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.

Table of Contents

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 _____.

(2) 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.

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

(1) 11. What was the significance of Koch's postulates at the time they were developed?

(1) 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?

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

(1) 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.

(2) 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.

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

(1) 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.

(2) 18. _____ was a Scottish bacteriologist and medical practicioner recognized for his discovery of _____, one of the first antibiotics obtained from fungi in the genus *Penicillium*.

(4) 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 _____.

(2) 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.

(3) 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 _____.

(1) 11. Carbohydrates such as arabinose, ribose and deoxyribose contain five carbon atoms and are referred to as _____.

(2) 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.

(2) 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.

(1) 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.

(1) 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.

(2) 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.

(2) 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 _____.

(1) 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 <u>other</u> 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.
	I. Lactose, sucrose and maltose
____ Quaternary structure	J. Negatively charged particle of matter.
	K. Glycogen, cellulose, chitin and starch.
____ Triglycerides	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 _____.

(1) 8. Cells placed into a hypotonic environment would tend to _____ water via a passive transport process called osmosis.

(1) 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.

(1) 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 _____.

(1) 11. Explain briefly how facilitated diffusion differs from active transport.

(2) 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".

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

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

(1) 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.

(2) 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? _____

(2) 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 _____.

(3) 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 _____.

(1) 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.

(3) 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 _____.

(2) 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.

(1) 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.

(3) 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.

(2) 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.

(2) 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? _____

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

(2) 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.

____ Histones ____	A. Contain enzymes involved in hydrogen peroxide metabolism.
____ Ribosomes ____	B. Made of protein pilin, allow for attachment and genetic exchange.
____ Cell walls ____	C. Homogeneous proteins associated with linear strands of chromatin.
____ Glycocalyx ____	D. Reservoir of stored food which aids in attachment and pathogenicity.
____ Thylakoids ____	E. Extrachromosomal loops of DNA carrying genes not essential to cell function.
____ Peroxisomes ____	F. Composed of RNA and protein, are the site of all protein synthesis.
____ Nucleoid ____	G. A non-living extracellular layer which provides support and protection.
____ Golgi complex ____	H. Primary site of packaging, secretion, and polysaccharide synthesis.
____ Pili ____	I. Not bounded by a membrane, contains one circular chromosome.
____ Plasmids ____	J. Membranous structures with enzymes for ATP synthesis via photophosphorylation.

(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.

(3) 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 _____.

(2) 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? _____ _____

(2) 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.

(1) 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.

(2) 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.

(1) 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.

(2) 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?

(2) 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.

(2) 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. Exospores 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? _____
_____

(2) 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.

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

(2) 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 _____.

(1) 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 then they are in the identification of bacteria.

(2) 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.

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

(2) 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 _____.

(1) 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.

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

(2) 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.

(1) 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.

(2) 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.

(2) 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 _____.

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

(1) 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.

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

(2) 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 _____.

(1) 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.

(2) 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.

(2) 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 _____.

(1) 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 _____.

(2) 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.

(1) 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.

(1) 26. What is the human genome project?

Introduction to Archaea and Bacteria:

(5) 1. Define:

Archaea

Bacteriarhodopsin

Luciferase

Leghemoglobin

Anoxygenic

(2) 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) 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.

(2) 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.

(1) 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.

(2) 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.

(1) 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 improtant human diseases, but can also cause severe inflammation in humans infected by roundworms.

(3) 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.

(2) 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.

(2) 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. _____ _____

(2) 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 _____.

(1) 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).

(1) 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.

(1) 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 0.25μm in diameter), they often pass through filters designed to remove bacteria.

(1) 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.

(2) 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 organsims is a primary flavor component in such foods.

(1) 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 _____.

(1) 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.

(2) 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.

(1) 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.

(1) 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.

(5) 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.

____ *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 (Myceteae) 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.

(2) 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.

(1) 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 _____.

(2) 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 _____.

