic compounds (glycoproteins, glycolipids, etc.). What other functions are associated with this organelle? __________________________________________________________________________

(3) 6. Most microbiologists believe the organelles called ________________________________ and ________________________________ were originally prokaryotic cells that were taken in by, and formed symbiotic relationships with larger eukaryotic cells. What evidence is available to support this theory? __________________________________________________________________________

(3) 7. Almost all types of eukaryotic cells carry organelles called ________________________________ that evolved from prokaryotic organisms (Proteobacteria), acquired through endocytosis. These organelles have inner folded membranes called cristae, that carry enzymes involved in _______ synthesis (something eukaryotic membranes cannot do), lack cholesterol, and have a 60:40 protein to lipid ratio. These organelles have additional features characteristic of prokaryotic cells; what are they? __________________________________________________________________________
8. Both mitochondria and chloroplasts contain inner membranes that are involved in ATP synthesis. In mitochondria these inner membranes form folds called ____________________, while in chloroplasts they are arranged in flat, membranous vesicles called ________________.

9. Single-celled organisms digest the "food" materials they consume by bringing them into contact with digestive enzymes called _________________________. These enzymes are stored within organelles called _______________________. Organelles containing enzymes involved in hydrogen peroxide metabolism are referred to as _________________.

10. Many types of fresh-water protozoa contain osmoregulatory organelles called ______________ ___________________. These structures serve to pump excess water out of cells, but also play a role in intracellular circulation and excretion.

11. Most eukaryotic cells contain many tubular protein complexes called ______________________ that provide structural support and allow for intracellular motion. These tubular structures appear to be arranged in a characteristic pattern (nine groups of two plus an additional two in the middle) within locomotor structures called ______________ and ________________.

12. Microtubules are made up of proteins called ________________________ arranged in linear strands (protofilaments) that can be taken apart or assembled as needed. Microtubule associated proteins such as ______________________ can move materials along these strands and play a major role in intracellular transport.

13. Some eukaryotic cells contain bodies called ______________ ___________________ that are made up of microtubules arranged in nine groups of three. These give rise to spindle fibers and aid in chromosome separation during mitosis and meiosis.

14. All single-celled eukaryotic microorganisms contain one or more centrally located structures called ________________________ that serve to control all cellular activities (they are sometimes referred to as the "brains" of these cells). The protoplasm within these structures is largely composed of a thread-like material called ________________________ that is made up of ____________________ and protein.

15. A dark staining body within the nucleus is known as the ________________________ and serves as the site of _______________________________ synthesis, and the assembly of 60s and 40s subunits.

16. The cells of most "plant-like" microorganisms (algae and fungi) are surrounded by rigid layers of non-living material known as _________________________. What functions do these layers of material serve? ______________ ___________________

17. Describe two ways (other than size) in which Eukaryotic cells differ from prokaryotic cells.

18. Many microorganisms (both eukaryotic and prokaryotic cells) are motile by means of locomotor structures called _________________________. In eukaryotic cells, these structures are surrounded by membrane, contain a cytoskeleton made of ________________________, and move with a whip-like motion.
(2) 19. Prokaryotic cells that have flagella distributed fairly evenly all over their cell surfaces are said to have a ____________________________ flagellar arrangement. If a cell has flagella located at both ends it is said to have a polar or ____________________________ flagellar arrangement.

(3) 20. Describe three ways in which the flagella of prokaryotic cells differ from the flagella of eukaryotic cells.

(1) 21. Many types of Gram negative bacteria are covered with thin, hair-like structures called ___________________________ that serve to attach the bacteria to various surfaces including other cells.

(1) 22. Gram negative bacteria such as Escherichia coli sometimes produce one or more thin, tube-like structures called ___________________________ from proteins called pilins. Cells making these structures can attach to other cells, then pull them close for the cell-to-cell transfer of DNA.

(1) 23. Structures known as ___________________________ or as periplasmic flagella occur within the periplasmic space of spirochete bacteria.

(10) 24. Matching - Cell Structure and Function:
Match the term or terms on the left with the most appropriate functions on the right, and indicate in the second blank provided whether the structure is most commonly found in association with eukaryotic cells, prokaryotic cells, or both.

<table>
<thead>
<tr>
<th>Term</th>
<th>Function</th>
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<tr>
<td>Histones</td>
<td>A. Contain enzymes involved in hydrogen peroxide metabolism.</td>
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<tr>
<td>Ribosomes</td>
<td>B. Made of protein pilin, allow for attachment and genetic exchange.</td>
</tr>
<tr>
<td>Cell walls</td>
<td>C. Homogeneous proteins associated with linear strands of chromatin.</td>
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<tr>
<td>Glycocalyx</td>
<td>D. Reservoir of stored food which aids in attachment and pathogenicity.</td>
</tr>
<tr>
<td>Thylakoids</td>
<td>E. Extrachromosomal loops of DNA carrying genes not essential to cell function.</td>
</tr>
<tr>
<td>Peroxisomes</td>
<td>F. Composed of RNA and protein, are the site of all protein synthesis.</td>
</tr>
<tr>
<td>Nucleoid</td>
<td>G. A non-living extracellular layer which provides support and protection.</td>
</tr>
<tr>
<td>Golgi complex</td>
<td>H. Primary site of packaging, secretion, and polysaccharide synthesis.</td>
</tr>
<tr>
<td>Pili</td>
<td>I. Not bounded by a membrane, contains one circular chromosome.</td>
</tr>
<tr>
<td>Plasmids</td>
<td>J. Membranous structures with enzymes for ATP synthesis via photophosphorylation.</td>
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(1) 25. The rigid cell walls of typical Gram positive and Gram negative bacteria are composed of a unique organic material known as ___________________________. In Gram positive cells this layer is quite thick, while in Gram negative cells it is thin.
26. A layer of organic material (glycocalyx) found outside the cell wall of a prokaryotic cell may be a dense, well-organized _______________________ or a loosely organized ______________________. This layer is produced only under certain circumstances and may serve a variety of functions. It may aid in attachment and in pathogenicity, may protect the cell against dehydration, and serves as a _____________________________.

27. A ____________________________ is a layer of material found outside the prokaryotic cell wall, and is usually composed of protein or polysaccharide. If this layer is dense and well organized it is called a capsule and if it is lose it is called a slime layer. What functions are associated with this layer? _____________________________.

28. Gram positive bacteria have thicker cell walls and are generally more resistant to ________________ than are ordinary Gram negative bacteria. The Gram negative cells have an outer membrane associated with their cell wall and so are more resistant to ________________ than are Gram positive cells.

29. The cell membranes of some prokaryotic cells appear to fold inward forming structures known as ___________________________. Like the cristae of mitochondria, these would contain enzymes involved in phosphorylation. Most investigators now believe these membrane folds are actually artifacts formed during the preparation of cells for electron microscopy.

30. Cyanobacteria are photosynthetic prokaryotes that contain membranous vesicles called ___________________________________. These are usually located parallel to the cell membrane, but appear to be separate from it. They contain enzymes involved in the formation of ________________ via photophosphorylation.

31. The cytoplasmic region of a prokaryotic cell contains no membrane bound organelles, but does contain ribosomes and a variety of inclusions. ____________________________ are inclusions which contain enzymes involved in "fixing" carbon dioxide into organic compounds.

32. Some bacteria contain cytoplasmic inclusions called ___________________________ that serve as a source of intracellular phosphate (a sort of energy reserve). Bacteria living in water environments often contain inclusions called gas vacuoles. What function do these serve?

33. The chromosome of a prokaryotic cell is not contained within a nucleus with a nuclear membrane, but is contained within a region known as the nuclear region (area, body) or ___________________________. Many bacteria also contain one or more small extrachromosomal loops of DNA known as ___________________________. These generally carry genetic information that is not essential to the survival of the cell under most circumstances.

34. Some types of Gram positive bacteria produce dormant structures called _______________________ that allow these cells to survive periods of unfavorable conditions. These dormant structures are highly resistant to environmental factors such as ___________________________ and may live in a nearly inactive state for hundreds or thousands of years.
(2) 35. Explain how endospores differ from vegetative cells in terms of their composition and function. Include information about nucleic acids, water, calcium, dipicolinic acid, resistance to environmental factors, metabolic activity levels and longevity.

(2) 36. Bacteria in the genera *Bacillus* and *Clostridium* often form dormant structures known as __________________ . These contain high levels of DNA, calcium and dipicolinic acid but very little RNA and almost no water. They are metabolically inactive and resistant to both physical and chemical damage. What is the overall function of these structures?

(1) 37. Exospories undergo a process called ___________________________ when they find an environment suitable for their growth, and have been subjected to the proper "triggering" stimuli.

(2) 38. Cyanobacteria such as *Anabaena* often form specialized cell types that carry out specific functions. Specialized cells called ___________________________ contain enzymes involved in nitrogen fixation, while thick-walled, granular-looking cells called ___________________________ are highly resistant to cold and desiccation thus allowing bacteria to survive cold winters and/or dry summers.

**Taxonomy and Classification:**

(5) 1. Define:

   Taxonomy

   Binomial nomenclature

   Psychrophile

   Photoheterotroph

   Microaerophile

(3) 2. ___________________________ may be defined as the science or study of biological classification, and provides a system for arranging living organisms into specific groups or categories. According to the five kingdom system proposed in 1969 by R.H. Whittaker, all prokaryotic organisms belong within the kingdom ___________________________. Why is this classification no longer considered valid? ___________________________
3. According to the binomial system of nomenclature as developed by Carolus Linnaeus (1735), the scientific name of an organism is composed of the _____________________________ and the _____________________________ names of that organism.

4. A taxonomic rank that is less encompassing than a class but more encompassing than a family would be known as a/an _____________________________.

5. The five kingdom system of classification was proposed in 1969 by ______________________. According to this system, all single-celled eukaryotic organisms such as algae and protozoa belong within the kingdom _____________________________.

6. Features such as size, shape, arrangement, color, etc. are generally referred to as _____________________________ characteristics, and are more useful in the identification of multicellular organisms than they are in the identification of bacteria.

7. Organisms which can obtain their energy from chemicals, but require organic compounds for their source of carbon are nutritionally categorized as _____________________________. If these organisms can only grow and reproduce while inside another living cell they are said to be _____________________________ or obligate intracellular parasites.

8. Algae are nutritionally categorized as _____________________________ because they can obtain energy from light, and use inorganic compounds as sources of carbon.

9. Chemoheterotrophs that live on or inside other organisms and obtain their carbon and energy from these organisms are referred to as _____________________________. Most fungi obtain their energy and carbon from dead and decaying organic materials and so are referred to as _____________________________.

10. Many types of bacteria are able to obtain both carbon and energy from inorganic chemicals. Organisms with this ability are nutritionally categorized as ____________________________ and often live in unusual environments.

11. Organisms that use inorganic compounds (usually oxygen) as their final electron acceptors are said to have a _____________________________ type metabolism.

12. An organism capable of growing with or without oxygen in its environment is said to be a _____________________________ organism and may use either a respiratory or a _____________________________ type metabolism.

13. Many types of bacteria can grow and reproduce only in environments containing very little or no oxygen. These organisms are referred to as _____________________________ and may be damaged or killed by exposure to molecular oxygen.

14. _____________________________ are organisms that grow best in cold environments (where temperatures range between -5 and 20 degrees C.). The term _____________________________ applies to organisms that can tolerate or endure very cold temperatures. These organisms are not able to grow at such cold temperatures, but they are not killed by them either. Most bacteria have this characteristic.
15. Organisms that grow best in hot environments (those with temperatures above 60°C) are referred to as _______________________________. Although most bacteria in the genera *Bacillus* and *Clostridium* do not fit within this category, they can survive exposure to high temperatures because they form endospores that are __________________________________.

16. The pH of an environment is a measure of what?

17. Chemical substances called ___________________________ change color in response to shifts in the acidity or alkalinity of their environment, and are often added to media as a means of monitoring the metabolic activity of microorganisms.

18. An organism is said to be an extreme _______________________________ if it can grow and reproduce only in extremely salty environments.

19. When two or more different types of organisms are found living together in a close association, they may be said to form a _______________________________ relationship. For example, many bacteria live on and inside our bodies and help us survive in our environment. Only a few types of bacteria cause disease and so are considered ________________________________.

20. In our microbiology laboratory we commonly use a type of biochemical analysis known as _______________________________ when attempting to identify various different types of bacteria. This method involves growing organisms on a variety of different media containing pH indicators, and sometimes exposing them to chemical reagents.

21. Two techniques used in the identification of unknown bacteria include _______________________________ (a method which involves mixing known antibodies with the bacteria) and phage typing, a method which involves mixing known types of ______________________ with unknown bacteria.

22. Bacteria are recognized as specific serotypes or serovars if they have been identified by being mixed with specific types of _______________________________. Bacteria can also be identified by mixing them with known types of viruses in a technique known as ________________________________.

23. Gel-electrophoresis, percent base composition (C+G and A+T), hybridization, “finger-printing” and nucleotide sequencing are all techniques used in the biochemical analysis of ____________.

24. A classification method that groups organisms based on unique but shared characteristics from a last common ancestor is called _______________________________, and can be used to generate tree-like, branching diagrams (cladograms) indicating the _________________________________ or evolutionary history of the organisms being studied.

25. ________________________________ is the study of the evolutionary relationships among groups of organisms as determined through molecular sequencing (e.g., 16S r-RNA genes) data and comparison of morphological features.

26. What is the human genome project?
Introduction to Archaea and Bacteria:

1. Define:
   - Archaea
   - Bacteriorhodopsin
   - Luciferase
   - Leghemoglobin
   - Anoxygenic

2. Prokaryotic organisms in the domain __________________________ are unlike bacteria in that their cell membranes contain unique lipids (mirror image glycerol molecules with ether linkages to isoprenoids), and their cell walls never contain ________________________________, a substance common within bacterial cell walls.

3. Although the Archaea were once considered ancient bacteria, they are now recognized as unique organisms that differ from bacteria more than humans differ from pine trees. Explain three ways in which Archaea differ from Bacteria.

4. Organisms categorized as ___________________________ Archaea produce methane using carbon dioxide and hydrogen. The methane generated by these organisms is potentially useful to humans. Archaea identified as Halobacterium salinarium (formerly H. halobium) are phototrophs that trap light energy using a pigment called ______________________________ that is similar to one of the pigments found in the human retina.

5. Some bacteria in the phylum __________________________ are polyextremophiles capable of surviving exposure to heat, pressure, desiccation and ionizing radiation; others are hyperthermophiles recognized as the source of enzymes most commonly used in the PCR.

6. Oxygenic photoautotrophs in the phylum __________________________ are sometimes called blue-green algae, but are actually bacteria. These organisms often form specialized cells called akinetes and heterocysts, and some of them form cyanotoxins called ________________ that if ingested can cause severe liver damage.

7. Bacteria in the genus __________________________ are Gram-negative parasites of insects, roundworms and other organisms that often cause reproductive changes in their hosts. These may provide a means for controlling insect vectors of important human diseases, but can also cause severe inflammation in humans infected by roundworms.
8. Although leguminous plants such as beans, clover and alfalfa are often credited with the ability to add nitrogen to soil, this activity is actually accomplished by soil bacteria in the genus ___________________________ that form symbiotic relationships with the plant roots. Many types of deep sea fish and squid are said to be bioluminescent, when actually the ability to produce light is associated with bacteria living on them. Bacteria in the genus ___________________________ are able to form a type of enzyme called ____________________________ that allows them to convert chemical energy into light energy, and so to produce light.

9. Aquatic bacteria in the genus ___________________________ form sheaths that resemble underwater spider webs and block water flow in well pumps, fishtank filters and slow moving streams. Soil bacteria in the genus ___________________________ produce sulfuric acid as a metabolic by-product, and are used in mining and to remove sulfur from coal.

10. Bacteria in the genus *Pseudomonas* are unique in that they can utilize a wide range of unusual organic compounds as "food" materials, and sometimes carry surface proteins that promote ice crystal formation. Explain how humans use these unique bacteria for our own purposes. ______________________________________________________________

11. Bacteria are said to be ___________________________ if they can produce their own light. The ability to convert chemical energy into light energy involves enzymes coded for by a gene known as the _____________________________.

12. ___________________________ is a type of Gram negative rod-shaped bacteria which is used extensively in genetic research, serves as an indicator in the bacteriological examination of water, and is known to be a significant pathogen (especially when consumed in raw or poorly cooked ground beef).

13. Bacteria in the genus ___________________________ are unique in that they attack and kill other types of Gram-negative bacteria. These comma-shaped cells drill through the host cell wall, reside within the periplasmic space, and use host cell nutrients to grow and reproduce.

14. Tiny bacteria in the genus ___________________________ lack peptidoglycan in their cell walls, and tend to be pleomorphic (variable in shape). Because of their small size (0.1 to 0.25 μm in diameter), they often pass through filters designed to remove bacteria.

15. Endospore-forming bacteria within the genus ___________________________ are of interest to scientists because they often infect and kill insect larvae and so can be used to make biological insecticides.

16. Two genera of bacteria commonly used in the production of cheese, yogurt, and sour-dough bread products are ___________________________ and ___________________________. The lactic acid made by these organisms is a primary flavor component in such foods.

17. Gram positive, catalase positive, aerobic cocci that commonly form bright colored colonies (red, yellow, orange, pink) on air plates are classified within the genus ___________________________.
18. Gram-positive bacteria in the genus ____________________________ are unique in that they contain high levels of mycolic acid in their cell walls and are acid-fast. Some species in this genus are important human pathogens.

19. Soil bacteria in the genus *Streptomyces* are currently the focus of extensive investigation since they have been found to be an important source of __________________________________. Organisms in this genus also produce ______________________ that give soil a characteristic earthy odor.

20. Bacteria in the genus _________________________ are hypotrophs with limited ability to produce ATP. Some are recognized as the etiological agents for one of the most common sexually transmitted diseases known.

21. The _______________________________ are thin, spiral-shaped bacteria with flexible cell walls and axial filaments (periplasmic flagella). Some organisms in this group are important human pathogens.

22. Match the bacteria on the left with the most appropriate characteristics on the right and place the letters of correct matches in the blanks provided.

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   Mycobacterium   | A. Nitrogen "fixing" bacteria that live in root nodules of leguminous plants.
 ___ Spirochetes  | B. Oxygenic phototrophic bacteria initially thought to be algae; some form toxic microcystins.
 ___ Clostridium  | C. Thin spirilla with flexible cell walls and axial filaments or periplasmic flagella.
 ___ Bdellovibrio | D. Soil bacteria that produce sulfuric acid used in mining operations and to remove sulfur from coal.
 ___ Mycoplasma   | E. Gram positive, catalase negative, cocci often used in food processing; some are important pathogens.
 ___ Cyanobacteria| F. Bacteria unique in their ability to use unusual organic compounds for carbon, used in bioremediation.
 ___ Rhizobium    | G. Acid-fast bacteria with high levels of mycolic acid in their cell walls. Some are important human pathogens.
 ___ Streptococcus| H. Gram negative bacteria that are known to attack and kill other bacteria.
 ___ Thiobacillus | I. Gram positive, endospore-forming soil bacteria that are anaerobic, some are important human pathogens.
 ___ Pseudomonas  | J. Tiny bacteria (0.1 to 0.25μm in diameter), that have no peptidoglycan in their cell walls.
Introduction to Fungi:

(5) 1. Define:

   Mycology

   Hyphae

   Karyogamy

   Dermatophytes

   Aflatoxin

(1) 2. ____________________________ may be defined as the science or study of fungi.

(2) 3. In the five kingdom system of classification, fungi are not classified as plants (kingdom Plantae) because they are ____________________________ (i.e., lack green pigments) and have a ____________________________ mode of nutrition.

(1) 4. Organisms in the kingdom Fungi (Mycetae) are achlorophyllous chemoheterotrophs that are made up of ____________________________ type cells, and that have cell walls containing cellulose, chitin, glucan or a combination of these polysaccharides.

(2) 5. The thallus of a mold-type fungus is made up of many thin thread-like filaments called ____________________________ that come together to form visible mats called mycelia. If the fungus is parasitic, and the filaments are specialized to penetrate host cells and absorb nutrients, they are referred to as ____________________________ .

(2) 6. The thallus of a mold-type fungus is made up of many thin thread-like filaments called hyphae that come together to form a mat-like structure called a ____________________________ . Hyphae may be septate or aseptate; but in either case, the cells involved are all connected, and so form a true ____________________________ or multinucleated mass of protoplasm.

(1) 7. Many types of fungi form symbiotic relationships with plant roots. Some fungi form specialized hyphae called ____________________________ that actually help plants to absorb water and minerals (e.g., phosphorous), from the soil. These fungi are essential to forest ecosystems.

(1) 8. Yeast type fungi typically reproduce asexually via a process called ____________________________ during which there is uneven division of the cytoplasm, one "daughter" cell being much smaller than the other.
9. That portion of a mold-type fungus that extends above the substrate upon which the fungus is growing (agar, soil, rotting wood, etc.) is referred to as the ___________________ and is involved in reproduction (either sexual or asexual). The portion of the fungus that extends into the substrate is called the ___________________ and is involved in food getting.

10. Fungi often reproduce asexually by budding, fragmentation, or by producing numerous reproductive structures called spores. If these spores are contained within a sac-like structure at the end of a filament, they are called ___________________.

11. The sexual reproductive process of fungi involves three steps or stages. The stage involving the union or joining of protoplasm is called ___________________ while the one allowing each diploid zygote to divide into four haploid cells is called ___________________.

12. Sexual reproduction as it occurs in fungi involves three stages or steps. The stage allowing nuclei to join forming a diploid zygote is called ___________________, while the stage allowing the zygote to return to the haploid state is called ___________________.

13. Explain two ways in which fungi are considered to be of value or beneficial to humans.

14. Fungi are used industrially in the production of food, antibiotics, ___________________, and a variety of ___________________.

15. Although some forms of fungi are the causative agents of ___________________ (fungal induced diseases) in man and other animals, others serve as a source of ___________________ (such as penicillin) that are used to treat disease.

16. ___________________ may be defined as infections or diseases that are caused by fungi. If these diseases involve the deep tissues of the body (blood, lungs, lymphatics, etc.) they are said to be deep or ___________________ infections and typically involve fungi that enter their host by ___________________.

17. Two factors that have contributed to an increase in fungal induced diseases in people living in the US during recent years are:
   a) ___________________
   b) ___________________

18. The primary factor determining if or not an individual will be likely to develop a serious mycosis after being exposed to potentially pathogenic fungi is ___________________.

19. Name two genera of fungi that are known to cause deep or systemic mycoses (you may include opportunistic pathogens).
   a) ___________________
   b) ___________________
(2) 20. Superficial fungal infections are often caused by fungi in the genera *Epidermophyton*, *Trichophyton* and *Microsporum*. These fungi are collectively referred to as the ____________________________ and cause a variety of infections including __________________________________________________________________________.

(2) 21. A soil fungus in the genus ____________________________ may cause deep or systemic infection in people with compromised immune function. These fungi typically enter the body via ____________________________________________________. (More than one answer is possible in the first blank above.)

(1) 22. ____________________________ is the name given to a mycotoxin produced by *Aspergillus flavus*.

(2) 23. Two genera of fungi known to produce mycotoxins that can cause severe illness or death when consumed by humans are ____________________________ and ____________________________.

(1) 24. An opportunistic pathogen known to cause the formation of fungus balls in the lungs of compromised individuals is classified within the genus ____________________________.

