FOOD MICROORGANISMS AND SAFETY

Unit. Chemistry of Food

Problem Area. Basic Principles of Food Science

Student Learning Objectives. Instruction in this lesson should result in a student achieving the following objectives:

1. **Explain the classification and naming of microorganisms.**
2. **Describe the structure of bacteria, yeasts and molds, and viruses.**
3. **Identify conditions for microbial growth and methods of assessing organisms present in food.**
4. **Describe the role of temperature, water activity, pH, oxygen, redox, and antimicrobials on microbes.**
5. **Explain food spoilage and how it impacts shelf life.**
6. **Describe how food is contaminated leading to food poisoning and food-borne infections.**

List of Resources. The following resources may be useful in teaching this lesson:

E-unit(s) corresponding to this lesson plan. CAERT, Inc. http://www.mycaert.com.

Glosson, Linda R., Nanci Burkhart, Anna Sue Couch, Brenda Barrinton Mendiola, Connie R. Sasse, and Lynn Steil. *Food for Today*, 4th edition. Peoria, Illinois: Glencoe/McGraw-Hill, 2000.

Medved, Eva. *The World of Food*. Englewood Cliffs, New Jersey: Prentice-Hall, Inc., 1990.

Nicklin, J., K. Graeme-Cook, T. Paget, and R. Killington. *Microbiology*. New York: Springer-Verlag, Inc., 1999.

Parker, Rick. *Introduction to Food Science*. Albany, New York: Delmar, 2003.

Potter, Norman N. and Joseph H. Hotchkiss. *Food Science*, 5th ed. New York: Springer, 1998.

Seperich, George J. *Food Science and Safety*, 2nd ed. Upper Saddle River, NJ: Pearson Prentice Hall Interstate, 2004.

Walker, Pam and Elaine Wood. *Biology in Our Lives*. Danville, Illinois: Interstate Publishers, Inc., 2001.

Wilbraham, Antony C., Dennis D. Staley, Michael S. Matta, and Edward L. Waterman. *Chemistry*, 7th ed. Boston, MA: Pearson Prentice Hall, 2008.

Wagner, Lynn A. and George J. Seperich. *Food Science and Technology*. Upper Saddle River, New Jersey: Pearson Prentice Hall, 2004.

____. *Concise Encyclopedia of Science and Technology*, fifth edition. New York: McGraw-Hill, Inc., 2005.

List of Equipment, Tools, Supplies, and Facilities

- ✓ Writing surface
- ✓ Overhead projector and screen
- ✓ Transparencies from attached masters
- ✓ Copies of student lab sheet (will include list of supplies and materials for the activity)

Terms. The following terms are presented in this lesson (shown in bold italics where defined):

- Agar
- Agar plate
- Antimicrobial
- Bacteria
- Binomial name
- Capsid
- Cell culture
- Contamination
- Culture medium
- Eukaryote
- Filamentous
- Food-borne infection
- Food microbiology

- Food poisoning
- Food safety
- Gram staining
- Habitat
- Inoculation
- Molds
- Morphology
- Nutrient agar
- Pasteurization
- Prokaryotic cell
- Protozoa
- Shelf life
- Spoilage
- Sterilization
- Stool culture
- Strain
- Substrate
- Taxonomy
- Virion
- Virus
- Yeasts

Interest Approach. Use an interest approach that will prepare the students for this lesson on food microorganisms and safety. Teachers often develop approaches for their unique class and student situations. Two possible approaches are included here.

Approach One: Ask one or more students to explain what is meant by "things you can't see can hurt you." After brief comments, have students apply this statement to the food that they eat. Follow by asking, "what could be in food that isn't readily seen that can cause problems?" Following comments, move into the next interest approach, cover the objectives for the lesson, and begin lesson content.

Approach Two: Ask one or more students to explain the meaning of microbiology. List possible definitions on the writing surface. Ask students to draw a consensus definition of microbiology from the discussion. Summarize with "Microbiology is the study of organisms that are visible only with a microscope." Discuss as needed and move into the lesson. Present the objectives for the lesson and begin covering the objectives sequentially.

SUMMARY OF CONTENT AND TEACHING STRATEGIES

Objective 1: Explain the classification and naming of microorganisms.

Anticipated Problem: How are microorganisms classified and named?