(2) 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 _____.

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

(2) 14. Fungi are used industrially in the production of food, antibiotics, _____, and a variety of _____.

(2) 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.

(3) 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 _____.

(2) 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) _____

(1) 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 _____ _____.

(2) 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.

(2) 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.

(2) 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.

(3) 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.

(4) 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.
____ *Balandidium*	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. Sporozoeans 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.

(1) 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 hosts stomach.

(2) 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.

(2) 14. Two types of human diseases that can be caused by amoeba-like protozoa are _____ and _____.

(2) 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.

(2) 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.

(5) 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.

(1) 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 _____.

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

(2) 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 abcess and damage to other interanl organs can occur.

Introduction to Multicellular Parasites:

(5) 1. Define:

Helminth

Definitive host

Redia

Vector

Monoecious

(2) 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) 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.

(2) 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.

(2) 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? _____

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

(3) 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 _____.

(1) 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 cycts.
____ *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 _____ (cocci or bacilli). What arrangement would cocci achieve if their fission plane was initially horizontal, and then vertical? _____

(1) 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: _____
_____

(2) 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?
_____

(2) 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.

(2) 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.

(2) 13. Where in a bacterial colony would you expect to find the "youngest" cells and why? _____
_____

(2) 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?
_____

(1) 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)

(1) 8. A molecule is said to have been _____ if it has gained oxygen or has given up one or more electrons and hydrogen protons.

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

(4) 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.

(2) 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 _____ _____.

(2) 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.

(2) 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.

(1) 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.

(1) 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.

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

(2) 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 _____.

(1) 18. _____ is a coenzyme derived from the B-complex vitamin niacin.

(3) 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.

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

(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? _____
_____

(2) 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 _____.

(1) 10. Homofermentative organisms such as *Lactococcus lactis* produce _____ as the only product of their fermentation activities.

(3) 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).

(2) 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 _____.

(2) 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 _____.

(2) 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.

(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? _____ _____

(1) 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).

(3) 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.

(2) 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 $FADH_2$).

(1) 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.

(1) 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.

(2) 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? _____

(3) 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 _____.

(2) 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.

(3) 24. $NADH+H^+$ and $FADH_2$ 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 $FADH_2$ reoxidized yields enough energy to make _____ (#) ATP.

(2) 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.

(3) 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. _____.

(3) 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 _____ (#) 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 _____ and _____.

(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: $6CO_2 + 6H_2O$ + energy from light will yield $C_6H_{12}O_6 + 6O_2$. 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 a _____ 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? _____

(2) 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.

(2) 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.

(2) 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 _____.

(3) 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.

(2) 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 _____.

(3) 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.) _____

(2) 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.

(2) 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.

(2) 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.

(2) 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.

(2) 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.

(2) 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 _____.

(2) 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.

(2) 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 _____.

(2) 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 _____.

(1) 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.

(2) 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.

(2) 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.

(3) 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.

(1) 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?
_____.

(4) 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
TCTACAGTTTGGGCATACCTTACCAAC

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.

(1) 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).

(3) 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.

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

(2) 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.

(2) 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):

(5) 1. Define:

Operon

Promoter

Operator

Catabolite repression

Cyclic-AMP

(1) 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.

(2) 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? _____

(1) 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.

(2) 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?
_____

(2) 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.

(1) 7. The promoter site (P-site) of an operon has what function? _____
_____

(2) 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.

(2) 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.

(2) 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 _____.

(3) 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.

(3) 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.

(2) 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.

(2) 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? _____

(1) 15. In the inducible system controlling lactose utilization, the repressor protein is active under what circumstances? _____

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

(2) 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.

(1) 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.

(2) 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.

(1) 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.

(2) 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).

(2) 7. The total DNA content of the chromosomes an organism may be referred to as the _____ 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.

(3) 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? _____ _____

(1) 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.

(2) 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.