(4) 25. Matching, Fungi - Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

_____ Dermatophytes  A. Soil fungus that causes San Joaquin Valley fever or desert rheumatism.

_____ Tinea corporis  B. Pathogenic fungus often associated with soil rich in bird or bat feces.

_____ Chromomycosis  C. Genus of yeast-like fungi known to be a pathogen in immunocompromised individuals.

_____ Coccidioides  D. Recognized as the source of aflatoxin and sometimes a respiratory pathogen.

_____ Cryptococcus  E. Fleshy fungus known to produce a mycotoxin that sometimes causes fatal intoxication in humans.

_____ Histoplasma  F. Superficial mycosis sometimes caused by fungi in the genus *Epidermophyton*.

_____ Aspergillus  G. A subcutaneous mycosis caused by various pigmented fungi called black molds.

_____ Amanita  H. Group of fungi known to be the cause of superficial mycoses.
Introduction to Algae:

(5) 1. Define:

Phycology

Endophytic

Eutrophication

Paralytic shellfish poisoning

Diatomaceous earth

(1) 2. __________________________________ may be defined as the science or study of algae, and in this class will be restricted to microscopic varieties.

(2) 3. Algae contain light trapping pigments and use inorganic carbon (carbon dioxide) to form organic compounds, so are nutritionally categorized as ___________________________.
Algae are beneficial to other organisms in that they produce __________________________ and serve as a source of __________________________________________.

(1) 4. Algae (probably with the help of certain bacteria) are thought to produce as much as 70% of the ___________________________________________ on this planet.

(2) 5. Although algae are very important environmentally in that they are the primary source of ____________________________________ present in our atmosphere, they can also cause problems to fish by using up this substance at night (when no light is available) if water bodies have undergone ____________________________.

(2) 6. All Algae contain green-colored pigments called ____________________________, but these are sometimes masked by red-colored pigments called ____________________________.

(1) 7. Many types of single-celled algae contain yellow or orange-colored pigments called ____________________________ and so appear golden or brownish rather than green.

(1) 8. Filamentous algae typically reproduce asexually via a process known as __________________________.

(2) 9. Sexual reproduction as it occurs in algae (like fungi) involves three stages or steps. The joining of the nuclei is called ____________________________ and results in the formation of a diploid cell or zygote. In algae, these diploid cells make up what is known as the ____________________________ generation, and are morphologically indistinguishable from haploid cells.
(2) 10. ___________________________ may be defined as an increase in algae population within a body of water. Although this may be considered beneficial in the sense that algae provide food for various other organisms, it may also be considered detrimental. Why? ___________________________

(2) 11. Certain types of green algae live symbiotically with fungi in organisms known as ___________________________. Other types of algae live inside organisms such as anemones, flatworms and protozoa and so are said to be ___________________________.

(1) 12. Although algae are not usually considered pathogens, toxins produced by Alexandrium and Gymnodinium can cause a disease called ___________________________ which causes muscle paralysis in humans and other animals.

(1) 13. The glass cell walls of algae known as ___________________________ are used as a filter material, as insulation, as an abrasive in cleansers, and in reflective paint.

(1) 14. Why are algae in the phylum Rhodophyta (Rhodophycophyta) important to bacteriologists?

(1) 15. The glass cell walls of algae called diatoms are used in a variety of ways including:

________________________________________________________________________

(1) 16. Although algae are usually photoautotrophs, those in the genus ___________________________ sometimes kill and feed on fish. These algae also produce toxins that can cause neurological symptoms (dizziness, memory loss, etc.), in humans.

(1) 17. Bioluminescent algae in the genus ___________________________ can cause ocean waves to light up, and sparkle under the feet of people running along wet ocean beaches.

(3) 18. Matching, Algae - Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

   ___ Alexandrium   A. Associated with harmful algae blooms, these can kill fish, and may produce toxins harmful to humans.
   ___ Diatoms       B. Filamentous green algae common in freshwater streams locally, these have spiral chloroplasts.
   ___ Noctiluca     C. Endophytic algae known to live within marine animals such as giant green sea anemones.
   ___ Pfiesteria    D. Formerly called Gonyaulax, these can form saxitoxin and can cause paralytic shellfish poisoning.
   ___ Spirogyra    E. A common form of phytoplankton, these have glass cell walls that form diatomaceous earth.
   ___ Zoochlorella F. Bioluminescent algae that cause waves to light up and ships to leave tracks through the sea.
Introduction to Protozoa:

(5) 1. Define:

Cirri

Trophozoite

Cytostome

Contractile vacuole

Schizogony

(2) 2. Most protozoa, like fungi, are eukaryotic organisms that can be nutritionally categorized as ______________________________ because they obtain their carbon and energy from preformed organic compounds. Some protozoa are parasites, a few cause disease in humans and other animals, so are important as ______________________________; but most are free-living organisms. Some protozoa function as photoautotrophs.

(1) 3. Amoeba-like protozoa move about by extending portions of their protoplasm called ______________________________ (false feet) into the environment, and flowing along after them.

(3) 4. Protozoa have a number of specializations that allow them to move about and obtain food in fresh-water environments. Amoeba-like protozoa have locomotor structures called ______________________________ that can be extended out and around prey organisms during phagocytosis. Organisms such as Paramecium swim through water by means of short, hair-like structures called ______________________________. These may also be arranged in tufts called ______________________________ and can be used for walking or jumping.

(2) 5. Protozoa often possess structures which serve both for locomotion and for food getting. Two examples of such dual-purpose structures are ______________________________ and ______________________________.

(2) 6. Most protozoa are chemoheterotrophs, and many of them are predators. Protozoa that catch and consume whole organisms as their prey are said to have a ______________________________ type of nutrition. Food materials are often taken into these cells via a specific site known as the ______________________________ or cell mouth.

(1) 7. Fresh water protozoa usually contain osmoregulatory organelles known as ______________________________ that pump excess water out of their cells.
8. Several types of protozoa are intestinal pathogens, and enter their host in their inactive or ______________ stage because these are resistant to the acidic conditions of the stomach. Once they reach the intestine, the protozoa emerge as __________________________, the active (feeding, growing, reproducing, etc.) form of the parasite.

9. Many protozoa produce structures that can be used for protection or for defense. The outer surfaces of ciliated protozoa are protected by a tough, flexible layer called a ____________________________, radiolarians have shells or external skeletons made of glass and foraminiferans live inside tests made of calcium carbonate. Some ciliates such as Paramecium form and use dart-like protective structures called __________________________ that can be shot out in response to certain stimuli.

10. Most protozoa reproduce asexually by undergoing __________________________, although some protozoa form buds, and some undergo schizogony. Sexual reproduction most commonly involves __________________________ a process that involves the exchange of genetic material between two cells or __________________________, a process that results in the formation of a diploid zygote.

11. Matching, medical protozoology – Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

   ___ Toxoplasma   A. Ciliated gut parasite capable of causing dysentery in humans and other animals.
   ___ Entamoeba   B. Vaginal flagellates that may be acquired through sexual contact or poorly maintained pools and hot tubs
   ___ Trypanosoma   C. Locally encountered gut parasites acquired by drinking contaminated water; these lack mitochondria.
   ___ Balanididium   D. Hemoflagellates recognized as the causative agents of African sleeping sickness and Chaga’s disease.
   ___ Trichomonas   E. Enter their host as sporozoites, exit RBCs as merozoites and enter mosquitoes as gametocytes.
   ___ Plasmodium   F. Live in soil or hot springs; enter hosts via nasal passages and cause meningoencephalitis.
   ___ Giardia   G. Sporozoans that can cause severe nervous system damage in fetuses and cause rodents to love cats.
   ___ Naegleria   H. Cysts ingested with water or vegetable materials carry four trophozoites that cause gut lesions and dysentery.

12. Protozoa can enter into a dormant stage known as the __________________________ stage in order to survive periods of unfavorable conditions, such as hot dry summer weather, or the acid conditions of a host's stomach.

13. Protozoa in the genus Plasmodium reproduce sexually via a process called ______________ while within their mosquito host, and reproduce asexually via __________________________ or multiple fission while within their mammalian host.

14. Two types of human diseases that can be caused by amoeba-like protozoa are __________________________ and __________________________.
15. Although the hemoflagellates that cause African sleeping sickness and Chaga’s disease occur only in tropical areas, there are some flagellated parasites that occur locally. Intestinal parasites identified as __________________________ are common in rivers, streams and lakes in Placer County, while a vaginal parasite called __________________________ may be acquired by swimming or bathing in pools or hot tubs that are not properly maintained.

16. *Plasmodium* organisms enter their mammalian host while in their __________________________ stage and then travel to the deep tissues of the body (e.g., liver). After a period of time, they enter RBCs and reproduce asexually. Each infected RBC will eventually rupture and release many protozoa in their __________________________ stage. These infect and eventually destroy other RBCs.

17. Describe the life cycle of *Plasmodium vivax* as explained in lecture. Be sure to include the mechanisms of sexual and asexual reproduction, and where they occur.

18. Malaria, African sleeping sickness and Chaga’s disease are similar in that all are caused by protozoa that are transmitted from one host to another by __________________________.

19. Amoeba-like organisms in the genus __________________________ are now recognized as the most common cause of primary amoebic meningoencephalitis.

20. Mature cysts of __________________________ can each release eight trophozoites. These can feed on intestinal bacteria, but can also cause lesions in the gut wall resulting in tissue damage and __________________________. If amoebas enter the blood stream, liver abscess and damage to other internal organs can occur.
**Introduction to Multicellular Parasites:**

1. Define:
   - Helminth
   - Definitive host
   - Redia
   - Vector
   - Monoecious

2. Multicellular endoparasites (flatworms and roundworms) are commonly referred to as ______________________ (a term meaning worms) and are adapted to a life within the gut, bloodstream, or tissues of another organism. One of the characteristics common to these organisms is a highly developed ______________________ system.

3. Organisms within the phylum Platyhelminthes have very poorly developed ______________________ and ______________________ systems, but often have both male and female reproductive systems so are said to be ______________________ or hermaphroditic.

4. Many helminths have complex life cycles that involve more than one host. The host in which the adult form of a parasite is found is called the ______________________ host, while the host in which the larval parasites are found is called the ______________________ host.

5. Many helminth parasites have complex life cycles that allow their offspring or larvae to live in an environment or host organism away from the parents. What are two advantages of this arrangement? __________________________________________________________

6. Describe the life cycle of *Fasciola hepatica* as presented in lecture and explain why having alternate hosts is an advantage to this organism.

7. Each egg of the sheep liver fluke, *Fasciola hepatica*, may hatch into a ciliated larval form called a ______________________. If the environmental conditions are appropriate, these larvae swim about in search of an intermediate host, which is a ______________________. Then they burrow in, undergo asexual reproduction, and eventually exit their host as tadpole-like fluke larvae called ______________________.

8. Blood flukes, *Schistosoma*, are important human pathogens in some parts of the world. Why is it hazardous to swim or wade in fresh-water lakes or rivers where blood flukes are prevalent?
(2)  9. Hookworms and filarial worms both belong to the phylum Aschelminthes (roundworms). Hookworms enter their hosts by ___________________________________________, while filarial worms enter via _______________________________________.

(2)  10. Two multicellular parasites which may be acquired by eating raw or poorly cooked pork are ____________________ and ____________________. (Common names are worth half credit here.)

(2)  11. Roundworm parasites identified as Trichinella spiralis and Necator americanus, can both use humans as their definitive hosts. In both cases the adult parasites live in the intestine. How do these parasites differ in terms of how their larvae reach a new host?

(4)  12. Describe the life cycle of the hookworm, Necator americanus, as presented in lecture.

(1)  13. Ectoparasites in the phylum Arthropoda are of interest to microbiologists because they often serve as ________________________________.

(1)  14. Multicellular organisms such as fleas, ticks, lice and mosquitoes are referred to as ________________________________ because they live outside their host.

(1)  15. The canine heartworm, Dirofilaria immitis, is an important parasite known to exist in this area. Why is it so important that dog owners treat their animals for this parasite?

(3)  16. Matching, multicellular parasites - Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

_____ Enterobius A. Female worms bare live larvae that enter the bloodstream and then burrow into striated muscles where they form cysts.

_____ Trichinella B. Larvae exit the bloodstream, enter the lungs, crawl up airways, tickle the pharynx, are coughed up and swallowed.

_____ Fasciola C. Intestinal parasites of pigs, cattle and other animals; adults can be several feet long and live for years.

_____ Schistosoma D. Eggs hatch into ciliated miracidia that enter water snails, form sporocysts, redia and cercaria that can burrow through the skin.

_____ Dirofilaria E. Adults reside in liver, mate and produce thousands of eggs, larvae form cysts on vegetation and are eaten by new hosts.

_____ Necator F. Adults collected in nodules under the skin, mate and release live larvae; these can enter the eye causing blindness.

_____ Taenia G. Filaria larvae of these parasites are transmitted by mosquitoes; adult worms fill heart and vessels blocking blood circulation.

_____ Onchocerca H. Eggs laid near anus itch, tiny fingers scratch, and spread eggs around. Eggs are ingested and adult worms inhabit intestines.
Microbial Nutrition and Growth:

(5) 1. Define:

- Enriched media
- Fastidious (microbe)
- Lag phase (microbial growth)
- Stationary phase (microbial growth)
- Carrying capacity

(2) 2. Most of the bacteria grown in our microbiology laboratory use preformed organic compounds for both carbon and energy so are nutritionally categorized as ___________________________. The mixture of materials that provides all the nutrients these organisms need to grow and reproduce is referred to as the ____________________________________.

(2) 3. Most bacteria reproduce asexually by means of _______________________________. What is the first thing bacteria must do in order to begin this process? ________________________

(1) 4. The term growth, when applied to a bacterial culture, refers to an increase in _______________ ________________ rather than to an increase in the size of a single organism.

(2) 5. Most bacteria reproduce via a process called ________________________________ during which one cell becomes two. The _________________________________ portion of this process requires that sections of the peptidoglycan wall be broken down and that new wall material be synthesized.

(1) 6. During the elongation phase of its reproductive cycle, a bacterial cell undergoes a number of changes. Describe the elongation process as it was explained in lecture. ________________ ________________ ________________

(1) 7. Bacterial cells placed into a container of sterile nutrient medium do not increase in number immediately, but enter into a phase known as the lag phase during which there is an increase in ___________________________________________ but not in cell number.

(1) 8. Bacterial cells may not undergo physical separation at the end of their fission process. If they don’t, and fission always occurs in the same plane, the cells will form an arrangement known as ___________ (c cocci or bacilli). What arrangement would cocci achieve if their fission plane was initially horizontal, and then vertical? ________________________
9. During the lag phase of its growth, an in vitro bacterial population does not increase or decrease in number, but does change in many other ways including: _____________________________
____________________________________________________________________________

10. Bacteria growing in vitro can reproduce exponentially only for a limited period of time. What causes an in vitro bacterial population to stabilize and then go into an exponential death phase?
___________________________________________________________________________

11. The maximum number of cells that can be supported in vitro is referred to as the m-concentration and is usually around _____________________ cells per ml. This number cannot be exceeded because the bacteria have reached the _____________________________
of their environment and one or more essential factors have become limited in supply.

12. The _____________________________ phase is a phase of bacterial growth during which the number of new cells being formed equals the number of cells dying. The bacterial population cannot exceed m-concentration because they have reached the _____________________________ of their environment, and factors critical to their survival have become limited.

13. Where in a bacterial colony would you expect to find the “youngest” cells and why? ______
____________________________________________________________________________

14. A batch culture of bacteria will eventually exit stationary phase and enter into an _____________________________ because the cells present have run out of nutrients and are being damaged by the build up of toxic metabolic waste products. It is possible to maintain bacteria in an exponential growth phase for an unlimited period time by doing what?
____________________________________________________________________________

15. The growth curve demonstrated by a bacterial population grown in a closed system has what implications relative to the human population on the planet earth?
**Microbial Metabolism, Enzymes and ATP:**

(5) 1. Define:

   - **Metabolism**
   - **Endergonic**
   - **Ribozyme**
   - **Constitutive enzyme**
   - **Oxidation**

(2) 2. _____________________________________ may be defined as energy transfer mechanisms occurring within living organisms, and is closely associated with metabolism. Chemical reactions that are anabolic (building reactions) require an input of energy (take up energy) and so are said to be ________________________________ while catabolic (breakdown) reactions give off more energy than is required to initiate them.

(3) 3. __________________________ may be defined as all the chemical reactions that occur within living organisms and includes both building and breakdown reactions. Different types of reaction are catalyzed by globular protein molecules known as ____________________ or by RNA molecules called ________________________.

(1) 4. Anabolic reactions that result in the formation of ATP (or other high energy phosphate compounds) are referred to as __________________________ reactions and may be categorized as substrate level, oxidative, or photo.

(2) 5. Chemical reactions that result in the release of energy (give off more energy than was required to get them started) are catabolic in nature and are called __________________________ reactions. The energy released during the catabolism of organic compounds such as carbohydrates is not used directly to drive cell processes, but is instead used to form high energy phosphate compounds such as ATP. Such reactions are called __________________________ reactions and may be categorized as substrate level, oxidative or photo.

(2) 6. Living cells use a type of energy “currency” known as ________________ to drive physiological processes such as flagellar movement, active transport and anabolic reactions. A single eukaryotic cell may use as many as 2 million of these molecules per second. What other types of high energy compounds may be found within living cells? __________________________

(1) 7. When an atom or molecule loses oxygen, or if it gains electrons and hydrogen protons, it is said to have been ________________________________. (Remember, LEO-GER)
8. A molecule is said to have been ______________________________ if it has gained oxygen or has given up one or more electrons and hydrogen protons.

9. According to the models presented in lecture, enzymes exert their influence by: 1) __________ _________________ and 2) _____________________________________________________________________.

10. Globular proteins that serve as biochemical catalysts and increase the rate at which chemical reactions occur within living cells are called ______________________________. The function of these molecules can be influenced by a variety of factors including _________________, _________________, __________ and concentration.

11. Both enzymes and ribozymes are molecules that serve as biochemical catalysts. Their overall function is to ___________________________________________________________________________________________________________________________________________. These catalysts are usually very specific in their action, but are not changed by the reactions they catalyze, so can be _________________________________________________________________________________________.

12. Enzymes that are functional as proteins alone are called simple enzymes, but if they require some type of “helper” group, they are called ______________________________ enzymes. Inorganic "helper" groups (Ca++, Mg++, etc.) are usually referred to as _________________ and may be associated with more than one type of enzyme.

13. The rate at which enzymes catalyze chemical reactions is influenced by factors such as pH, temperature, light, and the _________________________________ (of enzyme or substrate). Enzyme activity may also be influenced by chemicals known as _________________________________ (e.g., cyanide and arsenic), that can bind to the active site of an enzyme in place of the normal substrate molecules and block the enzyme’s action.

14. An enzyme inhibitor that binds to a site other than the active site, and thereby changes the active site so it can no longer function is called an __________________________________ inhibitor.

15. Enzymes that are released into the environment and are active outside the cell are called ____________________________________________. Bacteria use this type of enzyme to break down food materials, so they are more easily transported across the cell membrane.

16. A ____________________________________ is a non-protein organic group that can bind to an apoenzyme and convert it into a holoenzyme.

17. NAD, FAD, and NADP are examples of non-protein organic "helpers" known as _________________________________ and are much less specific than are enzymes. FAD is derived from the B-complex vitamin _________________________________.

18. _______________________________________________________________________________ is a coenzyme derived from the B-complex vitamin niacin.

19. A group of enzymes called ____________________________ (cell colors), are pigmented proteins with iron _____________________________. These enzymes help transfer electrons down the electron transport chains, but also move ______________________________ across membranes.
(5) 20. Matching - Enzymes: Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

<table>
<thead>
<tr>
<th>Term</th>
<th>Statement</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coenzyme</td>
<td>A. Catalyzes the transfer of phosphate groups between organic compounds.</td>
</tr>
<tr>
<td>Holoenzyme</td>
<td>B. Catalyzes the conversion of organic compounds into their chemical isomers.</td>
</tr>
<tr>
<td>Competitive</td>
<td>C. Pigmented enzyme which has an iron prosthetic group that can accept or donate electrons.</td>
</tr>
<tr>
<td>Allosteric</td>
<td>D. Active form of a conjugated enzyme, made up of protein plus a &quot;helper&quot; of some type.</td>
</tr>
<tr>
<td>Kinase</td>
<td>E. Protein portion of a conjugated enzyme. This portion is inactive alone.</td>
</tr>
<tr>
<td>Apoenzyme</td>
<td>F. Non-protein organic group which can bind with an apoenzyme to form a holoenzyme.</td>
</tr>
<tr>
<td>Cytochrome</td>
<td>G. Enzyme which is active as a protein alone, i.e., does not require a &quot;helper&quot;.</td>
</tr>
<tr>
<td>Simple enzyme</td>
<td>H. Inhibitor that binds to a site other than the active site and changes the configuration of the active site.</td>
</tr>
<tr>
<td>Isomerase</td>
<td>I. Inorganic group which can bind with an apoenzyme to form a holoenzyme.</td>
</tr>
<tr>
<td>Cofactor</td>
<td>J. Inhibitor which binds to the active site in place of the normal substrate.</td>
</tr>
</tbody>
</table>

(1) 21. Enzymes that are always present within the cell (are not inducible nor repressible) are referred to as ___________________________ enzymes.

(3) 22. Enzyme names often end in “ase” and provide information about the enzyme (what it acts on or what it does). Proteinases and lipases are enzymes that catalyze ___________________________. An enzyme called a carboxylase would be expected to ___________________________ to molecules, and a polymerase is an enzyme that ___________________________.
Fermentation and Cellular Respiration:

(5) 1. Define:

Glycolysis

Fermentation

Heterofermentative

Kreb's cycle

Cytochromes

(1) 2. ???????????????? may be defined as the catabolism of glucose resulting in the formation of two pyruvic acid molecules with the associated production of two molecules of ATP and reduction of two molecules of NAD to NADH + H⁺.

(1) 3. A biochemical pathway known as glycolysis allows glucose to be catabolized into molecules of ???????????????? with the associated formation of 2 ATP (net) and the reduction of two NAD to NADH + H⁺.

(2) 4. Toward the beginning of the glycolysis pathway, glucose is converted into glucose-6-phosphate, and later fructose-6-phosphate is converted into fructose-1, 6-bisphosphate. These reactions are catalyzed by ?????????????? enzymes and require energy (energy of activation) that is provided by ??????????????.

(2) 5. During glycolysis, ???????????????? is catabolized into pyruvic acids, two molecules of ATP are formed and two molecules of NAD gain electrons so are said to be ??????????????.

(1) 6. Kinase enzymes are those that catalyze reactions involving the transfer of ?????????????? groups between organic compounds.

(2) 7. Enzymes catalyzing the transfer of phosphate groups between organic compounds are called ?????????????? enzymes, while those catalyzing the reconfiguration of molecules into their chemical isomers are called ?????????????? enzymes.

(1) 8. The series of chemical reactions known as glycolysis cannot continue unless they are linked to one or more additional reactions, i.e., glycolysis occurs as a part of fermentation or cellular respiration. Why is this so? ????????????????
9. ____________________________________________________________________________ may be defined as the anaerobic decomposition of organic compounds that involves an organic compound (e.g., pyruvic acid) as the final electron acceptor. If organisms can form only one end product (lactic acid) via this pathway, they are said to be ____________________________________________________________________________.