I. All known living things have been investigated by scientists and classified in some way. The classification of living things, including microorganisms, is known as ***taxonomy***. In some cases, taxonomy is referred to as systematics though systematics may have a somewhat broader meaning. Scientific taxonomy involves grouping together organisms that are alike in certain characteristics, including evolutionary relationships. Taxonomy helps communicate organism similarities and differences as well as provides for a system of naming. (Note: Taxonomy remains in a state of flux. As scientists learn new information about organisms, classifications may change.)

A. The modern classification system involves grouping organisms into hierarchical categories. This is sometimes known as biological classification. Sorting the organisms into groups is a major role of biological classification. Scientists use information gained from developmental patterns, biochemistry, molecular biology, genetics, and detailed morphology. DNA study is increasingly used in the classification of organisms. (Note: The system described here is widely used in the United States though adjustments are continually being proposed.)

B. Scientists use a system that consists of seven divisions or stages of similarities and differences. The seven stages are: kingdom, phylum or division, class, order, family, genus, and species. These stages are arranged from the broadest to the most specific. Microorganisms in a kingdom have shared broad characteristics while those in a species are very similar on many characteristics. Beyond species, some organism categories are further broken into strains or varieties. A ***strain*** is a subspecies within a species that have unique characteristics from others in the species. Three kingdoms have numerous species of microorganisms that serve many roles and may pose problems with food quality: monera, protista, and fungi. In addition, the viruses are not classified into a kingdom. The kingdom Animalia and kingdom Plantae are comprised of organisms made up of many cells.

1. The kingdom Monera is comprised of simple, one-celled microorganisms. It has four phyla. The phylum Proteobacteria contains the ***bacteria***, which are of most concern with food quality. With incomplete cells, they are known as prokaryotic organisms. Many strains of bacteria are found. The strains are distinguished by cell shape, the structures the cells form, DNA and RNA, and how the cells react to stain. Bacteria perform many useful functions as well as those that are bad. For example, one species lives in the human intestine and manufactures vitamin K, which is needed for blood clotting.

a. Nucleic acid analysis may be used to distinguish strains of bacteria. Nucleic acid is found in every living cell. There are two main types of nucleic acid: DNA (Deoxyribonucleic acid–the material that carries the genetic information in an organism) and RNA (ribonucleic acid–nucleic acid that translates genetic information into proteins). RNA translates genetic information in proteins. Several kinds of RNA are found, including transfer RNA, ribosomal RNA, messenger RNA, and small nuclear RNA. RNA is quite similar in structure to DNA. The content and sequence of RNA and DNA are used to distinguish between strains of bacteria. The use of RNA and DNA testing is more definitive than other methods, such as visual microscopic comparisons. On the basis of RNA sequencing, bacteria are in two major groups: eubacteria and archaebacteria. Each of these groups is further divided based on RNA sequencing.

b. In 1884, Danish scientist Christian Gram developed a stain that would aid in classifying bacteria into one of two groups, hence the Gram-stain test is used. ***Gram staining*** is a method of differentiating between bacterial species based on chemical and physical properties of cell walls. The method allows bacteria cells to be placed in two groups: Gram-positive and Gram-negative. Bacteria with cell walls containing small amounts of peptidoglycan and, usually, lipopolysaccharide are Gram-negative. Bacteria with walls containing large amounts of peptidoglycan and no lipopolysaccharide are Gram-positive. The exact staining reaction is not understood but differences in cell wall reactivity are highly useful. Examples of Gram-negative bacteria include typhoid (*Salmonella typhi*) and widely-known intestinal bacteria referred to as *E. coli (Escherichia coli*). Examples of Gram-positive bacteria include mouth-living actinomyces (*Actinomyces odontolyticus*) and oral streptococci (*Streptococcus pyogenes*), which causes common sore throat.

c. Bacteria are also classified based on their response to oxygen and how they obtain their energy.

    (1) Bacteria species may be grouped into three classes based on their response to gaseous oxygen. Aerobic bacteria must have oxygen to exist. Anaerobic bacteria cannot survive in the presence of gaseous oxygen. Facultative bacteria prefer to live in gaseous oxygen but can do with out it.

    (2) Bacteria species are in two classes based on how they obtain their energy. Heterotrophic bacteria consume organic compounds and break down the materials to gain nutrients. Bacteria that live in decaying material or that are involved in fermentation and respiration are heterotrophs. Autotrophic bacteria create their own energy by using light or chemical reactions.

d. Depending on the strain, bacteria may cause food to spoil and disease as well as serve useful roles in the environment. Some bacteria are used in food production, such as those used in making cheese, vinegar, and yogurt.