(2) 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 it's m-concentration in 24 hours, how many mutations would be expected to have occurred within the population during that time period? _____

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

(2) 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".

(3) 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.

(1) 15. A _____ is a chemical or physical factor that increases the rate at which mutations occur.

(1) 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.

(2) 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.

(3) 18. Given a DNA strand with the base sequence TACCTGGCACGTTGGGACCTAC, 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.

(2) 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 _____.

(2) 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.

(2) 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 _____.

(2) 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 _____ or "jumping genes". 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".

(2) 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? _____

(3) 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 _____.

(2) 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.

(2) 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 _____.

(2) 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 _____.

(1) 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.

(1) 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.

(3) 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 _____.

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

(1) 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).

(1) 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.

(1) 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.

(2) 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. _____

(5) 20. 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.

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

Introduction to Prions, Viroids and Viruses:

(5) 1. Define:

Prion

Prophage

Lysogenic conversion

Retrovirus

Reverse transcriptase

(1) 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.

(2) 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 _____.

(2) 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.

(3) 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.

(2) 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. _____ _____

(2) 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.

(2) 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.

(2) 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).

(2) 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 _____.

(3) 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) _____

(2) 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? _____ _____

(2) 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 _____.

(1) 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.

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

(3) 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.

(3) 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? _____ _____

(1) 18. The adsorption process of HIV involves binding of spike proteins on the _____ _____ with specific receptors on the host cell surface.

(1) 19. HIV is a retrovirus which must _____ it's genetic information from RNA into DNA before it can become incorporated into the hosts chromosome.

(1) 20. HIV uses an enzyme called _____ to incorporate the DNA version of its viral genome into the chromosome of its host.

(3) 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.

(1) 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.

(1) 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 Ortho-myxoviruses 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.

(2) 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.

(2) 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.

(2) 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) _____

(1) 12. A/an _____ is a chemical agent used to control pathogenic microorganisms on non-living surfaces.

(2) 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.

(5) 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	I. Chemical agent used to control pathogens on living surfaces (skin).
____ Halogens	J. Heat treatment that will kill vegetative cells made by bacteria, but not endospores.
____ Sterilization	K. Mercury, lead, zinc, copper and silver.

(2) 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.

(2) 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 _____.

(2) 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.

(3) 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) _____

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

(3) 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? _____ _____

(1) 21. The sulfa drugs are synthetic antimicrobial agents that inhibit cell growth by _____ _____.

(2) 22. The _____ drugs are synthetic antimicrobial agents which exert their influence by blocking the enzymatic pathway converting _____ _____ within prokaryotic cells.

(2) 23. Antimicrobial drugs known as _____ kill actively growing cells by inhibiting the formation of peptidoglycan. Tetracycline and aminoglycosides exert their antimicrobial effects by _____.

(2) 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 _____.

(2) 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 _____.

(2) 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.

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

(2) 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.

(2) 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.

(2) 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? _____ _____

(1) 15. Chemical substances called _____ tend to elevate the temperature within a tissue or within the body overall.

(1) 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.

(1) 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.

(1) 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.

(2) 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 _____.

(2) 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).

(2) 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.

(2) 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 _____.

(1) 23. Describe the "defensive" role played by the normal flora associated with the human body. ____ _____

Specific Defense and Immunology:

(5) 1. Define:

Adaptive or Acquired immunity

Immunoglobulin

Epitope

Helper-T Lymphocyte

Interleukin

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

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

(1) 4. Adaptive or acquired immunity may be gained passively through the placenta or breast milk or by receiving _____.

(1) 5. 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.

(3) 6. 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.

(2) 7. 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.

(2) 8. 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.

(3) 9. 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).

(2) 10. 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. _____ _____

(2) 11. 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.

(2) 12. 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.

(1) 13. 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.

(2) 14. 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.

(2) 8. Hypersensitivity reactions that occur within minutes of exposure to the allergen are categorized as _____ reactions and involve _____.