10. Homofermentative organisms such as *Lactococcus lactis* produce ____________________________________________________________________________ as the only product of their fermentation activities.

11. The fermentation of glucose by bacteria such as *Lactococcus lactis* results in a net yield of ______ (#) ATP molecules per glucose. These ATP are produced via ____________________________________________________________________________ phosphorylation. The coenzymes reduced during the glycolysis portion of fermentation are reoxidized by passing their electrons to ____________________________________________________________________________ (the final electron acceptor).

12. Organisms that yield lactic acid as their only fermentation product are said to be ____________________________________________________________________________ and are often used in food processing. One organism which has this characteristic is ___________________________________________.

13. Organisms that can produce a variety of end products in association with fermentation are said to be ____________________________________________________________________________. One microorganism which has this characteristic and is often used in food processing is _________________________________________.

14. Chemoheterotrophs that have a _____________________________________________________________________________ type metabolism, can catabolize glucose (and other organic compounds) more completely than can fermentative organisms. They can also capture more of the energy available in these compounds and so can make more ____________________________________________________________________________ molecules per glucose catabolized, i.e., 36/38 rather than 2.

15. Bacteria such as *Pseudomonas* can completely catabolize glucose forming carbon dioxide and water by means of a metabolic process called _____________________________________________________________________________. This process is often divided into three steps or stages; what are they? ___________________________________________________________ ____________________________________________________________________________

16. During cellular respiration, pyruvic acid is decarboxylated (has a carboxyl group removed) and then binds with a coenzyme to form ____________________________________________________________________________ (a high energy compound).

17. Following glycolysis, and prior to the Kreb's cycle, a ____________________________________________________________________________ group is removed from pyruvate, and the remaining two-carbon unit is bound to ____________________________________________________________________________ forming acetyl-coA, a high energy compound. The enzyme complex responsible for catalyzing these reactions is called the ____________________________________________________________________________ complex.

18. The ____________________________________________________________________________ is a cyclic series of chemical reactions catalyzed by enzymes found in the matrix of mitochondria and in the cytoplasm of prokaryotic cells. These reactions allow organic acids to be catabolized and most of the energy released to be captured in the form of __________________________________ (NADH + H⁺ and FADH₂).
19. The carboxyl groups removed from various acids just prior to and during the Kreb's cycle are released from cells as ________________, a gaseous waste product.

20. The energy needed to bind the two-carbon remains of pyruvic acid to oxaloacetic acid at the beginning of the Kreb’s cycle is provided by the catabolism of ________________, a high energy compound.

21. One major function of the Kreb’s cycle (citric acid cycle or tricarboxylic acid cycle) is __________________________, a process that allows cells to release the energy stored in these molecules. What additional functions can be associated with this metabolic pathway? ___________________________________________________________________

22. The enzyme helper most often required for the reactions of the Kreb's cycle is a coenzyme called __________ and is derived from the B-complex vitamin _________________.

23. In eukaryotic cells the enzymes associated with glycolysis are found throughout the cytoplasm, those associated with the Kreb's cycle are found in the ________________________________ and those associated with the electron transport chain are bound to ________________________________ of these same organelles.

24. NADH+H+ and FADH2 can be reoxidized by passing their electrons to the electron transport chain. This series of reactions involves a number of pigmented enzymes called _________________ that have iron prosthetic groups, and are bound to membranes (most of them are integral proteins). Each NADH+H+ that is reoxidized via this pathway yields enough energy to make _____ (#) ATP and each FADH2 reoxidized yields enough energy to make _____ (#) ATP.

25. In respiratory organisms, the coenzymes reduced during the reactions of glycolysis and the Kreb's cycle are eventually reoxidized by passing their electrons to the ____________________________. This involves a series of membrane bound proteins (enzymes), most of which have iron prosthetic groups that can be alternately oxidized and reduced. The final electron acceptor at the end of this chain is an exogenous oxidizing agent such as ________________ or nitrate.

26. When cellular respiration occurs under aerobic conditions, the final electron acceptor is usually _________________. This inorganic compound picks up electrons and hydrogen protons to form _________________. Explain briefly why a final electron acceptor is needed, i.e., explain what function it serves. ___________________________________________________________________.

27. The ATP molecules formed in association with the electron transport chain of respiration are produced via __________________ phosphorylation, and the energy required is provided by the flow of ________________ across a membrane. Prokaryotic organisms produce approximately ____________ (#) molecules of ATP for each molecule of glucose they catabolize.
(2) 28. The proton motive force generated in association with electron transport chains is the gradient that causes hydrogen protons to flow through the ________________________ enzymes of membranes, thus providing the energy to make ATP. Research indicates that __________ (number) hydrogen protons must cross the membrane in order to make one molecule of ATP.

(3) 29. Most of the electron acceptors in the electron transport chain are ________________________ (pigmented proteins with iron prosthetic groups). The passage of electrons along this chain of proteins provides energy used to pump hydrogen protons across the membrane and to create a concentration and electrical gradient known as the ________________________. This gradient then causes protons to flow through an enzyme complex called __________________________ thus providing the energy needed to make ATP.

(2) 30. In eukaryotic cells, the passage of electrons along the electron transport chain causes ________________________ to accumulate within the intermembrane space of a mitochondrion. The reoxidation of one molecule of FADH$_2$ creates enough of an electrical and concentration gradient to drive the synthesis of (what and how many?) __________________.

(3) 31. Lipid catabolism involves many of the same biochemical pathways used in the catabolism of carbohydrates. When a triglyceride is catabolized, the three-carbon glycerol “backbone” can be phosphorylated to form dihydroxyacetone phosphate (DHAP) and then catabolized via the ________________________ pathway. The fatty acid chains can then undergo a process called __________________________ during which they are cut into two-carbon units and bound to a coenzyme to form __________________________. This can then be catabolized by the reactions of the Kreb’s cycle.

(2) 32. During protein catabolism, the amino acids are separated and ________________________ to yield substances that can be catabolized via glycolysis or the Kreb’s cycle, thus yielding energy and metabolic intermediates. During anabolism, metabolites such as pyruvate, oxaloacetate and α-ketoglutarate are used to make ________________________ that are then assembled into new proteins.
**Photosynthesis and Biosynthesis:**

(5) 1. Define:

   - Photophosphorylation
   - Ferredoxin
   - Bacteriochlorophyll
   - Anoxygenic
   - Calvin-Benson cycle

(3) 2. Phototrophic microorganisms use light energy to produce ATP by means of a process called _______________________________. The reactions involved, sometimes called the “light reactions” of photosynthesis, require light-sensitive molecules such as the chlorophylls of algae, cyanobacteria and green plants, the ________________ molecules of anoxygenic phototrophic bacteria, and the ___________________________ of certain types of Archaea.

(2) 3. Photophosphorylation as it occurs in the green and purple sulfur bacteria involves light trapping pigment molecules called ________________________________ and is cyclic when the electrons that leave the pigments eventually return to them. Because these bacteria do not produce oxygen as a by product of their photophosphorylation activities, they are said to be _______________ phototrophs.

(2) 4. ATP production via photophosphorylation (in algae and cyanobacteria) is very similar to ATP production via oxidative phosphorylation. In both processes, the flow of electrons along an electron transport chain provides energy used to “pump” hydrogen protons across a membrane to form a concentration and electrical gradient known as the __________________________. When these hydrogen ions flow back across the membrane “down hill” they pass through an enzyme complex called _________________________ and provide the energy required for the conversion of ADP + Pi into ATP.

(2) 5. Archae such as *Halobacterium* use a pigmented protein called ___________________________ to capture light energy. This membrane bound pigment “pumps” hydrogen protons across membranes when activated by light, and generates a proton motive force. Protons flow back across the membrane through ___________________________ enzymes, and provide the energy needed to make ATP.

(1) 6. Although most phototrophs use electron transport chains to generate a proton motive force, Archaea in the genus *Halobacterium* use ___________________________ instead.
(3) 7. Algae and cyanobacteria use non-cyclic photophosphorylation reactions to capture light energy. During these reactions, electrons "bounce" away from certain atoms in green pigment molecules called _______________________________ and are passed to electron acceptors. Electrons captured by ________________________ are passed along a series of cytochromes and then to other green pigments. Electrons that are captured by ___________________________ are ultimately passed to NADP and are not returned to the pigments.

(2) 8. The light trapping pigments and electron acceptors involved in photophosphorylation are found in association with membranes called ______________________________ in both cyanobacteria and eukaryotic cells. Similar pigments and electron acceptors are found in association with the ______________________________ of anoxygenic phototrophic bacteria.

(2) 9. Algae and cyanobacteria utilize an electron acceptor called ___________________________ to trap electrons leaving the pigment molecules of photosystem II. Since the electrons are not returned, the chlorophyll molecules "pull" replacement electrons away from water molecules and form __________________________ as a by-product.

(2) 10. The pigments associated with photosystem I (pigment system I) in algae and cyanobacteria pass their electrons to an acceptor molecule known as _______________________________. These electrons do not return, but are passed to a coenzyme called ____________________________.

(1) 11. The oxygen formed by oxygenic phototrophs such as algae and cyanobacteria are produced by splitting ________________________________ molecules.

(2) 12. The cytochromes involved in photophosphorylation are bound to membranes called ______________________________ in both cyanobacteria and eukaryotic cells. Enzymes which catalyze the reactions of the Calvin-Benson cycle are associated with inclusions called ______________________________ in prokaryotic cells and with the stroma of chloroplasts in eukaryotic cells.

(1) 13. A series of chemical reactions known as the ______________________________ are catalyzed by enzymes found within the stroma of chloroplasts or within carboxysomes.

(2) 14. The chemical reactions associated with non-cyclic photophosphorylation trap light energy in two ways, i.e., result in the formation of two types of energy-rich compounds, these are ___________________________ and ___________________________.

(3) 15. Autotrophic organisms use an anabolic pathway known as the ______________________________ to "fix" inorganic carbon (in the form of carbon dioxide) into organic compounds (sugars). This pathway requires energy which is provided by ___________________________.

(2) 16. Autotrophic microorganisms use an enzyme called ______________________________ to bind carbon dioxide to a five carbon sugar molecule called ribulose bisphosphate at one point in the Calvin-Benson cycle. How many reactions must be catalyzed in order to form one molecule of fructose? ______________________________
(2) 17. The Calvin-Benson cycle is a series of chemical reactions which allow autotrophic organisms to ____________________________. The enzymes needed to run this pathway are associated with structures (inclusions) called __________________ in prokaryotic cells.

(2) 18. A series of chemical reactions which allow autotrophic organisms to "fix" carbon from carbon dioxide into organic compounds (sugars) is known as the __________________________. The enzymes which catalyze the reactions of this cycle are found in association with the __________________________ in eukaryotic cells.

(2) 19. The chemical reactions of photosynthesis are sometimes represented by the following formula: 6CO₂ + 6H₂O + energy from light will yield C₆H₁₂O₆ + 6O₂. Although this is generally true for algae and cyanobacteria, it does not apply to all organisms. Bacteria that are nutritionally categorized as photoheterotrophs are able to make ATP using light energy, but cannot __________________________. Bacteria that are categorized as chemoautotrophs can use inorganic carbon to make sugar, but cannot __________________________. Some bacteria are able to run both the light reactions and dark reactions of photosynthesis, but are anoxygenic.

(1) 20. Phototrophic bacteria in the genus *Halobacterium* use a light sensitive pigment called __________________________ to capture light energy and to pump hydrogen protons across membranes. ATP production in these organisms does not require cytochromes in an electron transport chain.

(5) 21. Matching - Photophosphorylation: Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

___ Bacteriophaeophytin  A. Membranes of algae and cyanobacteria containing pigments, electron acceptors and cytochromes.

___ Thylakoids  B. Can donate electrons and hydrogen protons to bacteriochlorophylls, leaving sulfur behind.

___ Chlorophylls  C. Pigmented enzymes that can receive electrons passed from plastoquinone, and give them to chlorophylls.

___ Bacteriochlorophylls  D. Electron acceptor associated with anoxygenic phototrophic bacteria; can pass electrons to cytochromes.

___ Ferredoxin  E. Pigments of anoxygenic phototrophic bacteria; can pass electrons to ubiquinone and then to NADP.

___ NADPH + H⁺  F. Green pigments of algae and cyanobacteria capable of responding to light by giving up electrons.

___ Cytochromes  G. Can be split by oxygenic phototrophic organisms, and will donate electrons and H⁺ to pigments.

___ Hydrogen sulfide  H. High-energy compound formed during photophosphorylation, but not a nucleoside triphosphate.

___ Bacteriorhodopsin  I. Iron-sulfur protein capable of accepting electrons from photosystem I and passing them to a coenzyme.

___ Water  J. Light sensitive protein capable of transporting hydrogen protons across cellular membranes.
DNA, RNA and Protein Synthesis:

(5) 1. Define:

Nucleotide

Okazaki fragments

Sigma factor

Aminoacyl-t-RNA-synthetase

Peptidyl-transferase

(2) 2. The nucleic acids, DNA and RNA, are long chain molecules made up of smaller units called ________________ that are connected together by phosphodiester bonds. In RNA these smaller units contain a sugar called ________________ while in DNA they do not.

(2) 3. ________________ are long chain molecules made up of small repeating units called nucleotides. The 5' end of each nucleotide is bound to the 3' end of the next via a covalent bond known as ________________ bond.

(2) 4. Cellular DNA molecules differ from cellular RNA molecules in that they are double stranded rather than single, are much larger (longer), contain the pentose sugar ________________ and the pyrimidine base ________________.

(2) 5. Cellular DNA molecules (and some viral RNA molecules) are double stranded, i.e., the purine bases in one strand are complimentary to specific ________________ bases in the other and are bound to them by ________________ bonds.

(2) 6. In the DNA double helix (duplex), the nitrogenous bases Adenine and Guanine (bases with two rings in their structure) are called ________________. These will form hydrogen bonds with their ________________ bases Thymine and Cytosine, respectively.

(1) 7. The two strands of a DNA double helix are oriented in opposite directions 5’ to 3’, or are “up-side-down” relative to one another, and so are said to be ________________.

(2) 8. A ________________ is made up of a pentose sugar, a nitrogenous base, and a phosphate group. If a polymer is formed by connecting a series of these small molecules together, what chemical group would be located at the 5’ end? ________________
9. The process by which DNA molecules reproduce themselves is sometimes called semi-conservative ________________ because each new duplex formed contains half of the original DNA. This process is initiated at sites on an existing DNA strand called _________________. Circular chromosomes usually have only one of these, while linear chromosomes have many.

10. DNA replication as it occurs in *E. coli*, requires a number of enzymes. These include DNA and RNA ______________________________, enzymes that can catalyze the attachment of nucleotides to the free 3' ends of existing nucleotide strands; and _________________, an enzyme that serves to bind the fragments of each lagging strand into a single long chain.

11. Prokaryotic microorganisms reproduce their chromosomal and plasmid DNA by a process called _________________. This process begins at a site called the origin, and usually proceeds around the loop in both directions. Since polymerase enzymes can only “build” DNA in the 5' to 3’ direction, one strand is formed in a continuous sequence, and the other is formed in a series of segments called _________________.

12. The polymerase enzymes involved in building DNA can only “build” in one direction because they can only add nucleotides (bases) to the _______ ends of growing nucleotide strands. As a consequence of this, the leading strand is built as a continuous sequence, while the lagging strand must be built in segments as the duplex unwinds. In *E. coli*, the enzyme required to start each Okazaki fragment is DNA dependent _________________. The Okazaki fragments are eventually spliced together by _________________ enzymes, and the DNA duplex is completed.

13. The process by which cellular RNA molecules are formed is called RNA synthesis or _________________ and is similar to replication in that it requires a single strand of DNA as a template (pattern) and energy as provided by ________________.

14. Transcription is the process by which ____________________________ are made, and is similar to semi-conservative replication in that:

1.) it requires a single strand of DNA to serve as a template.
2.) ________________
3.) ________________

15. Transcription requires DNA as a template or pattern and can only be initiated at specific locations. The region on a DNA strand where transcription begins is called the ________________ site and is recognized by a protein called ________________ (which is a portion of the RNA polymerase enzyme complex). Transcription also requires ________________ which is provided by the nucleotides (rNTPs) involved in the process.

16. The portion of DNA dependent RNA polymerase that determines where transcription will begin and in which direction it will proceed is called _________________. Once this protein binds to the promoter site of the DNA molecule, the core enzyme can bind, and transcription can proceed. Each _________________ molecule formed via this process is essentially a copy of one small segment of one strand of the DNA.
17. In eukaryotic cells most genes are split genes, so transcription is followed by a process called __________________________________ during which RNA molecules are modified by having their __________________________ removed and by having a cap and a poly-A tail added. This process is accomplished in part by s-RNA.

18. Eukaryotic cells produce small or short RNA molecules (s-RNA) that bind to proteins to form structures called ___________________________. These are involved in post-transcriptional modification, and apparently recognize where RNA molecules are to be cut and spliced. After the intervening regions are removed, segments of RNA called __________________________ are spliced together and the resulting molecule is given a cap and a poly-A tail.

19. RNA molecules that coordinate the attachment of t-RNA to m-RNA during protein synthesis are called __________________________ while RNA molecules called __________________________ carry individual amino acids to the ribosomes for protein synthesis.

20. RNA molecules known as __________________________ are actually copies of small segments of DNA known as structural genes. In prokaryotic cells, these RNA molecules often contain information allowing them to encode more than one polypeptide chain (protein) because transcription is _______________________________.

21. Individual t-RNA molecules carry specific amino acids to the ribosome during the synthesis of proteins. The enzymes catalyzing reactions attaching specific amino acids to t-RNA are called __________________________. The portion of the t-RNA that determines which amino acid is added to the polypeptide next is called the ______________ region and forms hydrogen bonds with m-RNA at the ribosome.

22. The sequence of codons on a ______________________ molecule determines the sequence of amino acids in a polypeptide, but the amino acids cannot recognize nor bind to these molecules. Instead, each amino acid is carried by a specific t-RNA molecule. The factor that insures each t-RNA is carrying the correct amino acid is __________________________.

23. The primary factor determining which t-RNA will bind to the A-site of the ribosome at any given moment is whether or not the ______________________ region of that t-RNA can form hydrogen bonds with the complimentary bases of m-RNA. The enzymes that insure each t-RNA is carrying the correct amino acid are called __________________________.

24. A ______________ may be defined as a set of three bases on m-RNA that encodes a specific amino acid. This same term can also be applied to a set of three bases on a DNA strand.

25. The process by which proteins are made is called protein synthesis or __________________________ and occurs in association with structures called __________________________ in both prokaryotic and eukaryotic cells.

26. The primary factor determining which t-RNA will bind to the A-site of the ribosome at any given moment is whether or not the ______________________ region of that t-RNA can form hydrogen bonds with the ______________________ (set of three complimentary bases) of m-RNA.
27. During the translation process, the ______________________ regions of a t-RNA molecules form hydrogen bonds with the codons of ______________________ as they pass through the ribosome. The molecule that catalyzes the formation of a peptide bond between two adjacent amino acids is called ______________________ and is a ribozyme and part of the ribosome itself.

28. Three sets of nucleotides (nitrogenous bases) known as "ocher", "amber" and "umber" do not encode individual amino acids, but instead provide cells with what information? ________________________________________.

29. Use the DNA sequence shown and the genetic code handout provided to complete the statements below. If the sense strand of DNA has the base sequence TCTACGTTTGGGCATACCTTACCAAC

Transcription of this DNA will yield ________________________________________________________________

Translation of the RNA represented above will yield ________________________________________________________________

Does the polypeptide represented above contain the same number of amino acids as there are codons in the m-RNA formed? ________ Explain why or why not.

30. Explain briefly how the nucleotide sequence of a structural gene can have a significant influence on metabolism (i.e., how genetic information influences cell activity).

31. Each amino acid being added to a growing polypeptide chain is bonded to the previous amino acid by a covalent bond called a ____________________ bond. This bonding is catalyzed by a ribozyme called peptidyl transferase that is part of the ______________________. The termination of a growing polypeptide chain is signaled by the presence of a __________________________________ on the m-RNA molecule.

32. The term ______________________ may be used to describe a unit formed by many ribosomes attached to and translating a single m-RNA molecule.

33. A single m-RNA molecule is normally attached to several ribosomes during the translation process, forming a unit referred to as a ______________________. Each ribosome provides a ribozyme called ______________________ that catalyzes the formation of peptide bonds between the individual amino acids thus forming a polypeptide chain.

34. The ribozyme responsible for catalyzing the formation of each peptide bond at a ribosome is actually a ______________________ molecule (be specific). Name a substance composed of short amino acid chains that are not formed in association with ribosomes (hint, one type was mentioned during the lecture on cyanobacteria). __________________________________
Regulation of Gene expression (Genetic Control):

1. Define:
   - Operon
   - Promoter
   - Operator
   - Catabolite repression
   - Cyclic-AMP

2. Early investigations into the regulation of metabolic processes indicated that many enzymatic pathways are controlled by _______________________, a process which allows the product of a metabolic pathway to influence the activity of the enzymes in that pathway.

3. Both prokaryotic and eukaryotic cells can regulate some of their metabolic activity at the enzyme level by means of a mechanism called feedback inhibition. This regulation involves the ______________________ (be specific) of an enzyme, usually the first in a series, by the end product of the metabolic pathway. In what way might this process be considered less efficient than end-product repression? _____________________________

4. ______________________ is a process or mechanism that allows the end-product of a metabolic pathway to inhibit its own production by acting as an allosteric inhibitor for the first enzyme in the pathway.

5. In eukaryotic organisms, one m-RNA molecule is essentially a copy of the genetic information contained within an area of DNA known as a ______________________. In prokaryotic cells, transcription is often polycistronic. How does this influence the resulting m-RNA molecule? _____________________________________________

6. In prokaryotic cells, the genes encoding enzymes involved in a single metabolic pathway are often located adjacent to one another within a single ______________________ (region of DNA containing structural genes and the control elements regulating their transcription). The transcription of these genes is said to be ______________________ because they are not transcribed individually, but are transcribed together as a single unit. Translation of the resulting m-RNA often begins before transcription is completed.

7. The promoter site (P-site) of an operon has what function? _____________________________
8. An _____________________________ is a segment of DNA that includes a series of structural genes plus the control elements involved in regulating their transcription. RNA polymerase binds and begins transcription at a site within this segment known as the ________________ site.

9. In the repressible system controlling tryptophan biosynthesis, the R-gene encodes a protein known as the ___________________________. This protein is not active alone, so can bind to the ________________ site only when it has been activated by the corepressor tryptophan.

10. In the repressible system controlling tryptophan biosynthesis in E. coli, the R-gene (regulatory gene) encodes a protein called the ___________________________. This protein can bind to the operator site and block transcription only if it is aided by ________________.

11. Tryptophan biosynthesis in E. coli is regulated at the genetic level through a ___________________________. The genes encoding enzymes involved in tryptophan biosynthesis are transcribed together as one large m-RNA (transcription is polycistronic), or are not transcribed at all. If excess tryptophan is present within the cell, it acts as a ________________ to activate the repressor protein. The repressor-tryptophan complex then binds to the ___________________________ site (be specific) on the DNA and blocks the transcription of all the genes coding for enzymes used in tryptophan biosynthesis.