2. The kingdom of Protista has more than 65,000 different species of eukaryotic organisms. The kingdom is usually organized into nine phyla, including Sarcodina, which include amoeba, and five phyla containing algae. Protista are unicellular though some form colonies in which cells may specialize. Many algae have the ability to capture sunlight and use it to convert materials into chemical energy through a

process of photosynthesis. Species in the kingdom Protista are not as prominent in food spoilage as are species in other microbe kingdoms. However, some cause illness. An example is amebiasis, which is caused by one-celled parasite named *Entamoeba histolytica*.

a. Protozoa are often classified with the Protista. ***Protozoa*** are eukaryotic microorganisms that are sometimes classified in the kingdom Animalia and sometimes in the kingdom Plantae. Some protozoa may be transported with food materials and cause human disease, as parasites in the human body. Others are transmitted in other ways, such as those that cause malaria that are transmitted by mosquitoes.

3. The kingdom of Fungi has about 100,000 species in four phyla: zygomycota (black bread mold often found on bread, fruit, and other foods), ascomycota (includes yeasts, truffles, and others), basidiomycota (mushrooms), and deuteromycota (includes disease-causing species such as ringworm and is the source of penicillin though some are used to flavor cheese). Some fungi are multi-cellular, as are the molds, and others are uni-cellular (one-celled), as are the yeasts. Fungi are eukaryotic, as they have complete cells. Many fungi are saprophytic, which means that they feed on dead organic material including human food materials.

a. As with bacteria, DNA and RNA sequencing can be used to establish evolutionary relationships in the classification of Fungi. Once this has been done, a tree of evolutionary sequence can be prepared to depict relationships, similarities, and differences.

b. ***Molds*** are among the easiest Fungi to find and study as related to food. A fuzzy substance growing on the surface of bread is mold. Some mold organisms typically grow end to end to form filaments or hyphae. A mass of hyphae forms a mycelium. Other mold organisms produce stalks and spores, as with mushrooms. ***Yeasts*** are more difficult to identify because they are smaller, unicellular organisms.

4. A ***virus*** is a biological particle comprised of genetic material and protein. Most scientists do not consider viruses to be living organisms; others disagree. Since viruses are not cells, they reproduce only by invading another cell such as bacteria. Viruses are not readily classified into existing systems and are often not included in modern classification systems.

a. A general system to classify viruses has been developed that is quite similar to living organisms. The International Committee on Taxonomy of Viruses has developed a classification system using five stages. The stages are: order, family, subfamily, genus, and species. Another classification has been developed using seven groups which distinguish viruses based on their mode of replication and genome type. Today, the two systems are used together in classifying viruses.

b. Viruses can also be classified on the basis of structure and observed characteristics. These are: helical, icosahedral, enveloped, and complex viruses (these are covered in the next objective). Observation of structure is needed for this classification.

C. Microorganisms are named using the binomial system that is derived from the scientific classification. This is known as a ***binomial name***, which consists of two-word names.

The first word is the genus of the organism; the second word is the species. In writing binomial names, the first letter of the genus is capitalized; all other letters are in lower case. The entire name is in italics type face or underlined. An example is the bacterium that causes whooping cough: *Bordetella pertussis*.

Use TM–A, TM–B, and TM–C to outline and present the content of this lesson. Use TM–B to present the seven-stage modern classification system. Students may be referred to biology or microbiology textbooks for additional information on the classification of microorganisms. Students may be assigned to individually research selected microorganisms and prepare written or oral reports on their findings.

Objective 2: Describe the structure of bacteria, yeasts and molds, and viruses.

Anticipated Problem: What is the structure of bacteria, yeasts and molds, and viruses?

II. Bacteria, yeasts, and molds can usually be seen with a microscope. Viruses are typically too small for an ordinary microscope and must be studied with an electron microscope and, now, tissue culture may be used with virus study. Structure involves morphological features. ***Morphology*** is the study of the form and structure of organisms. A major distinction among microorganisms is that of cell structure: prokaryotic and eukaryotic.

A. The general structure of bacteria is that of a ***prokaryotic cell***, lacking well-defined nuclei and membrane-bound organelles. The chromosomes are made of a sing closed circle of DNA. Beyond that, they are of many shapes and sizes, though all are microscopic.