(2) 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 compliment proteins is referred to as a _____ response.

(2) 10. _____ is a type I hypersensitivity reaction involving immunoglobulins in the class _____ and the release of histamine body wide.

(2) 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.

(1) 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.

(1) 13. A type II hypersensitivity reaction known as a cytotoxic response involves IgG and plasma proteins known as _____ factors.

(2) 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).

(2) 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 _____.

(2) 16. A type III hypersensitivity reaction that is localized is called a/an _____ reaction, while one that occurs body wide is referred to as _____.

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

(2) 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 _____.

(1) 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?
_____

(1) 20. Although immunizations can sometimes cause hypersensitivity reactions, the most common causes of allergic reactions are _____.

(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.

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

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 _____.

(2) 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.

(2) 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).

(2) 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).

(2) 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.

(5) 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.

(1) 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 _____.

(2) 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 _____.

(1) 11. _____ play a dual role in disease transmission because they serve both as reservoirs and as vectors.

(2) 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.

(1) 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?

(1) 14. _____ transmission is that which involves contact between a susceptible host and a living reservoir.

(1) 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 _____.

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

(3) 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 _____.

(2) 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.

(2) 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.

(1) 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 _____.

(1) 10. _____ may be defined as a degree of pathogenicity and may be expressed in terms of ID_{50} or LD_{50}.

(2) 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 _____.

(2) 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.

(2) 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) _____

(2) 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.

(2) 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) _____

(2) 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:
_____

(3) 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.

(3) 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.

(1) 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.

(1) 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.

(1) 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.

(1) 22. Gram negative, areobic 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.

(2) 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 _____.

(2) 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) _____

(3) 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.

(2) 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).

____ Dysentery	A. *Bacillus*	a. *pyogenes*
____ Gas gangrene	B. *Francisella*	b. *pertussis*
____ Diphtheria	C. *Mycobacterium*	c. *anthracis*
____ Pneumonia	D. *Corynebacterium*	d. *tetani*
____ Typhoid	E. *Chlamydia*	e. *tularensis*
____ Scrub typhus	F. *Shigella*	f. *perfringens*
____ Leprosy	G. *Yersinia*	g. *menengitidis*
____ Cholera	H. *Bordetella*	h. *gonorrheae*
____ Gonorrhea	I. *Streptococcus*	i. *tuberculosis*
____ Tularemia	J. *Borrelia*	j. *pallidum*
____ Tetanus	K. *Vibrio*	k. *pneumoniae*
____ Tuberculosis	L. *Rickettsia*	l. *pneumophila*
____ Syphilis	M. *Pasteurella*	m. *tsutsugamushi*
____ Meningitis	N. *Treponema*	n. *typhi*
____ Lyme disease	O. *Neisseria*	o. *burgdorferi*
____ Plague	P. *Legionella*	p. *diphtheriae*
____ Legionnaire's disease	Q. *Orientia*	q. *cholerae*
____ Anthrax	R. *Staphylococcus*	r. *pestis*
____ Whooping cough	S. *Clostridium*	s. *leprae*
____ Scarlet fever	T. *Salmonella*	t. *dysenteriae*

Sierra College Name: _____
Microbiology Laboratory Exam 1 October 11, 2012

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 _____.

* (2) 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.

(2) 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. _____

(3) 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? _____

(2) 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.

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

(2) 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? _____
_____

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

(2) 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? _____
_____

(2) 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. 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.

* (1) 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.) _____

* (3) 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.

(2) 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 peptidogylcan. Cyanobacteria and other types of autotrophs can obtain the _____ they need from air.

(3) 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 _____

(2) 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? _____

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

(2) 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? _____

* (2) 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? _____

* (3) 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 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? _____
_____

(2) 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.

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

(3) 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.

(2) 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? _____
_____

(1) 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?
_____

* (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.
a) _____ 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? _____
_____.

(9) 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.