12. In E. coli the utilization of lactose is controlled at the genetic level through an ________________ _________________. A type of enzyme called β-galactoside ________________ allows lactose to enter the cell and a type of enzyme called thiogalactoside transacetylase converts some of the lactose to _________________. This molecule then acts as an inducer for the lactose utilization system.

13. The transcription of genes needed in the utilization of lactose can be partially induced by binding ________________ to the repressor protein, thus inactivating it. However, even after the repressor is removed, transcription will be minimal until cyclic-AMP and a protein called ________________ bind near the promoter site and enhance the attachment of sigma factor.

14. The operon controlling lactose utilization in E. coli contains three structural genes that encode enzymes involved in lactose utilization. The enzymes include a permease (to bring lactose into the cell), one called ___________________________ (that breaks lactose into glucose and galactose) and a transacetylase involved in forming allolactose. What is the function of the allolactose formed? ___________________________

15. In the inducible system controlling lactose utilization, the repressor protein is active under what circumstances? ____________________________

16. The β-galactoside permease encoded by a gene within the lactose utilization operon (lac operon) has what function? ____________________________

17. Thiogalactoside transacetylase is an enzyme encoded by a gene in the ________________ _________________. The function of this enzyme is to convert lactose into ________________.
(2) 18. ____________ is a mechanism that allows organisms to utilize enzymatic pathways involving constitutive enzymes in favor of using those for which the enzymes must be induced. A nucleotide known as ____________ serves as a regulatory molecule in this process.

(3) 19. Catabolite repression is a mechanism that allows organisms such as E. coli to utilize ____________ in favor of a catabolite such as lactose. The enzymes involved in glucose catabolism are ____________ (always present) so do not require induction. In order to use lactose, an E. coli cell must induce the lactose utilization operon with allolactose and must contain high levels of ____________ (a regulatory nucleotide).

(2) 20. Enzymes always present within prokaryotic cells, i.e., are neither inducible nor repressible are said to be ____________. A mechanism that allows cells to utilize these enzymes in favor of those which must be induced is called ____________________________.

(2) 21. High glucose levels within E. coli cells will inhibit the activity of two enzymes required to induce lactose utilization, specifically β-galactoside permease, and ____________, the enzyme responsible for the formation of cyclic-AMP. For this reason, glucose (an important carbon source) plays a major role in ____________, the regulatory mechanism allowing bacteria to use constitutive enzymes in favor of inducible ones.

**Genes and Mutations:**

(5) 1. Define:

Genetics

Phenotype

Point mutation

Translocation

Mutagenic agent (mutagen)

(2) 2. ____________ may be defined as the science or study of heredity and is concerned with the physical and chemical properties of hereditary material (_________________________), and how this material is transmitted from one generation to the next.
3. Individual genes generally occupy specific locations within a chromosome, plasmid or viral genome. This is known as the _______________. In some cases one or more genes may overlap within the same nucleotide sequence.

4. A sequence of nucleotides (DNA or RNA) that encodes a functional polypeptide or for a functional RNA molecule can be referred to as a ________________. If this sequence includes one or more intervening regions (introns) it may be called a ________________. Such sequences are common in eukaryotic cells, but uncommon in ordinary bacteria.

5. The _______________ of a cell is the total DNA content of the chromosomes of that cell and includes control elements as well as structural genes.

6. The genetic constitution of a cell or organism (content of genetic information) may be referred to as the ________________ of that organism, or its genetic potential. Whether or not the genetic information is expressed is dependent upon the ________________ (both outside and inside the cell).

7. The _______________ of a cell is the total DNA content of the chromosomes of that organism. The observed behavior or appearance (physiological characteristics) of an organism is referred to as the ________________ of that organism, and is greatly influenced by the environment in which the organism is placed.

8. The ________________ of an organism, or the observed characteristics of that organism is dependent upon the genotype present and the ________________. Under what circumstances would genetic information not be expressed? ________________

9. A ________________ may be defined as any change in the nucleotide sequence of DNA (or RNA in the case of some viruses), within a cell.

10. Any change in the base sequence of DNA (excluding some changes brought about by genetic exchange) is referred to as a ________________ and is the basis for ________________ or changes in populations over time.

11. Spontaneous mutations, those which occur under natural conditions or for no discernible reason, occur at a rate of about ________________. If a bacterial population is able to reach its m-concentration in 24 hours, how many mutations would be expected to have occurred within the population during that time period? ________________

12. Point mutations known as ________________ are potentially less significant to cells than are addition or deletion type mutations. Why is this so? ________________

13. Mutations known as ________________ involve the movement of one or more genes from one place to another within a chromosome, between chromosomes or between chromosomes and plasmids. These are sometimes initiated by segments of DNA called ________________ or "jumping genes".
14. Frame-shift mutations may be caused by either ___________________________ DNA molecules. Either will cause a “shift” in the “reading frame” for all of the ___________________________ (sets of three bases) on the m-RNA molecule that determines the amino acid sequence of a given protein. A physical mutagen known as ___________________________ increases the frequency of such mutations by causing the formation of T-T dimers. Since these mutations are usually lethal to cells, there is a great deal of concern about the amount of this mutagen in the environment.

15. A ___________________________ is a chemical or physical factor that increases the rate at which mutations occur.

16. Chemical agents that are structurally similar to naturally occurring nitrogenous bases (A, T, C or G) and so can be incorporated into DNA are known as ___________________________. These chemicals often cause substitution-type point mutations.

17. Mutagens such as 5 bromo-uracil and 5 fluoro-uracil are called ___________________________ because they can be incorporated into DNA in the place of naturally occurring bases. Since these mutagens encode the wrong complimentary bases when the DNA replicates, they tend to cause ___________________________ type point mutations.

18. Given a DNA strand with the base sequence TACCTGGCAGTGGGACCTAC, determine what type of mutation (nonsense, missense, or silent), would occur if the second purine present were replaced by a pyrimidine. A ___________________________ mutation would occur.

19. Although point mutations known as ___________________________ may or may not result in a change in amino acid sequence, mutations known as "frame shifts" most certainly will. One example of a physical mutagen known to cause "frame shift" mutations is ___________________________.

20. ___________________________ is a physical factor known to cause the formation of thymine-thymine dimers within DNA. Such dimers will encode only one complimentary base instead of two, and so will result in a ___________________________ type point mutation.

21. ___________________________ or "jumping genes" are segments of DNA that can initiate their own movement from one place to another within chromosomes, between chromosomes, or between chromosomes and plasmids. Such segments may be responsible for causing non-point mutations known as ___________________________.

22. Segments of DNA capable of initiating their own movement from one place to another within a chromosome or between chromosomes are referred to as ___________________________. The enzymes responsible for “cutting” DNA and allowing these segments to move are called ___________________________ enzymes and may or may not be encoded by genes within the segment of DNA “jumping”.

23. Bacteria exposed to ultra violet are often killed by the formation of ___________________________ (These will encode an incorrect number of complimentary bases, and cause deletion type point mutations). Bacteria that contain enzymes activated by visible light can repair such damage by doing what? ___________________________
Mechanisms of Genetic Exchange and Recombinant DNA Techniques:

(5) 1. Define:

Transformation

Episome

Sexduction

Transduction

Restriction endonuclease

(2) 2. _______________________ may be defined as a combination of chromosomal DNA from more than one source. Such molecules can be constructed in vitro using a variety of genetic engineering techniques, or may occur naturally as the result of ______________________ (horizontal gene transfer).

(1) 3. DNA molecules containing chromosomal material from more than one source are referred to as _______________________ and may be formed in bacteria via a variety of genetic exchange processes.

(1) 4. In nature, prokaryotic cells are able to exchange DNA most efficiently if they are closely related (usually within the same species). This is because bacteria produce enzymes called _______________________ that can recognize and chop up foreign DNA.

(1) 5. In most instances, DNA that is transferred into a recipient cell will replace the _______________________ DNA already present (i.e., DNA that is not identical, but is encoding the same general characteristics).

(2) 6. The genetic exchange mechanisms used by bacteria are similar to one another in a number of ways. Explain any two ways in which all of these processes are similar.

a) __________________________________________________________

b) __________________________________________________________

(2) 7. All genetic exchange processes are similar in that the cells involved do not ________________ ________________________, the DNA transfer is one-way (from donor to recipient) and in most instances only a small portion of the genome is transferred. Under natural conditions, genetic exchange is most successful if the cells involved are closely related because? __________________________________________________________
8. ___________________________ is a genetic exchange process that involves the transfer of DNA from a dead donor cell to a live recipient. This process was first observed by ___________________________ and was occurring in bacteria belonging to the genus ___________________________.

9. During transformation, bacteria are able to pick up ___________________________. Bacteria are able to undergo transformation only if they are ___________________________, i.e., able to transport the DNA through their cell walls. This usually requires that they be undergoing the fission process.

10. A genetic exchange mechanism that involves the transfer of DNA from donor to recipient with the aid of structures called sex pili is known as ___________________________. If the cells involved in the "mating" process are F- (F minus) and F' (F prime), and if the recipient cell becomes recombinant and a partial diploid, the process is called ___________________________.

11. During conjugation, DNA is transferred from donor to recipient with the aid of structures known as ___________________________. These structures are coded for by genes carried on a plasmid called ___________________________.

12. A segment of extrachromosomal DNA that can become incorporated into the chromosome is called a/an ___________________________ and may be either a virus or a plasmid.

13. A "male-type" E. coli cell that carries its "male" genes within its chromosome is referred to as a ___________________________ because when "mated" to an F- (F minus) cell it is very likely to cause the formation of a recombinant cell.

14. A specific form of conjugation called ___________________________ involves a transfer of DNA from an F' (F prime) cell to an F- (F minus) cell and results in the formation of a “male” type cell that is ___________________________ and a ___________________________.

15. The transfer of DNA from donor to recipient via a virus which was initially cytolytic is referred to as ___________________________(be specific).

16. The process by which DNA can be transferred from one bacterial cell to another via a virus which is initially a prophage is referred to as ___________________________(be specific).

17. A piece of DNA that can initiate its own replication when placed into a host cell may be called a ___________________________ and may be either a plasmid or a virus.

18. A type of cloning vector known as a ___________________________ can be formed by combining plasmid DNA with bacteriophage lambda DNA. This type of vector can be used to transfer relatively large segments of DNA.

19. Bacteria such as E. coli can (and do) synthesize human proteins, but before human genes are placed into bacteria, they must be modified. Explain why this is necessary, and how the genes are typically modified. ___________________________.
Matching - Genetic exchange: Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

<table>
<thead>
<tr>
<th>Term</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Specialized transduction</td>
<td>A. The location occupied by a specific gene on a chromosome, a plasmid or virus genome.</td>
</tr>
<tr>
<td>Homologous DNA</td>
<td>B. An <em>E. coli</em> cell that is lacking sex pili and so is considered to be female.</td>
</tr>
<tr>
<td>Sexduction</td>
<td>C. Bacteriophage lambda DNA that is lacking lytic genes, and can be used as a cloning vector.</td>
</tr>
<tr>
<td>F- (F-minus)</td>
<td>D. A plasmid that carries genes encoding the formation of sex pili and the initiation of genetic exchange.</td>
</tr>
<tr>
<td>Cosmid</td>
<td>E. Segment of DNA (may be a virus or plasmid) that is a replicon, i.e., able to initiate its own replication.</td>
</tr>
<tr>
<td>Hfr</td>
<td>F. A genetic exchange process involving a virus that was initially cytolytic.</td>
</tr>
<tr>
<td>F-factor</td>
<td>G. A specific type of conjugation that yields a male, partially diploid, recombinant recipient cell.</td>
</tr>
<tr>
<td>Cloning vector</td>
<td>H. Genes that encode the same traits or characteristics in general, but are not identical (also called alleles).</td>
</tr>
<tr>
<td>Locus</td>
<td>I. A genetic exchange process that involves a virus that was initially a prophage.</td>
</tr>
<tr>
<td>Generalized transduction</td>
<td>J. When &quot;mated&quot; to F- (F minus) is very likely to yield a recombinant F- (F minus) cell.</td>
</tr>
</tbody>
</table>

Introduction to Prions, Viroids and Viruses:

1. Define:

   - Prion
   - Prophage
   - Lysogenic conversion
   - Retrovirus
   - Reverse transcriptase

2. The term virus is Latin for ________________________ and has been applied to a variety of different disease causing agents over time. Currently, the term virus refers specifically to a non-cellular entity with a nucleic acid core and protein capsid.
3. A complete virus particle as it exists outside of its host (the infective form of a virus) is referred to as a ______________ and typically includes a nucleic acid core that is surrounded by a _____________________________.

4. A typical virion includes a _______________ core that is surrounded by a protein coat or capsid made up of units called ___________________________. These are made up of still smaller units called protomers.

5. A typical virus particle is composed of a nucleic acid core containing ___________________________ that is surrounded by a protein covering called a ___________________________. In some cases, the nucleocapsid is covered by an envelope and in other cases it is equipped with an elaborate ____________________________ that includes a collar, core, sheath, base plate, fibers and pins.

6. The nucleic acid core of a virus may be composed of DNA or RNA but not usually both. In what other ways do the genomes of various viruses differ from one another. ______________

7. A _______________ is a virus that infects bacteria and causes lysis (cell death) at the completion of its life cycle. The process by which such a virus attaches to its host is called ___________________________ and involves chemical bonding between proteins (tail fibers, capsid, envelope) of the virus and specific receptor sites on the host cell surface.

8. A coliphage is said to be avirulent or _______________ if it does not cause lysis of its host. When such a virus has become incorporated into the host cells chromosome, it is called a ___________________________ and will be reproduced along with the host cells DNA.

9. The first two stages in the life cycle of a typical T-even virus are _______________ (during which the tail-fibers of the virus bind with specific receptor sites on the cell surface) and _______________ (during which the nucleic acid core is injected into the cell).

10. During the life cycle of a T-even bacteriophage, different viral genes are transcribed and translated at different times. The immediate early phage genes encode enzymes that _______________ while the late phage genes encode _______________.

11. The genes of a T-even bacteriophage are not all transcribed and translated at the same time. List the sequence in which these genes are activated, and explain what each set codes for within the host.
   a) _______________
   b) _______________
   c) _______________

12. After the genome of a cytolytic virus has entered its host, it uses the host cells metabolic processes to transcribe and translate its genes. What host cell materials are essential to viral reproduction? _______________
13. A temperate phage that has become incorporated into the chromosome of its host cell is referred to as a ___________________________. If the host takes on new phenotypic characteristics (such as the ability to produce toxins) due to the presence of the viral genes it is said to have undergone ____________________________________________.

14. A bacterial cell (lysogenic cell) is said to have undergone ______________________________ when it has been infected by a virus, and has acquired some characteristics that are encoded by viral genes. The ability of certain bacteria to produce ______________________________ is believed to be due to this process.

15. Animal viruses are sometimes surrounded by a flexible membrane-like layer called a/an ______________________________ that is acquired as they exit their host cell.

16. A coliphage called ___________________________________ is a temperate or avirulent phage that can enter the chromosome of it’s host to become a __________________________ as long as it’s lytic genes are not being expressed. If the host cell becomes stressed, a proteolytic enzyme encoded by the RecA gene will degrade the specific __________________ protein blocking the transcription of the viral lytic genes. The virus will then complete a lytic cycle and kill it’s host.

17. HIV is a single-stranded RNA type virus called a ________________________ because its genetic information is reverse transcribed into DNA after it enters its host. This virus carries with it an enzyme called RNA-dependent DNA polymerase or _______________________________. What happens to the viral DNA within the host cell? __________________________________________________________

18. The adsorption process of HIV involves binding of spike proteins on the ______________________________ with specific receptors on the host cell surface.

19. HIV is a retrovirus which must ______________________________ it’s genetic information from RNA into DNA before it can become incorporated into the hosts chromosome.

20. HIV uses an enzyme called __________________________ to incorporate the DNA version of its viral genome into the chromosome of its host.

21. An enzyme that allows viral RNA to be reverse transcribed into DNA and then replicated to form double-stranded DNA is called _______________________________ and is found in association a group of viruses called ______________________. Among these, the virus called ______________________ is of great concern to humans.

22. Viruses are known to cause a variety of diseases in humans including the common cold, chicken-pox, measles, hepatitis and AIDS. Certain viruses have also been shown to induce the formation of ______________________________ in humans.

23. Genes capable of causing the formation of tumors within humans and other animals are called ______________________________ and are sometimes viral in origin.
(2) 24. Vaccines produced to prevent infection with influenza viruses must be modified frequently because the viral receptors keep changing. When viral receptor proteins change due to mutations in the genes encoding them, the viruses experience ____________________________, but if the changes in receptor proteins are due to recombination of viral genes from multiple different sources, the viruses have experienced ____________________________.

(5) 25. Matching - Viruses:

Match the disease on the left with the virus most likely to be the causative agent, and place the letters of correct matches in the blanks provided.

_____ Hepatitis  A. Symptoms associated with infection by human herpesvirus types 1 and 2.
_____ Influenza  B. Nearly all cases correspond to infection with the human papilloma virus (HPV).
_____ Mumps  C. Zoonosis of the nervous system, caused by a mosquito-borne arbovirus.
_____ German measles  D. Highly contagious disease caused by the varicella-zoster virus.
_____ Oral & Genital lesions  E. Enlargement of the parotid salivary glands caused by a paramyxovirus.
_____ Encephalitis  F. Caused by a rhabdovirus transmitted via the bite (or lick) of an infected animal.
_____ Rabies  G. Acute illness caused by a paramyxovirus known as the rubella virus.
_____ Cervical cancer  H. Lower respiratory tract infection caused by Orthomyxoviruses carried by pigs, birds and humans.
_____ Common cold  I. Viral induced inflammation of the liver and reduced liver function.
_____ Chicken-pox/shingles  J. Upper respiratory tract infection caused by a rhinovirus.
Control of Microorganisms and Antimicrobial Chemotherapy:

(5) 1. Define:

Bactericidal

Disinfectant

Antibiotic

Penicillinase

Aminoglycoside

(2) 2. Microbial control methods that kill cells in large numbers are referred to as being ___________. When all viable cells have been eliminated from an object or material (glassware, media, etc.) that object or material is said to have been ________________________________.

(1) 3. Materials such as microbiological media, glassware, syringes, needles, etc. are said to have been ________________________________ if they have been subjected to a treatment which has left them virtually free of any viable cells.

(2) 4. A microbial control method that does not kill cells, but inhibits their growth is said to be _______________. One example of a physical factor that has this effect is ________________.

(1) 5. Why would pasteurization or boiling for one minute not be considered an effective means of sterilizing liquids? ______________________________________________________________________

(3) 6. Temperature is a physical factor often used to control microorganisms. High temperatures (heat) may be applied in a method called ________________________________ that involves the alternate boiling and cooling of liquids over a period of three days. Heat (steam) under pressure may be applied in a device called an ________________________________. Cold temperature (freezing) can also be used to control microorganisms, but is ________________ rather than cidal because most bacteria are psychroduric.

(2) 7. ________________________________ is a physical factor that can be used to sterilize heat sensitive materials such as plastic Petri dishes and pipettes. This control method is highly effective, but is used less commonly than heat because it is expensive and ________________________________.

(1) 8. Heat sensitive liquids and gasses can be sterilized by means of ________________________________, a method that is not bactericidal, but can be used to remove all viable cells.
9. Chemical agents that are used to control pathogenic microorganisms on non-living surfaces are referred to as ___________________________. Chemicals that are used systemically to control pathogens within the body are called ____________________________ agents and may be either antibiotics or synthetic drugs.

10. Many chemicals have been designed for use in the control of pathogenic microorganisms outside the body. If these chemicals are to be used on a regular basis, they must meet certain criteria. They must be _____________________________, non-hazardous to the person applying them, readily soluble, non-corrosive or not damaging to the surface they are being applied to, and ___________________________ within a reasonable period of time.

11. What two features of a chemical agent would you be most concerned about if you were expected to use the chemical to control microbes on a regular basis (every day).
   1) __________________________________________________________________________
   2) __________________________________________________________________________

12. A/an ____________________________ is a chemical agent used to control pathogenic microorganisms on non-living surfaces.

13. A chemical agent designed to control pathogenic microorganisms on living surfaces is referred to as a/an _____________________________, while one designed for use on non-living surfaces is called a/an _____________________________. In some cases, the same chemical may be used for both applications.

14. Matching: Match the terms on the left with the most appropriate statements on the right and place the letters of correct matches in the blanks provided.

   ____ Freezing  A. Treatment which renders materials or objects free of any viable cells.
   ____ Antiseptic  B. Cause the hydrogens of organic compounds to be replaced by methyl or ethyl groups.
   ____ Metal ions  C. Cidal physical factors that cause the formation of ions or free radicals; expensive & hazardous to apply.
   ____ Alkylating agents  D. Powerful oxidizing agents including chlorine, bromine, fluorine, and iodine.
   ____ Pasteurization  E. Chemical agent used to control pathogens on non-living surfaces.
   ____ X-rays  F. Can be used to sterilize media even though cells are not killed.
   ____ Filtration  G. Treatment that is bacteristatic, but not cidal to cells that are psychroduric (most bacteria are).
   ____ Disinfectant  H. Chemical agent used to control pathogens on living surfaces (skin).
   ____ Halogens  I. Heat treatment that will kill vegetative cells made by bacteria, but not endospores.
   ____ Sterilization  J. Mercury, lead, zinc, copper and silver.
15. Chemicals called _____________________________ are often mixed with other control agents because they decrease the surface tension of water and increase the penetrating ability of other disinfectants. A group of chemicals called ___________________________ are powerful oxidizing agents that exert their effect by causing the oxidation of cellular proteins. These are often used to disinfect water and non-living surfaces, but are sometimes used as antiseptics.

16. Chemicals designed for use in the control of pathogenic microorganisms inside the body are called _____________________________ drugs or agents. If these are (at least initially) produced by some type of microorganism, they are called _____________________________.

17. A variety of microbes have been found to be sources of new antibiotics. Bacteria in the genera ___________________________ and ___________________________ are two examples of organisms with this ability.

18. The concentration of an antimicrobial agent needed to gain clinical control of a pathogen is referred to as the _____________________________ dose for that drug, and must be carefully monitored. What dangers exist if the concentration is too low, or too high? (explain both) ____________________________________________

19. Antibiotics that are effective against only a few types of microorganisms (sometimes only pathogens within a single genus) are referred to as _______________________________ drugs.

20. If an antimicrobial drug/agent is able to control pathogenic microorganisms without doing damage to human cells or tissues it is said to have good ______________________________. If it is effective against a wide range of microbes (controls both Gram positive and Gram negative cells) it is called a ______________________________ drug. Although drugs with this characteristic are useful when the identity of the pathogen is unknown, they also present a potential hazard. Why might using such drugs be less advantageous than using one with greater specificity? ____________________________________________

21. The sulfa drugs are synthetic antimicrobial agents that inhibit cell growth by ___________________________.

22. The _____________________________ drugs are synthetic antimicrobial agents which exert their influence by blocking the enzymatic pathway converting ___________________________ ___________________________ within prokaryotic cells.

23. Antimicrobial drugs known as ____________________________ kill actively growing cells by inhibiting the formation of peptidoglycan. Tetracycline and aminoglycosides exert their antimicrobial effects by _____________________________.