* (4) 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) _____

(2) 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:

	Genus name	Phylum
a)	_____	_____
b)	_____	_____
c)	_____	_____

* (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:

	Genus (or group name)	Phylum
a)	_____	_____
b)	_____	_____

* (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.

	Common name	Phylum
a)	_____	_____
b)	_____	_____

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

	Genus	Phylum
a.	_____	_____
b.	_____	_____
c.	_____	_____

* (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.

	Genus	Class
a.	_____	_____
b.	_____	_____

* (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^{o} 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. _____
_____

Sierra College Name: _____
Microbiology Laboratory Exam 1 April 4, 2013

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:

Calibration

Fungi

Fermented food

(1) 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.

(1) 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.

(2) 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 _____.

(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. 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? _____
_____

* (2) 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.

(2) 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? _____
_____

(1) 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.

(3) 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? _____

(2) 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.

(1) 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? _____

(2) 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? _____

* (3) 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. _____
_____

* (2) 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.) _____

(3) 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? _____

* (3) 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.) _____

(2) 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.

(4) 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 _____

(1) 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? _____
_____

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

(2) 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? _____

* (2) 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? _____

* (3) 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.

* (4) 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) _____

(2) 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.

* (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:

	Genus name	Phylum
a)	_____	_____
b)	_____	_____
c)	_____	_____

* (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:

	Genus (or group name)	Phylum
a)	_____	_____
b)	_____	_____

* (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.

	Common name	Phylum
a)	_____	_____
b)	_____	_____

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

	Genus	Phylum
a.	_____	_____
b.	_____	_____
c.	_____	_____

* (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. Genus Class
a. _____ _____
b. _____ _____

* (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. 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.

* (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. According to the cheese chart supplied within the laboratory syllabus, the bacteria most commonly used in the production of cheese are _____.

(1) 56. How do ripened cheeses differ from unripened cheeses? _____
_____

(2) 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.

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

Sierra College Name: _____
Microbiology Laboratory Exam II November 29, 2012

Directions: Read each question carefully and completely before you write your answer. Note that some of the questions require you to observe materials on the desktops. Those questions are marked with an asterisk in the margin. Please make your answers as complete as possible, DO NOT BE VAGUE!

(3) 1. Define:

Differential medium

Oxidase test

Thermus aquiticus

(2) 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? _____
_____

(4) 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? _____

(3) 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?_____
_____

(4) 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? _____
_____

(3) 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? _____.

(4) 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.

(2) 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? _____

(4) 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? _____

(4) 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? _____

(4) 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? _____

(2) 12. Both lysine and control tubes used in this laboratory are sealed with vaspar, a vasoline-parafin mix following inoculation. What two functions does vaspar serve? _____ _____

(2) 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 H_2O_2 on a clean glass slide.

(2) 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? _____

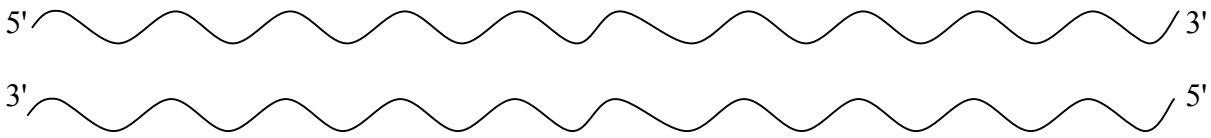
(4) 15. These media were used to determine if or not the bacteria present could form specific enzymes. Enzymes called _____ allow bacteria to transport citric acid across their cell membranes, while enzymes called _____ allow bacteria to carry out the hydrolysis of urea. Both of these enzymes allow bacteria 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? _____

(4) 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: _____

(2) 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.

(3) 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^{o}C _____
b) 55^{o}C _____
c) 72^{o}C _____

(2) 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.



(2) 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? _____
_____

(1) 21. Each ddNTP used in a sequencing reaction is tagged with a specific flurochrome, 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!

(2) 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.

(3) 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) _____

(3) 24. Define:

Cloning vector

Cytolytic bacteriophage

Agglutination

(2) 25. We use the NCBI BLAST to identify unknown types of microorganisms. What do these acronyms stand for? NCBI = _____
BLAST =_____

(2) 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? _____
_____

(1) 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.

(2) 28. The _____ identified as pUC19, pGEM and pGLO are small, extra-chromosomal loops of DNA (ccc-DNA) that carry genes not usually essential to cell survival. Each of these loops is equipped with it 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 gels? _____

(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 meningesii* 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.

(3) 38. The recognition sequence 5'…GAATTC…3' is bound and cut by *Eco*RI 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? _____

_____

(2) 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?

_____

_____

(3) 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^{o} 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? _____

_____

(2) 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.

(2) 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?

_____

(2) 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 anti-microbial drugs shown would be considered the most broad-spectrum? _____ Which of the pathogens listed would you least want to be infected by? _____

(3) 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) _____

(3) 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? _____

(3) 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 _____.

(2) 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.

(3) 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.

(3) 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? _____

(4) 59. Identify the **leukocytes** indicated at the pointer tips:
A) _____, B) _____
C) _____, D) _____

Answers to Extra Credit Questions go here:

Sierra College Name: _____
Microbiology Laboratory Exam II May 16, 2013

Directions: Read each question carefully and completely before you write your answer. Note that some of the questions require you to observe materials on the desktops. Those questions are marked with an asterisk in the margin. Please make your answers as complete as possible, DO NOT BE VAGUE!

(3) 1. Define:

Serial dilution

Confirmatory test (Bacteriological Examination of Water)

Oligonucleotide primer

(2) 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) 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? _____

(3) 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? _____
_____

(4) 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. _____
_____

(1) 6. A culture medium that promotes the growth of some organisms while inhibiting the growth of others is called a _____ medium.

(3) 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? _____.

(3) 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? _____

(2) 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? _____

(4) 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? _____

(4) 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? _____

(3) 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 (CO_2) and _____ called cadaverine. Which of these tube sets contains cadaverine? _____

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

(2) 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 of 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.

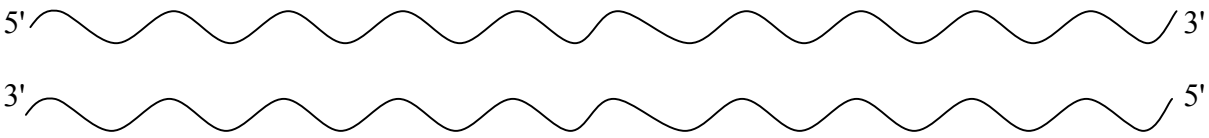
(5) 19. Using the data provided, identify the culture indicated (genus and species). Note - the culture present in these tubes will match exactly with one of the names on the chart. The bacteria present are identified as: _____

(2) 20. The acronym PCR stands for _____, a powerful diagnostic tool developed by Kary Mullis and his coworkers at Cetus Coorporation 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 <u>incomplete</u> and will be graded accordingly.
a) 94^{o}C _____
b) 55^{o}C _____
c) 72^{o}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.



(2) 24. What two functions do primers serve in addition to providing free, 3' ends? _____
_____

(3) 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 flurochrome, 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!

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

(2) 27. This nucleotide sequences 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? _____
_____

(3) 28. Define:

Expression vector

Latent period (burst time)

Alleles

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

(2) 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? _____
_____

(1) 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. *Hin*dIII 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) 41. 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.

(3) 42. 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, 128, 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) 43. 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 _____.

(5) 44. 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^{o} 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.

(2) 45. The clear areas on this plate are called _____ and indicate regions where cells have been killed by a _____ called X-174.

(2) 46. 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? _____.

(2) 47. 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? _____ _____ (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 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 **leukocyts** 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 !