24. Penicillin is one example of an antibiotic that kills actively growing bacterial cells by inhibiting the formation of ___________________________. This drug is said to have very good ___________________________ because it acts only on prokaryotic cells and is not damaging to eukaryotic cells (assuming no hypersensitivity reaction is involved).
(2) 25. Penicillins and cephalosporins (β-lactam drugs) are cidal to actively growing cells because they ________________________________. Although these drugs are effective against a variety of pathogenic microbes, some organisms have developed resistance. Organisms that are resistant to penicillins produce a type of enzyme called _______________________________ that allows them to degrade the drugs. Genes encoding this enzyme are often carried on R-factor plasmids.

(2) 26. Two examples of drugs that inhibit protein synthesis are the _______________________________ (which are cidal) and the _______________________________ (which are static).

(3) 27. _______________________________ and _______________________________ are two examples of antibiotics that control pathogens by inhibiting protein synthesis. Bacitracin exerts its effects by disrupting the _______________________________ function of susceptible cells.

(3) 28. A group of drugs known as _______________________________ inhibit protein synthesis by inhibiting the binding of aminoacyl-t-RNA molecules to the ribosomes. Since their effect is not permanent, these drugs are _______________________________ rather than cidal. Why are drugs that only inhibit cell growth (do not kill cells) effective in controlling pathogenic microorganisms? ________________________________________________________________________________

(2) 29. A group of antimicrobial agents known as aminoglycosides kill bacteria by _______________________________ (be specific). These drugs are all antibiotics, but may be produced by different types of bacteria. Streptomycin, Neomycin and Kanamycin are produced by bacteria in the genus _______________________________, while Gentamicin and Amikacin are produced by bacteria in the genus *Micromonospora*.

(2) 30. The Polymyxins are antimicrobial drugs produced (at least initially) by bacteria in the genus _______________________________. They exert their effects by _______________________________ and are usually cidal.

(1) 31. Rifampin and Actinomycin D are two types of antimicrobial drugs that control microbes by inhibiting _______________________________.

(1) 32. _______________________________ is an antimicrobial agent known to inhibit the formation of messenger RNA.

(2) 33. Prescription labels for antimicrobial drugs include the recommended dosage (amount to be taken and how often) and the length of time the drug use is to be continued. Why is it important that these guidelines be followed, i.e., what are the potential consequences of not following them? ________________________________________________________________________________

(1) 34. Why are most of the antimicrobial drugs described in lecture not effective against viruses? ___
Non-specific Resistance (Innate Immunity and Normal Flora):

(5) 1. Define:

- Inflammation
- Pyrogen
- Macrophage
- Interferon
- Bacteriocins

(1) 2. Immune mechanisms are said to be ______________________ or "built in" if they are present at birth, and require no previous exposure to a foreign agent. Most such mechanisms are non-specific.

(1) 3. One of the body's first lines of defense against microbial invasion is the skin. What features of this structure provide a mechanical barrier against infection? ________________________________

(2) 4. The epidermis of human skin provides a chemical barrier against infective agents because it is highly keratinize, ___________________________ and ________________________________.

(3) 5. Human ____________________ is a protective structure that is tough, keratinized, multi-layered and sheds its surface cells on a regular basis. Two chemical aspects of this structure that allow it to serve as an effective barrier against pathogenic microbes are ___________________________ and ________________________________.

(2) 6. Human skin provides the body with a physical barrier to many types of infective agents because it is tough, it has ________________________________ and the surface cells are __________________________ on a regular basis.

(2) 7. Moist surfaces of the body such as those lining the gastro-intestinal, urogenital and respiratory tracts are protected by ________________________________ membranes. In addition to their moist secretions which tend to trap microbes, these surfaces often have lysozyme enzymes that kill microorganisms, IgA type ___________________________ and sometimes cilia.

(2) 8. Mucous membranes produce moist secretions which tend to trap pathogenic microorganisms. In the respiratory system, these trapped microbes may be either killed by enzymes called ________________________________ or "swept" up and out of the airways by structures called ________________________________.
9. The body's second line of defense against foreign agents are cells known generally as ___________________________________ which seek out and consume infective agents. If such cells are agranular, and increase in number is association with chronic infections, they are called ________________________.

10. Neutrophils and monocytes are two types of leukocytes that are ______________________, i.e., able to consume dead cells and bacteria. The largest of these tend to leave the blood stream and take up residence within __________________________ tissues, masses of spongy-like tissues which serve to "filter" blood or lymph.

11. __________________________________ are medium-sized, granular leukocytes that tend to increase in number during acute infections, and are phagocytic.

12. __________________________________ may be defined as an increase in redness, swelling and temperature in an area of traumatized tissue. Much of this response is brought about by the release of ____________________________ a substance that dilates blood vessels and increases capillary permeability.

13. __________________________________ is a protective response characterized by an increase in redness, swelling and temperature in an area of traumatized tissue. This response involves the release of histamine, ___________________________ and leukotrienes by traumatized tissue cells, and increases blood flow to the traumatized area.

14. A protective response characterized by an increase in redness, swelling and temperature in an area of traumatized tissue is called an ________________________ response and involves (among other things) the release of histamine. What function does histamine serve, i.e., how does it help protect the body? ____________________________________________

15. Chemical substances called ___________________________ tend to elevate the temperature within a tissue or within the body overall.

16. Some cells within the human body respond to viral infection by releasing proteins called ______________________________ that act on other cells to inhibit the completion of virus life cycles.

17. ___________________________ are proteins produced by cells infected by virus, that can (when released) travel to other cells and initiate mechanisms within them that inhibit viral life cycles.

18. ___________________________ are plasma proteins that react with one another in a sequential manner to form "holes" through the cell membranes of pathogens or infected body cells.

19. Complement factors are plasma proteins (released by phagocytic WBCs), that can react with one another in a sequential manner. When initiated, the complement “cascade” can cause the formation of ________________________________________________ and can cause ________________________________________.
20. Lymphocytes called Natural Killer cells (NK cells) help provide non-specific defense by releasing a protein called ________________________, that makes holes (pores) through cell membranes, and enzymes called ____________________________, that destroy DNA and proteins within cells, causing apoptosis (cell death from within).

21. Microorganisms that are normally found living in and on the human body are referred to as ______________________ and play a major role in defending the body against pathogens. They do this by taking up ___________________________________________ and by producing chemical substances called bacteriocins.

22. Microorganisms referred to as "normal flora" can be found growing on or within various regions of the human body. These organisms do not usually cause disease, and can actually help defend the body by producing ______________________ and by competing against pathogens for available nutrients and ____________________________________________________.

23. Describe the "defensive" role played by the normal flora associated with the human body. _____________________________________________

**Specific Defense and Immunology:**

1. Define:

   Adaptive or Acquired immunity

   Immunoglobulin

   Epitope

   Helper-T Lymphocyte

   Interleukin

2. An adaptive or acquired immune response (specific immunity), may be actively acquired by ___________________________ or passively acquired by ____________________________________________.

3. Adaptive or acquired immunity (specific immunity), may be acquired (actively and artificially) by ________________________________.

4. Adaptive or acquired immunity may be gained passively through the placenta or breast milk or by receiving _____________________________.

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Adaptive or acquired immunity may be acquired in a number of ways. It is considered to be passive acquisition and natural if immunity is acquired by ____________________________ ___________________________. This type of immunity is available to most young mammals.

Adaptive or acquired immunity involves leukocytes called ____________________________ and may be divided into two categories. That which involves B-cells and the production of antibodies is referred to as ____________________________ immunity, while cellular or cell mediated immunity involves cells that have been processed within the ____________________________ gland.

Adaptive or acquired immunity involves leukocytes called ____________________________ that are produced originally within the red bone marrow. If these cells travel to the ____________________________ gland for processing, they emerge as T-cells and will be involved in cellular or cell mediated immunity.

Lymphocytes (like other blood cells) are produced initially within the ____________________________ by multipotent stem cells. Lymphocytes that travel to the ____________________________ gland for "processing" will participate in cellular immunity.

Adaptive or acquired immunity that involves B-cells and antibodies is referred to as antibody mediated or ____________________________ immunity. The initiation of this immune response involves cells other than B-cells including ____________________________ (that consume antigens and then present antigenic determinants on their cell surfaces) and ____________________________ (that release cytokines that act on the immunocompetent B-cells).

Immunocompetent B-lymphocytes cannot respond to foreign agents without the help of other cells. Name the cells that play a role in activating the B-cells, and explain what they do. ______

The activation of immunocompetent B-cells by specific antigens usually involves the assistance of two other cells types. These are ____________________________ that present antigenic determinants on their cell surfaces, and ____________________________ that bind the antigenic determinants in association with class II MHC proteins and then release cytokines that act on the B-cells.

The initiation of a humoral immune response usually involves the presence of cells other than B-cells. These include phagocytic white blood cells that ____________________________ ____________________________ and ____________________________ that stimulate the proliferation of B-cells into clones that give rise to plasma cells and B-memory cells.

Immunoglobulins are globular and quaternary proteins called ____________________________ that are produced in large quantities in response to specific antigens and which can bind specifically with those antigens.

Quaternary proteins that are produced by the body in response to foreign agents, and can bind specifically with those agents are called antibodies or _____________________________. A sudden increase in their titer following a second or subsequent exposure to the same antigen is called an ____________________________ response and is the basis for vaccination.
(2) 15. Immunoglobulins can be divided into five classes or __________________________ on the basis of the amino acid sequences of their constant regions. Those containing four polypeptides, able to cross the placenta and "fix" complement are called __________________ (be specific).

(1) 16. Immunoglobulins within a given class all have the same __________________________ within the constant regions of their heavy and light chains.

(2) 17. Immunoglobulins designated as isotype ________ can “fix” complement and play an important role in defending the body, but are too large to cross the placenta. Those designated as isotype ________ are involved in hypersensitivity reactions such as hay fever and anaphylaxis.

(2) 18. Foreign agents that enter the body and initiate the production of antibodies (and other immune substances) are referred to as __________________________ and have multiple chemically defined sites on their surfaces to which antibodies can bind. When these agents enter the body for the first time, they are typically consumed and "digested" by __________________________ before they can be recognized by immune cells.

(1) 19. Foreign agents that can enter the body and initiate the production of antibodies are called __________________________ and may be cells, viruses, or large molecules such as microbial toxins.

(2) 20. Foreign agents that enter the body and initiate the production of immune substances are referred to as __________________________ and have multiple chemically defined sites on their surfaces. These chemically defined sites are called antigenic determinant groups or __________________________, and are the sites that antibodies bind.

(2) 21. Serological reactions called __________________________ cause antigens (bacteria or blood cells) to clump together while those called __________________________ render toxic antigens non-toxic.

(1) 22. If an individual immunized against Rubeola virus at an early age were exposed to a live virulent strain of this same virus as an adult, he/she would probably experience an __________________________ response, i.e., a rapid increase in antibody titer, and would not develop disease symptoms.

(1) 23. A rapid increase in antibody titer following the second or subsequent exposure to the same antigen is called an __________________________ response and involves B-memory cells.

(3) 24. Chemical reactions involving the binding of antibodies to antigens in vitro are called __________________________ reactions and indicate how antibodies are able to protect the body. If such reactions cause soluble antigens to become insoluble and to "fall out of solution" they are called __________________________ reactions, and if they prevent pathogenic antigens from binding to cell surfaces, they are referred to as __________________________ reactions.

(2) 25. A retrovirus currently referred to as __________________________ has a devastating effect upon human immune function because it selectively infects (and ultimately kills) __________________________ lymphocytes. In most (if not all) instances, infection with this virus ultimately leads to death.
(1) 26. A cellular or cell mediated immune response cannot be initiated against bacteria or viruses alone, but can be initiated against ____________________________________________ because T-lymphocytes can respond to antigens only if they are found in combination with major histocompatibility complex proteins, and these are only found on eukaryotic cells.

(3) 27. T-cells exert their effects by releasing chemical substances called _____________________________________________. Two such chemicals are ____________________________________________ a substance that stimulates cell proliferation and ____________________________________________ one that kills tumor cells.

(3) 28. The only T-lymphocytes that actually defend the body by attacking and killing infected cells and eukaryotic pathogens are called ____________________________________________. They exert their influence by releasing cytokines called ____________________________________________ and _____________________________________________.

(2) 29. T-lymphocytes do not make antibodies, but exert their effects by releasing chemicals called ____________________________________________. A type of chemical called ____________________________________________ stimulates the activity of phagocytic white blood cells.

(2) 30. Cellular immunity can be directed only against eukaryotic cells (protozoa, fungi, infected cells and tumor cells) because T-cells can respond to antigens only when they are found in combination with _____________________________________________. Why do killer-T lymphocytes not attack and kill phagocytes presenting antigenic determinants from pathogens on their cell surfaces? _____________________________________________.

(5) 31. Matching - Match the terms on the left with the statements on the right.

___ Tumor necrosis factor A. Antibodies binding to toxic antigens (such as tetanus toxin) and rendering them non-toxic.
___ Opsonization B. Cytotoxic substances released by killer-T cells and NK cells; will cause apoptosis and destruction of viral components within cells.
___ Anamnestic response C. Cytokine that cause tumor regression by inducing cell lysis; can also prevent tumor genesis and viral replication.
___ Neutralization D. Cytokine released by T4 lymphocytes that attracts and activates phagocytes, and causes macrophages to kill bacteria.
___ Immobilization E. Rapid increase in antibody titer following a second or subsequent exposure to the same antigen.
___ Isotypes F. Also called human leukocyte antigens, these proteins make eukaryotic cells recognizable as "self" or "non-self".
___ Granzymes G. Can be triggered by complement factors or antibodies binding with antigens, makes particles more attractive to phagocytes.
___ Immune tolerance H. Antibodies binding to the receptor sites of pathogenic cells or viruses and preventing their attachment to host cells.
___ MHC proteins I. Categories or groups of antibodies (immunoglobulins), defined by common amino acid sequences.
___ Gamma interferon J. Our ability to not mount an immune response against "self" molecules, most foods, aerosols and foreign cells such as spermatozoa.
**Immunization and Hypersensitivity:**

(5) 1. Define:

Immunization

Vaccine

Toxoid

Hypersensitivity

Anaphylaxis

(3) 2. ________________ is the process of conferring specific immunity by artificial means and may be accomplished by administering vaccines, ____________________ (detoxified microbial toxins) or ____________________.

(3) 3. ________________ may be defined as the process of inducing active immunity by introducing microorganisms or their products into a host in a non-pathogenic form. A substance that contains killed or attenuated microorganisms is called a ________________, while a substance made from detoxified microbial toxin is called a ________________.

(2) 4. Immunization is considered appropriate for the mass of the population if ___________________________ and ________________________________

__________________________________________________________________________.

(2) 5. Two risks that may be associated with immunization include:

1) __________________________________________________________________________

2) __________________________________________________________________________

(3) 6. Although a number of important human diseases can be prevented by immunization, there are some potential risks associated with this procedure. Sometimes people who receive vaccines fail to develop immunity, sometimes hypersensitivity reactions are initiated. In some places, vaccines, toxoids or instruments may be ________________, and vaccines made with Gram negative bacteria can be ________________. Live viral vaccines should not be given to women during pregnancy because the virus may ________________.

(1) 7. An abnormal physiological state during which an immune reaction causes tissue damage or malfunction is referred to as ________________ or as an allergic reaction.
8. Hypersensitivity reactions that occur within minutes of exposure to the allergen are categorized as __________________ reactions and involve _______________________________.

9. Abnormal immune reactions that result in tissue damage or malfunction are known as __________________ reactions and may involve either antibodies or T-cells. That which can cause the destruction of fetal RBCs by initiating the action of complement proteins is referred to as a __________________________ response.

10. __________________________ is a type I hypersensitivity reaction involving immunoglobulins in the class _________ and the release of histamine body wide.

11. Anaphylaxis is an immediate hypersensitivity reaction that is initiated when allergens bind to ___________ attached to mast cells (be specific) throughout the body. The mast cells are then stimulated to release ________________ which causes life threatening changes within the circulatory system. This reaction can lead to circulatory shock within minutes.

12. A type I hypersensitivity reaction that is localized, i.e., tends to affect only the eyes, nasal passages or skin is referred to as ________________ and is usually not life threatening.

13. A type II hypersensitivity reaction known as a cytotoxic response involves IgG and plasma proteins known as _______________________________ factors.

14. During a type II hypersensitivity reaction known as a cytotoxic response, a mother carrying an Rh-positive fetus produces ___________________________ that cross the placenta and stimulate the activity of ___________________________. These bind to the fetal RBCs and cause them to lyse (by forming holes in their cell membranes).

15. If a person with type O, Rh-negative blood received a transfusion containing packed RBCs obtained from a person with type AB, Rh-negative blood, they would experience a ___________________________ due to the binding of anti-A and anti-B antibodies with the transfused cells, and the release of ________________.

16. A type III hypersensitivity reaction that is localized is called a/an __________________________ reaction, while one that occurs body wide is referred to as ___________________________.

17. Immediate hypersensitivity reactions that involve IgG, IgM and complement factors cause a set of symptoms referred to as ___________________________ disease.

18. A ___________________________ reaction is one for which symptoms often occur 24 or more hours after exposure to the allergen. Reaction to poison oak would fall into this category and involves the activity of T8 lymphocytes known as ___________________________.

19. People who receive organ transplants or tissue samples from other individuals must be immunosuppressed. This is because the rejection of such organs or tissues involves what?

20. Although immunizations can sometimes cause hypersensitivity reactions, the most common causes of allergic reactions are ___________________________.

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(4) 21. Matching - Immunology and hypersensitivity: Match the term or terms on the left with the most appropriate statements on the right, and place the letters of correct matches in the blanks provided.

<table>
<thead>
<tr>
<th>Term</th>
<th>Statement</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anaphylaxis</td>
<td>A. Substance which contains killed or weakened (attenuated) microorganisms.</td>
</tr>
<tr>
<td>Antiserum</td>
<td>B. Type III hypersensitivity reaction which tends to be localized within the body.</td>
</tr>
<tr>
<td>Cytotoxic response</td>
<td>C. Blood plasma that lacks clotting factors and contains a high titer of antibody.</td>
</tr>
<tr>
<td>Hypersensitivity</td>
<td>D. Life threatening hypersensitivity reaction that involves IgG and mast cells that release histamine body wide.</td>
</tr>
<tr>
<td>Vaccine</td>
<td>E. Hypersensitivity reaction that involves IgG and complement in the destruction of fetal RBCs</td>
</tr>
<tr>
<td>Type IV</td>
<td>F. Substance containing detoxified microbial toxin, and used to stimulate immunity against that toxin.</td>
</tr>
<tr>
<td>Arthus reaction</td>
<td>G. Reactions that involve T-cells and cytokine and occur 24-48 hours or more after exposure to the allergen.</td>
</tr>
<tr>
<td>Toxoid</td>
<td>H. Any abnormal immune reaction that causes tissue damage or malfunction within an individual.</td>
</tr>
</tbody>
</table>

**Epidemiology and Disease Transmission:**

(5) 1. Define:

Epidemiology

Endemic

Reservoir

Zoonosis

Morbidity rate

(2) 2. __________________________________ may be defined as the quantitative study of the occurrence of disease, and factors that influence disease frequency and distribution. What is the primary goal of people working in this field? __________________________________

(1) 3. The study of disease dealing with etiology, pathogenesis and the anatomical and physiological changes occurring within individuals experiencing disease is called ______________________.
4. Since epidemiology is a quantitative study, it involves the collection and analysis of large quantities of data, and the making of recommendations based on the information gathered. In the United States, the agency responsible for data collection, analysis and reporting (recommendations) is the ______________________________________ with it’s headquarters in Atlanta, Georgia. The agency responsible for coordinating similar activities throughout the world is the ______________________________________________ with it’s headquarters in Geneva, Switzerland.

5. A disease that tends to affect a relatively small percentage of a population, but at a constant rate, is said to be ______________________ to that population (an example would be cholera in Southeast Asia). A disease that occurs in small, localized, unpredictable outbreaks would be referred to as being __________________________ (Legionnaire’s disease has this characteristic).

6. Diseases such as chicken-pox and measles tend to be endemic to human populations that have not received immunization; however, when the number of new cases rises significantly above the expected “background” level, the disease is said to have become an ___________________________________________. If the disease is wide spread, such that it threatens people of many nations on more than one continent, it is said to be __________________________ (currently, AIDS falls into this category).

7. Air and water are non-living reservoirs that tend to pick up potential pathogens from living reservoirs and from _________________, a non-living reservoir that supports the growth of many microorganisms. Since air and water tend to carry microbes, sometimes for long distances, they can also be considered as _______________________ involved in disease transmission.

8. Matching - Epidemiology: Match the term or terms on the left with the most appropriate statements on the right, and place the letters of correct matches in the blanks provided.

   ____ Sporadic A. Combs, utensils, clothing, and other small items involved in disease transmission.
   ____ Mortality B. The sum of all potential sources for a specific disease agent.
   ____ Reservoir C. Disease which occurs in small isolated unpredictable outbreaks.
   ____ Morbidity D. Number of individuals dying from a disease within a given population and time period.
   ____ Vector E. Number of cases of disease is significantly above the expected "background" level.
   ____ Pandemic F. Epidemic threatening several nations on more than one continent.
   ____ Fomite G. An animal (usually an arthropod) which is involved in disease transmission.
   ____ Epidemic H. A disease usually associated with non-human animals but which can be transmitted to man.
   ____ Endemic I. Disease which affects a small % of the population on a fairly constant basis.
   ____ Zoonosis J. Number of individuals infected within a given population and time period.
9. Small objects such as utensils, clothing, bedding, toiletries and money may be involved in the
transmission of disease causing agents, and so are considered as _________________.

10. Non-human animals often play an important role in disease transmission because they serve as
__________________________. A disease which is normally associated with non-human
animals but which can be transmitted to man is called a _________________.

11. __________________________________________________ play a dual role in disease transmission
because they serve both as reservoirs and as vectors.

12. Sometimes virulent pathogens can colonize an individual without causing disease symptoms. When
this happens, the person involved becomes a reservoir and may transmit the pathogen
without being aware of it. Mary Mallon was such an individual, and became a carrier of
__________________________. She was unwittingly transmitting the etiological agent to
_________________________ and infected at least thirty people.

13. Smallpox was an important human disease that is now considered to have been vanquished. People
are no longer immunized against it because the virus has been eliminated from the
human population. Why is it highly unlikely that diseases such as bubonic plague, lyme disease
and rabies will be similarly eradicated?

14. __________________________ transmission is that which involves contact between a
susceptible host and a living reservoir.

15. If you were to acquire a Streptococcus infection by running barefoot on an ocean beach and
stepping on broken glass, the mode of transmission involved would be
__________________________.

16. Etiological agents require __________________________ transmission if they are very sensitive to
environmental factors such as drying, sunlight and variation in temperature.

17. The severity of epidemics within human populations is known to be influenced by a number of
factors including: 1) the number of individuals that been immunized, 2) the
___________________________ of the population, 3) the age,
nutritional status and general health of the population, 4) the__________________________
of the people, 5) the degree of exposure to the pathogen involved, and 6) the
___________________________.

18. Three methods used by epidemiologists in an attempt to prevent disease are: 1) by increasing
___________________________, 2) by decreasing the number of ________________________________
available, and 3) by segregating or restricting the interactions between
infected and non-infected persons.

19. The number of cases of a given disease within a specified population and time period is referred
to as the __________________________ rate. The number of people dying from that disease
within the same population and time period would be the __________________________ rate
Pathology, Mechanisms of Pathogenicity, and Disease Causing Agents:

(5) 1. Define:

Pathology

Latent disease

Septicemia

Leukocidin

Tetanospasmin

(2) 2. __________________________ may be defined as the science or study of disease and deals with the etiology, pathogenesis and the structural and functional changes brought about by disease.

(2) 3. __________________________ may be defined as a condition in which a pathogenic microorganisms has invaded or colonized a portion of the body (cells or tissues). If the presence and activities of these microbes causes a change in the overall health of the individual they have colonized, they are said to be causing a ____________________.

(1) 4. Many diseases that are caused by microorganisms are __________________________, i.e., readily passed from one individual to another, either directly or indirectly.

(2) 5. A disease is categorized as __________________ if it’s symptoms develop slowly, over a long period of time, and it persists within the body for months or years. If the symptoms of a disease develop rapidly and last a short period of time, the disease is said to be ________________.

(2) 6. __________________________ may be defined as the condition of having viable bacteria in the blood stream and does not necessarily indicate a disease state. If the bacteria present are reproducing and causing disease (septic) symptoms, the condition is termed ________________.

(2) 7. When bacteria are present within the bloodstream and are reproducing and causing septic symptoms, the condition is known as __________________________. If only the toxic products of bacteria are present in the bloodstream, the condition is known as ____________________.

(1) 8. Scarlet fever, rheumatic fever, diphtheria and tetanus are diseases that result from __________________________, i.e., the presence of bacterial toxins within the blood stream.
9. A single-celled microorganism that colonizes some part of another organism, and by direct interaction with that organism causes disease, is referred to as a _____________________.

10. ______________________________ may be defined as a degree of pathogenicity and may be expressed in terms of ID50 or LD50.

11. The dose or concentration of a pathogen that is required to infect 50% of a test population is referred to as the ______________________________. The dose or concentration expected to kill 50% of the test population is called the ______________________________.

12. In order to cause disease, pathogenic microorganisms must first gain access to the body. Those which enter via a cut, puncture, bite or other wound are said to require the _________________ route as a portal of entry. Pathogenic bacteria must also be able to __________________ faster than the body can eliminate them.

13. The virulence of pathogenic microbes is, to some extent, dependent upon their ability to avoid the body's immune responses. Explain two ways in which microbes may avoid or overcome normal immune mechanisms:
   a) _________________________________________________________
   b) _________________________________________________________________________

14. Bacteria produce a variety of enzymes that alter host function and so aid in their pathogenicity. These include enzymes called _____________________ which destroy erythrocytes (RBCs) and _______________________________ which degrade one of the components of dense connective tissue thus increasing the pathogens invasiveness.

15. Bacterial exotoxins are proteins which tend to have very specific actions within the body. Name two types of exotoxins and describe briefly what they do.
   a) _________________________________________________________________________
   b) _________________________________________________________________________

16. The exotoxins produced by ordinary Gram negative bacteria are all similar to one another in that they are made up of _____________________________________ that are part of the outer membrane of the bacterial cell wall. These toxins all tend to cause similar symptoms including: _________________________________________________________________________

17. Pathogenic bacteria in the genera Streptococcus and Staphylococcus are often highly virulent because they produce a number of factors that allow them to avoid body defenses and weaken host resistance. These bacteria produce _______________________________ (respectively) that allow them to avoid phagocytic WBCs, and they also produce _______________________________ that kill WBCs. They form hemolysins that destroy host RBCs and they form _______________________________ that break down fibrin and increase their invasiveness.

18. Bacteria in the genus Clostridium are responsible for both tetanus and botulism. Although the symptoms of these diseases are quite different, they are both caused by protein exotoxins that are nearly identical in structure. Describe the symptoms characteristic of botulism and tetanus, name the target (host) cells involved, and explain the action of the toxin in each case.
19. ___________________________________________________________________________ is the name given to Gram positive pyogenic cocci that are known to cause a variety of infections (carbuncles, furuncles, toxic shock syndrome, endocarditis, etc.) as well as bacterial intoxication or food poisoning due to a heat stable enterotoxin.

20. Gram negative, facultatively anaerobic bacteria identified as ______________________________ are sometimes associated with diarrhea, but can also cause bladder and kidney infections, meningitis (in neonates) and hemolytic uremic syndrome.

21. New warning labels have recently been applied to meat due to the actions of a virulent strain of ___________________________________________________________________________. These Gram negative enterics are also commonly associated with bladder and kidney infections.

22. Gram negative, aerobie bacteria identified as ___________________________________________________________________________ produce a blue-green pyocyanin pigment as well as toxins that inhibit protein synthesis. These bacteria may cause pneumonia, meningitis and infections of the skin, eyes, ears, etc.

23. Many bacteria live in the mouth and upper respiratory tract without causing much harm to their host. However, the acid produced by some bacteria can degrade tooth enamel and cause ____________________________________________________________________________, and bacteria are also a major cause of ____________________________________________________________________________.

24. Bacteria are known to cause a variety of problems in association with the mouth (gums and teeth). Name and briefly describe two such problems caused by mouth bacteria.
   a) __________________________________________________________________________
   b) __________________________________________________________________________

25. Three reasons why young sexually active females should be particularly concerned about sexually transmitted diseases are: 1) their __________________________ are not obvious and often go unnoticed, 2) they can cause __________________________ and 3) they leave their host much more susceptible to infection with ____________ and therefore a wide variety of other disease agents.

26. Sexually transmitted diseases are a much more significant threat to females than they are to males for a number of reasons. Name any two: __________________________________________________________________________
   __________________________________________________________________________
**Bacterial Diseases and Agents:**

(20) 1. Matching - Bacterial Diseases and Agents:

Match the disease on the left with the bacteria most likely to be the causative agent. **NOTE** that the genus and species names are in separate columns, so it will be necessary to place two letters in each blank provided. Genus names require capital letters (A-P); species names small letters (a-t).

<table>
<thead>
<tr>
<th>Disease</th>
<th>A. Bacillus</th>
<th>a. pyogenes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dysentery</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gas gangrene</td>
<td>B. Francisella</td>
<td>b. pertussis</td>
</tr>
<tr>
<td>Diphtheria</td>
<td>C. Mycobacterium</td>
<td>c. anthracis</td>
</tr>
<tr>
<td>Pneumonia</td>
<td>D. Corynebacterium</td>
<td>d. tetani</td>
</tr>
<tr>
<td>Typhoid</td>
<td>E. Chlamydia</td>
<td>e. tularensis</td>
</tr>
<tr>
<td>Scrub typhus</td>
<td>F. Shigella</td>
<td>f. perfringens</td>
</tr>
<tr>
<td>Leprosy</td>
<td>G. Yersinia</td>
<td>g. menengitidis</td>
</tr>
<tr>
<td>Cholera</td>
<td>H. Bordetella</td>
<td>h. gonorrheae</td>
</tr>
<tr>
<td>Gonorrea</td>
<td>I. Streptococcus</td>
<td>i. tuberculosis</td>
</tr>
<tr>
<td>Tularemia</td>
<td>J. Borrelia</td>
<td>j. pallidum</td>
</tr>
<tr>
<td>Tetanus</td>
<td>K. Vibrio</td>
<td>k. pneumonias</td>
</tr>
<tr>
<td>Tuberculosis</td>
<td>L. Rickettsia</td>
<td>l. pneumophila</td>
</tr>
<tr>
<td>Syphilis</td>
<td>M. Pasteurella</td>
<td>m. tsutsugamushi</td>
</tr>
<tr>
<td>Meningitis</td>
<td>N. Treponema</td>
<td>n. typhi</td>
</tr>
<tr>
<td>Lyme disease</td>
<td>O. Neisseria</td>
<td>o. burgdorferi</td>
</tr>
<tr>
<td>Plague</td>
<td>P. Legionella</td>
<td>p. diphtheriae</td>
</tr>
<tr>
<td>Legionnaire's disease</td>
<td>Q. Orientia</td>
<td>q. cholerae</td>
</tr>
<tr>
<td>Anthrax</td>
<td>R. Staphylococcus</td>
<td>r. pestis</td>
</tr>
<tr>
<td>Whooping cough</td>
<td>S. Clostridium</td>
<td>s. leprae</td>
</tr>
<tr>
<td>Scarlet fever</td>
<td>T. Salmonella</td>
<td>t. dysenteriae</td>
</tr>
</tbody>
</table>
Directions: Read each question carefully and completely before you write your answer. Note that the point values for all questions are listed in the margin, and that those requiring observation of laboratory preparations are marked with a * symbol. Answer all questions as specifically as possible, and DO NOT BE VAGUE - GOOD LUCK!

(3) 1. Define:

Transient microbiota

Microbial enrichment

Cultured food

(1) 2. The laboratory disinfectant used in this laboratory is applied to the bench surface when, and for what purpose? _________________________________________________________________ _________________________________________________________________

(1) 3. Students consuming food materials, snacks, gum, drinks, etc. in the microbiology laboratory are breaking a basic safety rule; i.e., no food or drinks in the micro lab. What potential hazard does this rule address? _________________________________________________________________

(2) 4. The Bunsen burners used in this laboratory represent a potential hazard to microbiology students because their flames are difficult to see in the room light, and they are frequently lit. Describe at least two precautions students were given to prevent accidental burns when using these pieces of equipment. _________________________________________________________________

(1) 5. If you had just been splashed in the face with crystal violet, and you suspected the chemical had entered your eyes, where would you go within the laboratory, and what would you do there?

______________________________________________________________

(1) 6. Student working in this laboratory were told to exit the room, pass through the hallway to the left, walk down the concrete steps and gather on the lawn in the event of a ____________________.
7. Assuming this collection of used materials is ready for discard, indicate by placing letters in the correct blanks, where you would dispose of each item.

- In the large metal bins on the bottom shelf inside the discard cabinet.
- On the top or bench surface of the discard cabinet.
- In the plastic racks on the upper shelf inside the discard cabinet.
- In the small metal bin on the upper shelf inside the discard cabinet.

8. According to information provided by the Centers for Disease Control and Prevention (CDC), ___________________________ is the single most effective procedure for preventing nosocomial infections. Name one individual recognized as a pioneer in this arena, i.e., a person who strongly advocated physicians use this procedure. ____________________________________

9. Each compound microscope used in this laboratory is equipped with four _________________ lenses mounted on a revolving nosepiece, and two _________________ lenses (located nearest the viewers eyes). What are the four degrees of magnification possible with this dual lens system? ______________________________________________________________________

10. The nosepiece on each of our compound microscopes is spring-loaded, and can be moved by grasping it with the fingers and gently lifting it upward. Students applying _________________ to a slide on the microscope stage were told to use this method for moving the revolving nosepiece out of the way, because it minimizes the extent of _________________ adjustment required to bring the image sharply into view once the lens is repositioned.

11. When using a compound microscope, students will notice that the _________________ and _________________ of their viewing field will increase as magnification is decreased.

12. Immersion oil is applied to prepared slides being viewed with the 100X lens because it prevents the _____________________________________, thereby increasing resolution and allowing tiny objects such as bacteria to be seen clearly. Before any microscope is put away, lenses must be cleaned with optical lens wipes, but Kimwipes or VWR light-duty tissue wipers are also available. These can be used to clean what? _________________________________________

13. Student using the compound microscopes to view bacteria in this laboratory were instructed to begin focusing while using which lens, and why? _____________________________________

14. The small silver knob on the left-hand side of your microscope can be used to adjust the position of the _________________ lens, and thereby increase or decrease the amount of light focused on the specimen. What are the disadvantages of viewing specimens using very high light intensity, i.e., very bright light? ____________________________________________

15. The measuring device located within the eyepiece of a compound microscope is called a/an ___________________________ lens, and is a circle of glass with two scales etched into its surface. The unit values for these scales must be determined for each power of magnification using a process called _________________________, because the unit values change as magnification is changed.
16. What is the size (length and diameter) of the cells shown here? (Be sure to check the power of magnification being used, and to include units.) _____________________________________

17. A mixture of materials designed to provide all the nutrients necessary for the growth of microorganisms in vitro is called ______________________ and in this laboratory is sometimes solid and sometimes broth. If this mixture contains nutrients in pure chemical form and in specified quantities, it is called a ______________________. Assuming you wanted to make 100ml of the material indicated by the label on this container, you would add _____________ of dry powder to 100ml of water, and mix.

18. Although most of the bacteria growing in this laboratory are chemoheterotrophs that must be provided with essential elements, many soil organisms including Rhizobium, Agrobacterium and Anabaena can fix ______________________ from the atmosphere and use it in the formation of amino acids, nucleotides and peptidoglycan. Cyanobacteria and other types of autotrophs can obtain the ______________________ they need from air.

19. Students working in the microbiology laboratory were advised to apply ______________________ whenever they were manipulating live microbial cultures. These procedures were designed to prevent infection and avoid contamination of the cultures being used. Describe the proper procedures as they are applied to each of the following:
   Wire loops _________________________________________________________________
   Petri plates _______________________________________________________________

20. The mouths of glass culture tubes are rolled in the flame of a Bunsen burner before and after each microbial transfer (into or out of the tube). What is this action expected to accomplish? _________________________________________________________________

   What were students told to do with the plastic caps removed from glass culture tubes during the time the tube mouth was being flamed? _________________________________________________________________

21. Which of the plates shown here has been properly streaked? ________ What corrections would you make to this plate's label? _________________________________________________________________

22. Students working in this laboratory were told to always incubate agar plates in what position? ______________________. The primary reason for storing Petri plates in this position is to prevent condensation from collecting on the lid and dripping onto the agar surface. Why would this matter? _________________________________________________________________

23. What is a pure microbial culture? (Remember a pure culture can exist in broth or on an agar plate) _________________________________________________________________

   Which of the plates shown here contains a pure culture? ________________

24. ______________________ can be defined as the study of external features, and applies to both cells and colonies. Describe four cultural characteristics of the colonies presented here. _________________________________________________________________
25. Bacteria are most commonly killed and stained prior to being viewed with light microscopes. One advantage of staining is that it increases the ____________________ between cells and their background, making them easier to see. What additional advantages are gained by viewing dead stained cells rather than living ones?

26. Stain techniques that color the cells present, leaving the background uncolored or white in appearance are called ______________________ stains, while those that color the background, leaving the cells uncolored are called ______________________ stains.

27. Basic stains including crystal violet, methylene blue and safranin have their color associated with charged particles called ____________________. Why do these stains bind cell surfaces and not the glass slides the cells are bound to?

28. Nigrosin preparations are more likely to provide an accurate indication of cell ______________________ than will Gram stains or Acid-fast preparations, because the cells are not distorted by heat or alcohol before the stain is set.

29. Bacteria being prepared for Gram or acid-fast staining are typically mixed with liquid (usually water), and then the smear is ______________________ before the primary stain is applied. Both the Gram stain and the acid-fast stain are examples of ______________________ stains, i.e., ones that will cause cells to look different even though all cells are treated the same way.

30. In this stain preparation the larger of the two cell types indicated are human cheek cells and are ______________________ type cells, the smaller cells are _______________________.

31. Almost all bacteria have cell walls containing ______________________, a unique complex of polysaccharides and amino acids. The polysaccharides are made up of two sugars called N-acetyl muramic acid and ______________________. An outer membrane of complex composition surrounds a typical Gram-negative bacterium. This membrane has clinical significance because the ______________________ is toxic to mammalian cells and can cause considerable damage to humans even if the bacteria are dead.

32. What is the function of the Gram's iodine solution applied to cells during the Gram staining procedure? ______________________. If you had prepared a Gram stain using a Gram-negative culture, had correctly applied the first three reagents, but forgot to apply the last one, what would you expect to see and why?

33. In this laboratory we sometimes mix 3% potassium hydroxide with cell samples placed on clean glass slides as an alternative method for determining wall composition. Cells capable of forming thick (multi-layered) walls without outer membranes would do what when mixed with 3% KOH?

34. Describe the shape and Gram stain quality (positive VS negative) of the cells demonstrated. Shape = ______________________ Gram stain characteristic = ______________________.
35. These cells were stained with a technique known as the _______________________ stain. The cells retaining carbol fuchsin differ from the others present in that their cell walls contain a high percentage of a wax-like lipid called ________________________ acid. This substance makes the cells hard to stain, but very resistant to being decolorized.

36. Identify the special (unique) cell structures represented in each of these slides. (Note: - the techniques used to stain these structures were all very different from one another.
   a) _______________________  b) _______________________  c) _______________________ 

37. The dormant structures within the cells represented here were stained over steam heat with a stain reagent called ___________________________. What is the shape and location of the dormant structures? __________________________________________________________
   __________________________________________________________

38. Name the three genera we obtained from soil using enrichment procedures, explain what techniques or special media were used in each case, and explain why these were effective, i.e., what characteristics of the organisms were taken advantage of in order to separate them from other forms present in soil.

Note that for all identifications beyond this point you may gain one extra credit point for each correct species name given, with an overall maximum of five points possible.

39. The organisms shown here are oxygenic photoautotrophs sometimes referred to as blue-green algae. They are classified within the Domain ______________________ and within the phylum ____________________________. Identify the two examples shown as to genus:
   a) ___________________________  b) ___________________________

40. Fungi can be divided into three groups or categories based on their morphology, but in this laboratory, we were primarily interested in single-celled forms called _______________________ and the fuzzy-looking forms called _______________________ that form microscopic hyphae and mycelia that extend only short distances into the air or substrate they are growing in/on.
(1) 41. Fungi previously categorized within the phylum Deuteromycota (fungi imperfecti) were believed to reproduce only by asexual means, i.e., they were always in the _________________________ state. Penicillium and Aspergillus are two examples of fungi initially categorized this way.

* (2) 42. The fungus present on this slide is in the genus __________________________. Which type of reproductive process (sexual VS asexual) is indicated by the structures at the pointer tip, and what name is given to these spores? ________________________________

* (3) 43. The fungus represented here belongs to the genus __________________________, and the structures indicated at the pointer tip are called __________________________. These are produced via __________________________ reproduction.

* (6) 44. Identify the fungi represented here as to genus and phylum as indicated below:

<table>
<thead>
<tr>
<th>Genus name</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>b) __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>c) __________________________</td>
<td>__________________________</td>
</tr>
</tbody>
</table>

* (4) 45. These organisms belong to the phylum __________________________. Identify them as to genus:

| a) __________________________ | b) __________________________ |
| c) __________________________ |

* (4) 46. Identify these unicellular algae as to genus (or group name) and phylum as indicated below:

<table>
<thead>
<tr>
<th>Genus (or group name)</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>b) __________________________</td>
<td>__________________________</td>
</tr>
</tbody>
</table>

* (6) 47. The organisms shown here are all classified within the kingdom __________________________ and have layers (skeletons or walls) containing __________________________ outside their cell membranes. Provide the common names and phyla of these organisms.

<table>
<thead>
<tr>
<th>Common name</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>b) __________________________</td>
<td>__________________________</td>
</tr>
</tbody>
</table>

* (6) 48. Identify the following as to genus and phylum as indicated below:

<table>
<thead>
<tr>
<th>Genus</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a. __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>b. __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>c. __________________________</td>
<td>__________________________</td>
</tr>
</tbody>
</table>

* (2) 49. The organisms shown here belong to the genus __________________________, and to the class __________________________. They enter their host with the aid of an infected mosquito.

* (5) 50. These organisms belong to the phylum __________________________. Identify them as to genus and class.

<table>
<thead>
<tr>
<th>Genus</th>
<th>Class</th>
</tr>
</thead>
<tbody>
<tr>
<td>a. __________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>b. __________________________</td>
<td>__________________________</td>
</tr>
</tbody>
</table>
* (3) 51. These organisms belong to the phylum ___________________. Identify them as to genus a.) ___________________________ and b.) ___________________________

* (6) 52. These organisms belong to the Kingdom ___________________ and to the phylum ___________________. Identify each of them as to genus.  
   a.) _________________________________  b.) _________________________________  
   c.) _________________________________  d.) _________________________________

* (3) 53. This sauerkraut was made by creating an enrichment designed to promote the growth of bacteria in the genera ________________________ and __________________________, while inhibiting the growth of fungi and less desirable bacteria. What fermentation product caused the pH within the inner beaker to change from 7 to 4? _________________________________

* (3) 54. The primary fermentation product remaining in association with this apple wine is a volatile liquid called ________________________. The _______________________________ (second fermentation product) initially formed by the microorganisms present was allowed to escape through the rubber tubing. What organisms were added to the apple juice in order to produce the substances named above? ________________________________.

* (1) 55. What fermentation product formed by organisms present in both buttermilk and yogurt caused the pH indicator in this medium to change color? ________________________________

1) 56. According to the cheese chart supplied within the laboratory syllabus, the bacteria most commonly used in the production of cheese are ___________________________.

1) 57. The production of cheese typically involves adding live bacteria and enzymes to milk, warming the mixture to about 37° C, and allowing it to stand for about one hour. Following this, the solidified milk protein is cut into cubes and heated until it constricts. The solid and liquid portions of the resulting mixture are called ________________________________ respectively, and these are usually separated from one another with cheesecloth.

2) 58. Soft cheeses contain between 50 and 80% _________________ while hard cheeses contain less than 40%. How do ripened cheeses differ from unripened cheeses? ____________________________

2) 59. The bacteria most commonly used in the production of yogurt include ________________________ and one or more different species of Lactobacillus. The sour flavor characteristic of yogurt is due to the lactic acid made by these bacteria, but most fresh yogurts also have a subtle nut-like flavor due to the ___________________________ present. This second fermentation product has a neutral pH.

1) 60. Describe one health benefit gained from the consumption of fresh yogurt in addition to the benefit gained from obtaining the protein associated with milk. ________________________________
1. Define:

Calibration

Fungi

Fermented food

2. Students working in this laboratory were advised to apply ____________________________ to the bench surface before beginning and after completing each laboratory session. Additional applications would be appropriate in the event of spills involving live microbial cultures.

3. Many of the bacteria used in this laboratory are potential gastrointestinal pathogens; therefore, students are reminded they are not allowed to ____________________________ in this room. The application of chapstick or lipstick; nail biting, pencil chewing and pipetting liquids by mouth, are also strongly discouraged.

4. Students working in the microbiology laboratory were advised to keep their ____________________________ near the center of their workbench, to never leave these unattended while lit, and to ____________________________ .

5. If you had just been splashed in the face with crystal violet, and you suspected the chemical had entered your eyes, where would you go within the laboratory, and what would you do there?

_____________________________________________________________________________
_____________________________________________________________________________

6. In the event of an emergency evacuation of this laboratory, students were told to exit through the door nearest the office, enter the hallway, turn in which direction and gather where? __________

_____________________________________________________________________________
7. Assuming this collection of used materials is ready for discard, indicate by placing letters in the correct blanks, where you would dispose of each item.

__________ In the large metal bins on the bottom shelf inside the discard cabinet.
__________ On the top or bench surface of the discard cabinet.
__________ In the plastic racks on the upper shelf inside the discard cabinet.
__________ In the small metal bin on the upper shelf inside the discard cabinet.

8. Joseph Lister and _____________________________________ were two individuals recognized for strongly advocating physicians wash their hands prior to surgical procedures and between patients. Currently the Centers for Disease Control and Prevention (CDC), recognizes handwashing as the single most effective procedure for preventing what? ________________

9. Bacteria not normally present on the surface of human skin, but picked up through contact are categorized as ____________________ microbiota, and according to the data obtained in this laboratory, are readily removed by thorough washing with plain soap and water.

10. Each compound microscope used in this laboratory is equipped with two ___________________ lenses (located nearest the viewers eyes) and a set of four ________________________ lenses mounted on a revolving nosepiece. What maximum degrees of magnification (four) are possible with this dual lens system? ________________

11. The nosepiece on each of our compound microscopes is spring-loaded, and can be moved by grasping it with the fingers and gently lifting it upward. Students applying _________________ _________ to a slide on the microscope stage were told to use this method for moving the revolving nosepiece out of the way, because it minimizes the extent of _______________________ adjustment required to bring the image sharply into view once the lens is repositioned.

12. The diameter and depth of the viewing field visible with a light microscope will both change as the degree of magnification is changed. In what ways do these features change as magnification is increased? __________________________________________________________________

13. Student using the compound microscopes in this laboratory were instructed to begin focusing on prepared slides containing bacteria, while using the ________________ objective. They were told to skip the "high dry", and apply immersion oil when using maximum magnification. What is the function of the immersion oil? ____________________________________

14. Indicate by letter which of the materials shown would be: 1) most appropriate for cleaning microscope lenses _________, 2) most appropriate for cleaning the microscope stage and/or prepared slides _________. Assuming all cleaning has been completed, explain what additional changes are required before returning this instrument to the cabinet. ______________________

15. The measuring device located within the eyepiece of a compound microscope is called a/an ________________________, and is a circle of glass with two scales etched into its surface. What is the size (length and diameter) of the cells shown here? (Be sure to check the power of magnification being used, and to include units.) _____________________
16. Light intensity can significantly influence image quality when using a compound microscope; therefore, light can be adjusted in three ways. The ______________________________ can be opened or closed using the metal lever under the front edge of the stage, the ______________________________ lens can be elevated or lowered using the small silver knob on the left-hand side of the microscope, and the light switch may have a rheostat. Students will experience what disadvantage, in addition to poor image quality, when using very high light intensity, i.e., very bright light? _______________________________________

* 17. These containers hold examples of ______________________________, i.e., mixtures of materials designed to provide all the nutrients necessary for the growth of microorganisms in vitro (sometimes as broth, and sometimes as solids). Assuming you wanted to make 100ml of example "A", the label indicates, you would add _______ grams of dry powder to 100ml of water, and mix. Would the resulting material be categorized as defined or would it be complex? (The correct answer here is not yes or no.) _______________________

18. Most of the bacteria grown in this laboratory are chemoheterotrophs, so must be provided with essential elements; however, soil organisms including Rhizobium, Agrobacterium and Anabaena can obtain the element _______________________ from the atmosphere, and autotrophs such as Nostoc and Anabaena can obtain the element _____________________ they need from air.

19. __________________________________ can be defined as a set of procedures applied in order to prevent infection and avoid contamination during the manipulation of live microbial cultures. Describe how this is applied to each of the following:
   Wire loops __________________________________________________________________
   Mouths of glass culture tubes ___________________________________________________
   Plastic snap-on caps ___________________________________________________________

20. Petri plates opened for inoculation, or during the time cultures are being transferred, should be exposed to room air for a minimal period of time. Why is this precaution necessary? ____________

* 21. Which of the plates shown here has been properly streaked? _________ What corrections would you make to this plate's label? __________________________________________

22. Students working in this laboratory were told to always incubate agar plates in what position? __________________________________. The primary reason for storing Petri plates in this position is to prevent condensation from collecting on the lid and dripping onto the agar surface. Why would this matter? __________________________________

* 23. A microbial culture is called a _________________ culture if it contains only one population, or one type of organism. Which of these plates contains such a culture? _________________

* 24. ______________________________ can be defined as the study of external features, and applies to both cells and colonies. Describe four cultural characteristics of the colonies presented here.

________________________________
(2) 25. Bacteria are most commonly viewed when dead and stained because they can be made to contrast with their backgrounds, they cannot ______________________________________ and are no longer capable of causing ____________________________________.

(2) 26. Stain techniques/preparations that color the background, leaving the cells uncolored (they appear white against a dark background) are called ________________________ stains, while those that color the cells present, leaving the background uncolored or white in appearance are called ________________________ stains.

(2) 27. Basic stains including crystal violet, methylene blue and safranin have their color associated with charged particles called ____________________. Why do these stains bind cell surfaces and not the glass slides the cells are bound to? ____________________________________________

(1) 28. Nigrosin preparations are more likely to provide an accurate indication of cell __________________________ than will Gram stains or Acid-fast preparations, because the cells are not distorted by heat or alcohol before the stain is set.

(2) 29. Bacteria being prepared for Gram or acid-fast staining are typically mixed with liquid (usually water), and then the smear is __________________________________________ before the primary stain is applied. Both the Gram stain and the acid-fast stain are examples of ________________________ stains, i.e., ones that will cause cells to look different even though all cells are treated the same way.

* (2) 30. The larger of the two cell types indicated here are human epithelial cells (cheek cells), so are ________________________ cells, while the smaller cells are ________________________.

(3) 31. Almost all bacteria have rigid walls containing ______________________, a unique complex of polysaccharides and amino acids. The polysaccharides are made up of two sugars called N-acetyl glucoseamine and ______________________. An outer membrane composed of protein, lipoprotein, phospholipids and lipopolysaccharide surrounds a typical Gram-negative bacterium. This has clinical significance because the ______________________ is toxic to mammalian cells and can cause damage to humans even if the bacteria are dead.

(2) 32. The function of the acetone-alcohol mix used during the Gram stain procedure is to quickly ______________________________________________________________. If you had prepared a Gram stain using a Gram-negative culture, had correctly applied the first three reagents, but forgot to apply the last one, what would you expect to see and why? __________________________________________________________

* (2) 33. Which of the reagents shown here could be used as an alternative to the Gram stain for determining wall composition of bacterial cells? __________________________ What would you expect to see if cells with thin walls were mixed with this reagent? ________________

* (2) 34. Describe the shape and Gram stain quality (positive VS negative) of the cells demonstrated. Shape = ________________________ Gram stain characteristic = ________________________
* (2) 35. These cells were stained with a technique known as the _______________________ stain. The cells retaining carbol fuchsin differ from the others present in that their cell walls contain a high percentage of a wax-like lipid called ________________________ acid. This substance makes the cells hard to stain, but very resistant to being decolorized.

* (3) 36. Identify the special (unique) cell structures represented in each of these slides. (Note: - the techniques used to stain these structures were all very different from one another.
   b) _______________________  b) _____________________  c) __________________

* (2) 37. The dormant structures within the cells represented here were stained over steam heat with a stain reagent called ___________________________. What is the shape and location of the dormant structures? ________________________________________

(1) 38. When dealing with microorganisms, the term enrichment refers to what? ________________________
   __________________________________________________________________________
   __________________________________________________________________________

(9) 39. Name the three genera we obtained from soil using enrichment procedures, explain what techniques or special media were used in each case, and explain why these were effective, i.e., what characteristics of the organisms were taken advantage of in order to separate them from other forms present in soil.

Note that for all identifications beyond this point you may gain one extra credit point for each correct species name given, with an overall maximum of five points possible.
40. The organisms shown here are oxygenic photoautotrophs sometimes referred to as blue-green algae. They are classified within the Domain ________________ and within the phylum ________________. Identify the two examples shown as to genus:
   a) __________________ b) __________________

41. Fungi can be divided into three groups or categories based on their morphology, but in this laboratory, we were primarily interested in single-celled forms called _______________ and the fuzzy-looking forms called ________________ that form microscopic hyphae and mycelia that extend only short distances into the air or substrate they are growing in/on.

42. The fungus present on this slide is in the genus ________________. Which type of reproductive process (sexual VS asexual) is indicated by the structures at the pointer tip, and what name is given to these spores? ________________________________

43. The fungus represented here belongs to the genus ________________, and the structures indicated at the pointer tip are called ________________. These are produced via ________________ reproduction.

44. Identify the fungi represented here as to genus and phylum as indicated below:

<table>
<thead>
<tr>
<th>Genus name</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
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<td></td>
</tr>
<tr>
<td>b)</td>
<td></td>
</tr>
<tr>
<td>c)</td>
<td></td>
</tr>
</tbody>
</table>

45. These organisms belong to the phylum ________________. Identify them as to genus:

<table>
<thead>
<tr>
<th>Common name</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a)</td>
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</tr>
<tr>
<td>b)</td>
<td></td>
</tr>
<tr>
<td>c.</td>
<td></td>
</tr>
</tbody>
</table>

46. Identify these unicellular algae as to genus (or group name) and phylum as indicated below:

<table>
<thead>
<tr>
<th>Genus (or group name)</th>
<th>Phylum</th>
</tr>
</thead>
<tbody>
<tr>
<td>a)</td>
<td></td>
</tr>
<tr>
<td>b)</td>
<td></td>
</tr>
</tbody>
</table>

47. The organisms shown here are all classified within the kingdom ________________ and have layers (skeletons or walls) containing ________________ outside their cell membranes. Provide the common names and phyla of these organisms.

<table>
<thead>
<tr>
<th>Common name</th>
<th>Phylum</th>
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</thead>
<tbody>
<tr>
<td>a)</td>
<td></td>
</tr>
<tr>
<td>b)</td>
<td></td>
</tr>
</tbody>
</table>

48. Identify the following as to genus and phylum as indicated below:

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<tr>
<td>b.</td>
<td></td>
</tr>
<tr>
<td>c.</td>
<td></td>
</tr>
</tbody>
</table>

49. The organisms shown here belong to the genus ________________, and to the class ________________. They enter their host with the aid of an infected mosquito.
50. These organisms belong to the phylum __________________________. Identify them as to genus and class.  
   a. ____________________________  
   b. ____________________________  

51. These organisms belong to the phylum __________________________. Identify them as to genus.  
   a.) ___________________________ and b.) ___________________________  

52. These organisms belong to the Kingdom __________________________ and to the phylum __________________________. Identify each of them as to genus.  
   a.) ___________________________  
   b.) ___________________________  
   c.) ___________________________  
   d.) ___________________________  

53. Sauerkraut made by methods similar to those used in this laboratory will typically contain bacteria in the genera __________________________ and __________________________. The sour flavor of this food is due to __________________________, a fermentation product that will typically cause the pH within a saurkraut container to drop from 6.5 to 4.  

54. The primary fermentation product remaining in association with this apple wine is a volatile liquid called __________________________. The __________________________ (second fermentation product) initially formed by the microorganisms present was allowed to escape through the rubber tubing. What organisms were added to the apple juice in order to produce the substances named above? __________________________.  

55. According to the cheese chart supplied within the laboratory syllabus, the bacteria most commonly used in the production of cheese are __________________________.  

56. How do ripened cheeses differ from unripened cheeses? __________________________.  

57. The bacteria most commonly used in the production of yogurt include __________________________ and one or more different species of Lactobacillus. The sour flavor characteristic of yogurt is due to the lactic acid made by these bacteria, but most fresh yogurts also have a subtle nut-like flavor due to the __________________________ present. This second fermentation product has a neutral pH.  

58. Name one type of bacteria recognized as being important food-borne pathogens or responsible for causing intoxication in humans.
1. Define:

Differential medium

Oxidase test

*Thermus aquaticus*

2. Although various methods, e.g., microscopic counts, spectrophotometry, etc. can be used to obtain estimates of cell density in broth media, the quantitative plating method or _______ cell count provides the best method for determining the number of living cells present. Why can only living cells be counted by this method? __________________________________________

3. These bottles were used to make a ____________________, i.e., to dilute a bacterial culture to a known extent, using the procedure indicated. What degree of dilution (total dilution) was achieved in the last bottle? ____________ What degree of dilution (total dilution) would be achieved if you transferred 0.1ml of liquid from bottle #2 onto an agar plate? ________________ Assuming the plate shown was inoculated with 0.1ml of liquid from Bottle #3, how many cfu/ml were present in the original broth culture? ___________

4. These materials were used to complete a procedure called ________________________, a technique recognized for saving both ____________ and media because individual colony types are not transferred one at a time onto separate plates. Why was nutrient agar, a type of non-selective medium, used as the last plate in the sequence inoculated? ________________

5. Mannitol salt agar is a ____________________ medium because it contains 7.5% salt, and will promote the growth of _______________ (salt-loving forms) like *Staphylococcus* while inhibiting the growth of many other bacteria. Which of these plates contains a culture capable of fermenting mannitol? ________ How do you know? ___________
6. These plates contain examples of media supporting the growth of ______________________ bacteria but inhibiting the growth of cells sensitive to chemicals. What is the carbohydrate present in these media? ______________________ Which plate contains a culture capable of fermenting this disaccharide? ______.

7. These tubes contain glucose, and were used to conduct two ____________________ tests (Please write out the complete name of the test). What is the name of the pH indicator present? ______________________ Which tube set was inoculated with an obligately aerobic culture? ______ The bacteria in the other tube set would be described as being ______________________ with respect to their gas requirements, because they can grow with or without oxygen present.

8. One of these tubes was inoculated with organisms capable of fermenting the carbohydrate present. What two types of products were formed, and what evidence (data) is visible to support your conclusion? _____________________________________________________

9. These tubes contain MR-VP medium. The tube set on the left was used to determine if the bacteria present could ferment glucose forming a large percentage of ______________________, enough to overcome the buffers in the medium. The pH indicator added to the broth was ______________________. The tube set on the right was used to complete the Voges Proskauer portion of the test, Barrit’s reagents A and B were added and the tubes were shaken vigorously to determine if or not the bacteria present could produce ______________________, a neutral end product. Which tube combination represents organisms that are positive for both the MR and the VP portion of this test? ____________________

10. Both TSI and SIM media can be used to indicate if bacteria are capable of catabolizing sulfur-containing ______________________ molecules. Bacteria that can, will form ________, a gaseous end product with a distinctly unpleasant odor. When this gas reacts with iron in the media, it forms ______________________ a precipitate readily visible toward the bottom of the culture tube. Which of these tubes show positive reactions? ____________________

11. These tubes indicate if or not the bacteria present can ______________________ the amino acid lysine, forming an alkaline ______________________ called cadaverine, and carbon dioxide. Which of these tube sets contains cadaverine? ______ What is the name of the pH indicator present? ______________________

12. Both lysine and control tubes used in this laboratory are sealed with vaspar, a vaseline-paraffin mix following inoculation. What two functions does vaspar serve? ______________________

13. In this laboratory, students used 3% hydrogen peroxide to determine if or not bacteria could form ______________________ enzymes. Those that could would ________________ when exposed to a drop of H₂O₂ on a clean glass slide.

14. These materials were used to test for the presence of ______________________, an enzyme capable of reacting with Rabbit plasma. Which tube contains bacteria capable of producing this enzyme? __________
15. These media were used to determine if or not the bacteria present could form specific enzymes. Enzymes called ________ allow bacteria to transport citric acid across their cell membranes, while enzymes called ________ allow bacteria to carry out the hydrolysis of urea. Both of these enzymes allow bacteria to form an alkaline end-product called ________ when grown on the media shown. Which of the tubes (as indicated by letter) contain this end product? _________________________

16. Using the data provided, identify the culture indicated (genus and species). Note - the culture present in these tubes should match exactly with one of the names on the chart. The bacteria present are identified as: ____________________________________________

17. The __________________________ or PCR is a method or technique used to amplify segments of DNA in vitro. Unlike DNA replication as it occurs in vivo, the PCR requires only one enzyme, called __________________________, and Okazaki fragments are not formed.

18. The thermal cycler used to amplify DNA during the PCR created a series of temperature fluctuations within a set of tubes containing the appropriate reaction mixture. Explain what occurred relative to the DNA during each temperature period. Note – Single-word answers in this section are incomplete and will be graded accordingly.
   a) 94°C __________________________________________________________________
   b) 55°C __________________________________________________________________
   c) 72°C ___________________________________________

19. Indicate below (by drawing DNA strands on the diagram), where the primers 8-forward and 1530-reverse would anneal to the template DNA shown.

20. The oligonucleotide primers added to the reaction mixture when students were amplifying PUNK2 genes using the PCR were __________________________ (name them). This primer mix was used to determine what genes were amplified, but have multiple additional functions; what are they? __________________________________________

21. Each ddNTP used in a sequencing reaction is tagged with a specific fluorochrome, so will emit a different-colored fluorescent signal when activated by UV light (in this case indicated by the colored ends on the paper strips). Assuming the DNA fragments provided here were generated during a sequencing reaction and were run in a gel within a capillary tube, what nucleotide sequence would be generated? __________________________ Please return the fragments to the beaker when you finish with this station. Thank you!
22. When nucleotide sequencing is conducted using the Sanger Chain Termination method, a small percentage of molecules called ____________________ nucleoside triphosphates (dd-NTPs) are added to each reaction mixture. These nucleotides will terminate DNA replication because they cannot form ____________________________ bonds at their 3’ ends.

23. The segments of DNA amplified during the PCR applied to PUNK2 (and to semester project cultures) were identified as ___________________________ genes. Explain why (giving at least two reasons), this genetic information is considered to be particularly valuable in the identification of bacteria.
   a) ___________________________________________ and
   b) ___________________________________________

24. Define:

   Cloning vector
   Cytolytic bacteriophage
   Agglutination

25. We use the NCBI BLAST to identify unknown types of microorganisms. What do these acronyms stand for? NCBI = ____________________________
   BLAST = ___________________________________________

26. Accessing NCBI and using a nucleotide-nucleotide BLAST to search public databases provided this data. Data obtained from what type of organisms shows significant alignment with the query sequence used? ____________________________ What is the lineage (taxonomic information) listed for this species? ____________________________

27. A new discipline allowing biologists to store, access, compare and analyze vast amounts of biological information using public databases equipped with sophisticated algorithms is called ____________________________ and would not be possible without computer technology. Genomics and Proteomics are two subcategories within this discipline.

28. The ____________________________ identified as pUC19, pGEM and pGLO are small, extrachromosomal loops of DNA (ccc-DNA) that carry genes not usually essential to cell survival. Each of these loops is equipped with its own ____________________________, so will tend to reproduce repeatedly until high copy numbers are reached.
(2) 29. The ccc-DNA identified as pUC19, pGEM and pGLO all carry a marker gene called Bla that encodes __________________________, a type of enzyme allowing cells carrying the gene to be selected for. Since pUC19, pGEM and pGLO can all transcribe and translate the Bla gene, they are considered __________________________ vectors as well as cloning vectors.

(3) 30. A procedure called ______________________________ allows DNA fragments to be separated on the basis of size by "running" them in an electric field. DNA fragments will travel toward the anode because each DNA molecule carries a slight __________________________ charge. Is the simulated agarose gel shown here oriented in the proper direction for conducting this procedure? ______________

(2) 31. What was the purpose of mixing DNA samples with dye (xylene cyanol and brom phenol blue) prior to loading it into the wells of an agarose gel? __________________________

 Which of these pipettes was used to mix DNA with dye, and to load the gel? ______________

(1) 32. In this laboratory, DNA samples are always stained with ethidium bromide after they have been run in agarose gels. What is the function of ethidium bromide? __________________________

(4) 33. Use the data provided to answer the following questions:

 a) Which lane contains bacteriophage lambda DNA? ______________

 b) Which lane contains the plasmid pUC19? ______________

 c) Which lane contains the plasmid pGLO? ______________

 c) Which lane or lanes contain(s) PCR-product DNA? ______________

(2) 34. Enzymes called _____________________________________________ are produced by bacteria, can recognize foreign DNA and chop it up (break phosphodiester bonds), and so restrict genetic exchange between unrelated cells. These enzymes most likely evolved as a defense against __________________________.

(1) 35. According to the naming procedure for restriction enzymes explained in class, the second enzyme isolated from Xenorhabdus meningitis strain N, would be called ______________.

(1) 36. Enzymes capable of cutting double-stranded DNA typically bind to and usually cut within specific recognition sequences. What feature do these recognition sequences have in common? 

 Modification enzymes prevent cells from cutting their own DNA.

(2) 37. The patterns shown here were generated by cutting PCR product DNA from PUNK2 with the enzyme AluI. What does the acronym RFLP stand for? ______________

 The 5th lane in this gel contains a RFLP pattern generated by running fragments of 352, 266, 231, 225, 207, 86, 79 and 33 base pairs. Explain (give two reasons) why the pattern in the gel does not quite match the pattern expected if these fragments were drawn on paper.
38. The recognition sequence 5'…GAATTC…3' is bound and cut by EcoRI between adenine and guanine. 3'…CTTAAG…5'

In the space provided (right),
diagram the cut DNA and then answer the questions below.
This uneven cut will generate short regions of single-stranded DNA called "sticky ends" because they are capable of hydrogen bonding with complementary DNA. What is the technical name for the "sticky ends" generated by this enzyme, and how long will they be?

____________________________________________________________________________

What type of enzyme can be used to "glue" DNA fragments into cloning vectors; i.e., what type of enzyme will form phosphodiester bonds between fragments? _________________________

____________________________________________________________________________

39. A genetic exchange mechanism called DNA mediated _______________________________
can be used to transfer DNA from dead donor cells into living recipients. Although this process occurs rarely under natural conditions, cells grown in the laboratory can be made competent, and will take up DNA much more readily. What did we do to make our E. coli cells competent?

_____________________________________________________________________________

_____________________________________________________________________________

40. All of these plates were inoculated with 50µl of E. coli cells. Use the data provided to answer the following questions:
a) Were the host strains JM-83 and DH5-α carrying Bla genes? __________
b) Did exposing E. coli cells to cold, centrifugation and 42°C for 90 sec. damage them? ______
c) Most of the TSA-AMP plates shown here contain isolated colonies, while the TSA plates contain lawn cultures. Why did far fewer cells grow on the TSA-AMP plates? __________

____________________________________________________________________________

41. The expression of GFP genes in E. coli is regulated by an inducible operon. Green fluorescent protein genes are only expressed when E. coli cells carrying pGLO are grown on TSA-AMP plates containing _______________. This pentose monosaccharide serves as an inducer. By binding with and inactivating the constitutive ____________ protein encoded by the AraC gene, it allows transcription of GFP genes to occur.

42. The clear areas on this plate are called ___________________ and indicate either the presence of free virions or the presence of viral-infected cells. Students in this lab mixed 200µl of E. coli cells with each sample of top agar prior to pouring it over bottom agar; why was this necessary?

____________________________________________________________________________

43. These plates were used to conduct a type of biochemical analysis called ____________________________ in order to identify two strains of E. coli labeled #1 and #2. The E. coli strain Pi can be infected by both Φ fish (large plaques) and Φ meerkat (small plaques), but the E. coli strain Richard Parker can only be infected by Φ meerkat. Which E. coli, #1 or #2 is E. coli strain Pi? ________.
(2) 44. According to the data shown here, what was the total dilution factor (TDF) attained on plate #3? ________________ Assuming these plates represent those made from a serial dilution, what was the concentration of virions in the 1ml KCl broth? ________________

(3) 45. This graph represents data collected during the exercise on Virus Reproduction. The time required for this virus to complete its life cycle is called the burst time or ________________ period, and is ________________ minutes. According to this data the burst size or burst number can be calculated as approximately ________________. Be sure to include units in your answer.

(2) 46. A bright, red-orange colored pigment called ____________________________ is produced by bacteria identified as Serratia marcescens when grown at certain temperatures. Which of these plates was grown at room temperature? ________________

(2) 47. During the exercise addressing the effects of temperature on cell viability, 100µl pond water samples were plated on nutrient agar. One sample was fresh (no treatment), one sample was ______________________ by placing it in boiling water for one minute, and one was boiled for 5 minutes. According to the information collected in our laboratory, did five minutes of boiling serve to sterilize the pond water? ________________

(3) 48. The physical factor used to control bacterial growth on these plates was ________________ radiation. How does this form of radiation kill cells? ________________________________ (Be sure your answer is complete here.) Why would it be inadvisable to use this type of radiation to sterilize plastic Petri plates inside plastic containers? ________________________________

(3) 49. Chemicals used to control potentially pathogenic bacteria outside the body, but on living surfaces are called ____________________________, while those used on non-living surfaces are called ____________________________. According to the data shown here, which general category of bacteria, Gram-positive or Gram-negative are more resistant to potentially toxic chemicals? ____________________________

(2) 50. A patient in a local hospital has been diagnosed with septicemia, and the causative agent has been identified as Escherichia coli. According to the antimicrobial sensitivity test data shown here, if this patient were given Polymyxin B (PB-300), would the pathogen be controlled? ________________ Could Penicillin (P-10) be used as an alternative? ________________

(2) 51. When applied to Antimicrobial Sensitivity Testing, the acronym MIC means ________________ ________________, and this would be found where? ________________________________. Determining the MIC for specific drugs as applied to specific pathogens is highly significant because this can be used to determine the optimum therapeutic dose for controlling pathogens within patients.

(2) 52. Assuming this data was obtained by students measuring zones of inhibition, which of the antimicrobial drugs shown would be considered the most broad-spectrum? ____________________________ Which of the pathogens listed would you least want to be infected by? ____________________________
53. When conducting Standard Methods testing during the Bacteriological Examination of Water, lab personnel are looking for an indicator species identified as _______________________. Explain (give at least two reasons) why these organisms were chosen as indicators rather than more significant pathogens such as *Giardia* or *Vibrio cholerae*.

1) __________________________________________________________________________
2) __________________________________________________________________________

54. These materials represent the first two steps in the Standard Methods procedure for the Bacteriological Examination of Water. The first step is the ____________________________ test, and the second step is the ____________________________ test. Assuming the materials shown here represent actual results obtained from testing water samples, which water sample (A or B) would be safe to drink? _______________

55. In humans, ABO blood groups are determined by the presence of antigenic determinant groups or epitopes called _________________________ that are located on the surfaces of erythrocytes (RBCs). The ability to form these epitopes is genetically determined by alternate forms of a gene or _________________________ designated as A, B, and O, and blood typing typically involves the use of anti-sera (agglutinins) in the isotype __________.

56. Would a person with Rh-negative blood be homozygous for the genes determining Rh? __________ A mother with type A Rh-positive blood and a father with type B Rh-positive blood could have a son with type O Rh-negative blood, only if both parents were ____________________________ for both gene pairs involved.

57. The blood type indicated here would be identified as ______________________ (include both ABO blood groups and Rh). What types of antibodies can a person with this blood type produce? _______________ Could this individual safely receive a transfusion from a person with type AB Rh-negative blood? ____________________________

Think about what will happen to transfused RBCs if they are agglutinated by antibodies.

58. The white bands visible in the agar shown here indicate where antibody-antigen complexes have formed a ____________________________, i.e., an insoluble complex. If the middle well contains antigens from *Coccidioides immitis*, and the outer wells contain serum samples from patients, which of the patients are producing antibodies against the fungus? _______________

59. Identify the leukocytes indicated at the pointer tips:
A) ____________________________ , B) ____________________________
C) ____________________________ , D) ____________________________

Answers to Extra Credit Questions go here:
1. Define:

Serial dilution

Confirmatory test (Bacteriological Examination of Water)

Oligonucleotide primer

2. Although various methods, e.g., microscopic counts, spectrophotometry, etc. can be used to obtain estimates of cell density in broth media, the quantitative plating method or ____________ cell count provides the best method for determining the number of living cells present. Why can only living cells be counted by this method? ________________________________

3. The bottles shown here were used to dilute a batch culture to a known extent, using the procedure indicated. What was the total dilution factor (TDF) achieved in the last bottle? ____________ What concentration would be achieved if you transferred 1.0ml of liquid from bottle #2 onto an agar plate? ____________ Assuming the plate shown was inoculated with 0.1ml of liquid from Bottle #3, how many cfu/ml were present in the original batch culture? ____________

4. These materials were used to conduct a procedure called ______________________________, a method recognized for saving both ______________________________. Given that the master plate used for this procedure contained five sets of colonies growing on nutrient agar, what was the significance of the last plate in the series, also containing nutrient agar? ____________

5. This mannitol salt agar contains 7.5% salt, so will promote the growth of ______________________________ like Staphylococcus, but inhibit the growth of many other bacteria. The pH indicator and sugar present allow the medium to also be ______________________________, i.e., it will cause colonies to look different based on carbohydrate utilization. Indicate which of the plates shown contains a culture that can ferment mannitol, and how you know. ________________________________
6. A culture medium that promotes the growth of some organisms while inhibiting the growth of others is called a ______________________ medium.

7. These plates contain examples of media supporting the growth of ______________________ bacteria but inhibiting the growth of cells sensitive to chemicals. What is the carbohydrate present in these media? ______________________ Which plate contains a culture capable of fermenting this disaccharide? ________.

8. This medium was used to conduct an O/F test; what does the acronym O/F stand for, and what bacterial characteristics can be determined with this medium? ___________________________ _______________________________________________________________________________

What is the pH indicator present? ____________________________. Which tube set contains fermentative organisms? ___________.

9. One of these tubes was inoculated with organisms capable of fermenting the carbohydrate present. What two types of products were formed, and what evidence (data) is visible to support your conclusion? ____________________________________________

10. These tubes contain MR-VP medium. The tube set on the left was used to determine if the bacteria present could ferment glucose forming a large percentage of ______________________, enough to overcome the buffers in the medium. The pH indicator added to the broth was ______________________. The tube set on the right was used to complete the Voges Proskauer portion of the test, Barrit’s reagents A and B were added and the tubes were shaken vigorously to determine if or not the bacteria present could produce ______________________, a neutral end product. Which tube combination represents organisms that are positive for both the MR and the VP portion of this test? ____________________

11. Bacteria capable of catabolizing sulfur-containing __________________________ molecules will form a gaseous end product called ______________________ that has a distinctly unpleasant odor. When this gas reacts with iron in SIM medium or TSI agar, it forms ______________________, a precipitate readily visible toward the bottom of the culture tube. Which of these tubes show positive reactions? ________________

12. All of the tubes shown here contain glucose, and some also contain the amino acid lysine. The purpose of this test is to determine if or not the bacteria present can carry out a type of catabolism called ______________________. Bacteria with this ability will form two types of end products, carbon dioxide (CO₂) and ______________________, called cadaverine. Which of these tube sets contains cadaverine? ________

13. Why does the test described in question #12 above require a control tube, i.e., what specific function does the control serve? ___________________________________________ ___________________________________________

14. The vaspar seal applied to lysine and amino acid control tubes serves what two functions? ___________________________________________
(3) 15. This Simmon’s citrate agar was used to determine if or not the bacteria present could produce an enzyme called ______________________________, associated with citrate transport; while the urea agar was used to determine if or not the bacteria present could form ______________________________, the enzyme responsible for urea hydrolysis. The presence of either enzyme will result in the formation of ______________________________, causing the pH of the medium to increase.

(2) 16. These materials were used to conduct multiple __________________________ tests. Samples of bacteria capable of forming the enzyme ________________________________ produced the dark purple spots when rubbed into the filter paper with clean toothpicks.

(2) 17. These materials were used to test for the presence of ________________________________, an enzyme capable of reacting with Rabbit plasma. Which tube contains bacteria capable of producing this enzyme? ___________

(1) 18. Bacteria are categorized as being __________________________ if they grow on blood agar and cause the hemoglobin present to be converted into methemoglobin, a green-colored substance.

(5) 19. Using the data provided, identify the culture indicated (genus and species). Note - the culture present in these tubes will match exactly with one of the names on the chart. The bacteria present are identified as: ______________________________________________________

(2) 20. The acronym PCR stands for _______________________________________________, a powerful diagnostic tool developed by Kary Mullis and his coworkers at Cetus Corporation in 1986. What is the function of the PCR? __________________________________________

(2) 21. The enzyme most commonly used during the PCR was obtained from hyperthermophilic bacteria identified as _____________________________ (genus and specific epithet please), initially found growing in the hot springs of Yellowstone National Park. Why did Kary Mullis select this type of bacteria as the source for enzymes to be used in the PCR? _________________

(3) 22. The thermal cycler used to amplify DNA during the PCR created a series of temperature fluctuations within a set of tubes containing the appropriate reaction mixture. Explain what occurred relative to the DNA (identify any molecules interacting with DNA) during each temperature period. Note – Single-word answers in this section are incomplete and will be graded accordingly.

a) 94°C __________________________________________________________________

b) 55°C __________________________________________________________________

c) 72°C __________________________________________________________________

(2) 23. Draw two small sections of complementary DNA, to indicate on the diagram below where the primers 8-forward and 1530-reverse would anneal to the template DNA strands shown.

```
5' ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~ 3'

3' ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~ 5'
```
24. What two functions do primers serve in addition to providing free, 3’ ends? ________________

25. The chain termination method of nucleotide sequencing requires that a small percentage of molecules called __________________ nucleoside triphosphates be added to each reaction mixture. These nucleotides will terminate DNA replication because they cannot form __________________ bonds at their 3’ ends. Each one is tagged with a specific fluorochrome, so will emit a different-colored fluorescent signal when activated by UV light (in this case indicated by the colored ends). Assuming the DNA fragments provided here were generated during a sequencing reaction and were run in a gel within a capillary tube, what nucleotide sequence would be generated? _____________________________ Please return the fragments to the beaker when you finish with this station. Thank you!

26. The colored pattern shown here is called a/an ____________________, and is a printed record obtained from an automated DNA sequencing system.

27. This nucleotide sequence was obtained from one of the Physiological Unknown 2 cultures used this semester. What gene product is encoded by this sequence? _____________________________ Why is this genetic information considered to be particularly valuable in the identification of bacteria? ______________________________________________________________________________________

28. Define:

   Expression vector

   Latent period (burst time)

   Alleles

29. Two acronyms (NCBI and BLAST) apply to the genomics exercise. What do these acronyms stand for? NCBI = _____________________________

   BLAST = _____________________________

30. Accessing NCBI and using a nucleotide-nucleotide BLAST to search public databases provided this data. Data obtained from what type of organisms shows significant alignment with the query sequence used? _____________________________ What is the lineage (taxonomic information) listed for this species? _____________________________

31. Small, extra-chromosomal loops of DNA are called ______________________ and can be extracted from cells using relatively simple methods.
(2) 32. Segments of ccc-DNA identified as pUC19, pGEM and pGLO are called cloning ________________, because they are capable of transferring DNA from various sources into host cells. These tiny loops of DNA reproduce independently of the cells chromosomal DNA, because each has its own ______________________ and because they are much smaller. They typically have high copy numbers (around 150 per cell for pUC19), and were used to amplify DNA within cells long before the PCR was developed.

(2) 33. Marker genes carried by pUC19, pGEM and pGLO encode _______________________, an enzyme allowing cells carrying the Bla-gene to grow on media containing Ampicillin. The expression of a marker gene requires transcription, so the Bla gene must be located behind (down stream of) a regions of DNA called the ______________________ site. Translation of the resulting m-RNA requires ribosomes, so can only occur within a living cell.

(3) 34. These materials were used to conduct a procedure called ______________________________, a method allowing DNA fragments to be separated on the basis of size by "running" them in an electric field. DNA fragments will travel toward the anode because each DNA molecule carries a slight ______________________ charge. Is the simulated agarose gel shown here oriented in the proper direction for conducting this procedure? _____________

(2) 35. Dye samples called xylene cyanol and brom phenol blue are mixed with DNA prior to loading it into the wells of agarose gels. What two functions do these dye samples serve? ________________

(4) 36. Use the data provided to answer the following questions:
   a) Which lane contains bacteriophage lambda DNA? ______________________
   b) Which lane contains the plasmid pUC19? _________________________
   c) Which lane contains the plasmid pGLO? _________________________
   c) Which lane or lanes contain(s) PCR-product DNA? ______________________

(2) 37. Bacteria produce enzymes called ______________________, that are capable of recognizing and "chopping up" foreign DNA molecules, i.e., those not protected by modification enzymes. Biologists believe these enzymes most likely evolved as a defense against _________________ with DNA genomes. This seems likely, because such entities can readily take over the function of and destroy cells from within.

(1) 38. Humans use enzymes to cut DNA for a number of reasons, explain one. __________________

(1) 39. According to the naming procedure for restriction enzymes explained in class, the third enzyme isolated from Iodobacter amylolyticus strain Ry-13, would be called ________________.

(1) 40. HindIII is an enzyme capable of recognizing the palindromic sequence 3'-AAGCTT-5' and "cutting" it between the two adenine bases on both strands. 5'-TTCGAA-3' This uneven "cut" will generate short regions of single-stranded DNA called "sticky ends" because they are capable of hydrogen bonding with complementary DNA. What is the technical name for the "sticky ends" generated by this enzyme, and how long will they be?
1. This illustration represents pGEM after being "cut" with EcoRI, and a segment of c-DNA taken from *Aequorea Victoria*, and cut with the same enzyme. The "sticky ends" generated by the enzyme can easily form hydrogen bonds, temporarily binding the two DNA segments together, but the formation of new phosphodiester bonds will require ____________________, an enzyme with glue-like action and the ability to change pGEM into pGLO.

2. What does the acronym RFLP stand for? __________________________. The RFLP patterns shown here were generated by cutting PCR product DNA from PUNK2 with the enzyme AluI. Which sample (as indicated by number) contains a RFLP pattern with fragment sizes 636, 387, 186, 113, 60, 57 and 48bp? __________. Sample #17 contains fragment sizes 310, 286, 209, 206, 189, 79, 44, and 35bp; why does the RFLP pattern generated in the gel not match what you would expect to see if you illustrated these fragments on paper? ____________________________________________________________________________

3. A genetic exchange mechanism allowing DNA fragments to be transferred from dead donor cells to living recipients is called __________________________ and occurs with limited frequency under natural conditions. In this laboratory, *E. coli* cells were made ______________, i.e., more capable of picking up DNA from their environment by growing them to the mid-log phase and treating them with ice cold ______________.

4. All of these plates were inoculated with 50µl of *E. coli* cells. Use the data provided to answer the following questions:
   a) Were the host strains JM-83 and DH5-α carrying Bla genes? __________
   b) Did exposing *E. coli* cells to cold, centrifugation and 42°C for 90 sec. damage them? _____
   c) Most of the TSA-AMP plates shown here contain isolated colonies indicating a significant decrease in population. Why did this occur? ____________________________________________________________________________
   d) Green fluorescent protein is produced only on the plate containing ________________, because this substance is the inducer capable of binding with and inactivating the constitutive ________________ protein encoded by the AraC gene.

5. The clear areas on this plate are called ________________ and indicate regions where cells have been killed by a ________________ called X-174.

6. These plates were used to conduct a type of biochemical analysis called ________________ in order to identify two strains of *E. coli* labeled #1 and #2. The *E. coli* strain Harry can be infected by both Φ Voldemort (large plaques) and Φ Snape (small plaques), but the *E. coli* strain Dumbledore can only be infected by Φ Snape. Which *E. coli*, #1 or #2 is *E. coli* strain Harry? _______.

7. According to the data shown here, what was the total dilution factor (TDF) attained on plate #3? _______________. Assuming these plates represent those made from our serial dilution, what was the concentration of virions in the 1ml KCl broth? _______________
(2) 48. This graph represents data collected during the exercise on Virus Reproduction. According to this data, the burst time or latent period is ________________ minutes and the burst size or burst number can be calculated as approximately ________________. Be sure to include units in your answer.

(2) 49. The bright red-orange colored pigment visible on one of the plates shown here is called __________________ and is produced by *Serratia marcescens* only when grown at certain temperatures. Which of these plates was grown in a lab drawer? ________________

(2) 50. These plates show the effects of temperature on cell viability, when 500µl pond water samples were plated on nutrient agar after being: a) not treated, b) Pasteurized, and c) sanitized. What types of bacteria (in general) survived Pasteurization? ________________. Did the results obtained in our laboratory indicate that boiling the pond water for 5 minutes would render it sterile? __________.

(3) 51. The physical factor used to control bacterial growth on these plates was ______________ radiation. How does this form of radiation kill cells? ________________. Why would it be inadvisable to use this type of radiation to sterilize plastic Petri plates inside plastic or cardboard containers? ________________

(2) 52. Chemicals used to control potentially pathogenic bacteria on living surfaces are called ______________ and are often considerably more toxic than chemicals used inside the body. According to the data collected in our laboratory, which category of bacteria, Gram-positive or Gram-negative are more sensitive to such chemicals? ________________

(2) 53. A patient in a local hospital has been diagnosed with septicemia, and the causative agent has been identified as *Staphylococcus aureus*. According to the antimicrobial sensitivity test data shown here, if this patient were given Tetracycline (TE-30), would the pathogen be controlled? __________. Could Rifampin (RA-5) be used as an alternative? __________

(3) 54. When applied to Antimicrobial Sensitivity Testing, the acronym MIC means ______________, and would be found where? ________________. Determining the MIC for specific drugs as applied to specific pathogens is highly significant because this can be used to determine the optimum ______________ dose for controlling pathogens within patients.

(1) 55. Assuming this data was obtained by students measuring zones of inhibition, which of the drugs shown would be considered the most narrow-spectrum antibiotic? ________________

(3) 56. Standard testing methods applied in the Bacteriological Examination of Water are designed to detect ______________. These organisms live in the gut, so are good indicators of fecal contamination. Two additional reasons we test for these specific organisms are: ________________
(3) 57. These tubes were used to conduct the first step in the Standard Methods procedure for the Bacteriological Examination of Water, or the ______________________________ test. Which tube contains water testing positive for fecal contamination, and what evidence is visible? ____

(2) 58. In humans, ABO blood groups are genetically determined by alternate forms of genes (homologous DNA) designated as A, B, and O. These encode antigenic determinant groups or epitopes called ______________________________ that appear on the surfaces of RBCs, and are capable of binding with specific ______________________________ in the isotype IgM called agglutinins.

(2) 59. A person with type O Rh-negative blood would be ______________________ recessive for both the O and d genes as explained in class, i.e., their genotype would be OOdd. If a mother with type A Rh-positive blood and a father with type B Rh-positive blood produced a daughter with type O Rh-negative blood, both parents would have to be ______________________ for both gene pairs involved.

(3) 60. The blood type indicated here would be identified as _______________________ (include both ABO blood groups and Rh). What types of antibodies can a person with this blood type produce? ______________________. Could this individual safely receive a transfusion from a person with type AB Rh-negative blood? ______________________

Think about what will happen to transfused RBCs if they are agglutinated by antibodies.

(3) 61. This photograph represents data collected with a type of serological test called an ______________________________ test, involving the diffusion of antibodies and antigens through agarose. The white bands visible in the agar indicate where antibody-antigen complexes have formed a ______________________, i.e., an insoluble complex. If the middle well contains antigens from Coccidioides immitis, and the outer wells contain serum samples from patients, which of the patients are producing antibodies against the fungus? __________

(4) 62. Identify the leukocytes indicated at the pointer tips:
   A) ______________________________, B) ______________________________
   C) ______________________________, D) ______________________________

Answers to Extra Credit Questions go here:
SUGGESTED STUDY TECHNIQUES

The conscientious use of proven study methods can help students to improve their mastery of subject materials and to improve their grades. In a physiological sense, although LEARNING involves understanding, it is mostly a matter of REPETITION: The more exposure students have to their subject materials, the greater their chances of retaining that information. The more times a neuronal circuit (pathway) is excited (used) in the central nervous system (brain and spinal cord) the more that circuit is facilitated (remembers).

When students begin to study Microbiology they must recognize that: 1) most of the information presented will be "new" to them, and 2) much of the vocabulary used to present this information will be unfamiliar. Studying a new course "in a different language" is more difficult than taking a "more conventional class". It is somewhat similar to studying the history of Czarist Russia with the instructor lecturing on the history in Russian. You would obviously need to master the language before you can master the history. Therefore, when studying Microbiology (as well as many other academic subjects) you must learn the language in order to master the information. The following techniques can be useful in any class where there is a large volume of information being presented and much of the vocabulary is new.

1. Establish a schedule (similar to your class schedule) and extend it for a full 24 hour day, 7 days a week. Know when you have free time to study and use it. Don't waste time sitting in the campus center, lounging on the lawn or watching mediocre TV. When you need to play, do so energetically, and when its time to study, do so intensely. Don't try to play while studying.

2. Understand your physiology. Recognize when you are really alert and when you are sluggish mentally (like after meals). STUDY ACCORDINGLY!

3. Complete all reading assignments before the lecture and/or lab. on that topic. You will find this makes the lecture and lab. material easier to understand.

4. Make sure your notes are complete and accurately represent the information being presented. (Most students write down less than half of the material they receive orally, and this is often not adequate.) If necessary, share notes with another student and/or invest in a small tape recorder and record lecture and lab. presentations. Taped information can be reviewed before the next class session, and will allow you to fill in any gaps present in your notes. Reviewing taped lectures increases accuracy, and allows for REPETITION of the subject material.

5. Use flash cards for vocabulary. REPETITION IS LEARNING, whether it is in college or in the second grade. Write each new term on one side of a 3X5 card along with the word define. On the other side of the card write out a complete definition for the term in the form of a question, leaving a blank space for the term being studied. This method will allow you to study for both definition and fill in the blank type questions. You will find that flash cards take time to make up, but are highly effective if used properly. For best results, do not attempt to put too much information (more than two new terms) on a single card, and remember that there is a difference between recognition and recall. When studying for “fill-in-the-blank” type tests, do not be fooled by recognition. If you must turn the 3X5 card over and look at the information before you can “recall” it, you DO NOT know it, and will not be able to recall it on a test.
6. Review is repetition; REPETITION IS LEARNING. Review each night before the next day's class. Pick up points on quizzes. KNOW WHAT TO STUDY, and if the instructor tests from lecture materials, don't waste time excessively studying other sources of information. Review several weeks ahead for finals. Get ahead of the crowd. Try simply reading and re-reading your notes from beginning to end at least three or four times a week (It will take less time each time you read them, and is more REPETITION.) If you have reviewed ahead of time you can come to the instructor and get questions answered while others are feebly cramming.

7. Rewrite your lecture notes. It is time consuming but it is repetition. REPETITION IS LEARNING. Look up unclear portions. Write a second set of notes which contains only that information you could not immediately remember from your original notes.

8. Set small, attainable study goals. If you are successful in attaining your initial goals, you will be encouraged to formulate and accomplish additional goals. Success feels good!

9. Review some school work each evening before going to sleep. This is usually a very efficient learning technique as there is little or no interference with the formation of memory. This method is particularly valuable when trying to comprehend difficult subjects.

These techniques, when used properly, yield a high degree of success. If you force yourself to use them (over and over again), establish a regular pattern and follow it, you will be successful. "Hit and miss" study habits result in "hit and miss" learning and yield "hit and miss" grades. Consider your role as a student analogous to a sharp-shooter; with practice you can be one of the best, but without it you're just average.

USE THIS STUDY GUIDE!