1. The cells of bacteria are incomplete when compared to most other cells. Bacterial cells do not have a nucleus or membrane structures known as organelles. Bacteria live at a wide range of temperatures and in conditions with or without oxygen. Bacteria reproduce by fission, which is splitting of a cell into identical daughter cells.
2. In general, the structure of bacteria involves the following structural features: (Note: Some structural features vary with the species being studied.)
    - capsule—protective covering that prevents drying out and protects it from being engulfed by larger microorganisms
    - cell envelope—typically a three-layered membrane that encloses the organism
    - cell wall—a rigid wall composed of protein-sugar (polysaccharide) molecule that encloses the cytoplasm
    - cytoplasm—also known as protoplasm, a gel-like composition of water, enzymes, nutrients, wastes, and gases that contain ribosomes, a chromosome, and plasmids
    - cytoplasmic membrane—a layer of phospholipids and proteins that encloses the interior and regulates the flow of materials in and out of the cell
    - flagella—some species use flagella for locomotion; some have one flagellum at an end, while others may have two or several flagella

- nucleoid—the region of the cytoplasm where the chromosomal DNA is located
- pili—present in some species, pili are hairlike projects on the outside of the cell; help bacteria attach to other cells and surfaces (Without pili, bacteria that cause disease lose their ability to infect because they are unable to attach themselves to host tissue.)
- ribosomes—structures inside bacteria that translate genetic code from nucleic acid to amino acids

B. Yeasts and molds are eukaryotic organisms. A ***eukaryote*** is an organism with complex cell(s). The genetic material is organized into membrane-bound organelles. The eukaryotes include the kingdoms of plants, animals, and fungi (the kingdom Fungi includes yeasts and molds). These cells are typically larger than prokaryotic cells. (Note: Eukaryotes are sometimes said to be a superkingdom, empire, or domain because three kingdoms have the shared cellular attribute.)

1. The overall structure of a eukaryote cell includes the following:
   - cell wall—cell walls provide rigidity for a cell and control water influx by osmosis; are present in fungi, algae, and plant cells
   - plasma membrane—present in all eukaryotic cells; located inside the cell wall on cells with walls; serves as a semipermeable barrier between outside and inside of cell
   - cytoplasm—watery substance that contains proteins, sugars, and salts; organelles are suspended in cytoplasm; fungi and algae have single membrane-bound vacuoles
   - nucleus—double membrane-bound organelle containing chromosomal DNA; contains nucleolus, which is where RNA synthesis occurs
   - endoplasmic reticulum—membrane tubes and plates which synthesize and transport proteins and lipids
   - golgi bodies—flattened, membrane-bound sacs and vesicles; vesicles secreted by endoplasmic reticulum fuse with Golgi and, after processing, are secreted into organelles or plasma membrane
   - peroxisomes—membrane-bound sacs secreted from Golgi; contain amino acid and fatty acid-degrading enzymes and catalase, the enzyme which detoxifies hydrogen
   - mitochondria—bound by a double membrane, mitochondria are involved in respiration and oxidative phosphorylation in aerobic organisms; ATP production occurs here
   - chloroplasts—found in cells which carry out photosynthesis; comprised of chlorophyl
   - flagella—found on some cells, particularly those without cell walls; tube-like extensions of the cell membrane which provide mobility
2. Yeasts are unicellular (one-celled) fungi. About 1,500 species of yeasts have been identified. Some yeasts are important in food production. One example is that used in baking and fermenting beverages: *Saccharomyces cerevisiae*. This species is also a

model used in cell biology research. A special database of *S. cerevisiae* is maintained: www.yeastgenome.org/

a. Yeasts reproduce by binary fission or budding to create new individuals from a parent cell. With binary fission, the parent divides into two parts that are of about equal size. Both cells are known as daughter cells; no parent cell remains. With budding, the nucleus has moved to the side of a cell and a small portion protrudes from the cell like an outgrowth or a small bud. The nucleus divides with a small portion (known as daughter nuclei) in the outgrowth. A cell wall grows between the parent cell and outgrowth resulting in separation of the outgrowth from the parent organism.

b. Yeasts are eukaryotic organisms and have cell structures similar to that outlined above.

3. Molds are multicellular, eukaryotic fungi. Most molds are filamentous. ***Filamentous*** means that the mold grows in a threadlike manner. Molds produce spores that are readily transported by air, water, and insects. In the right environment (such as on a slice of bread), a spore begins to grow and produces hyphae (threads). The hyphae form a tangled mass, which makes molded bread have a fuzzy appearance. The ends of the hyphae will produce more spores, which continue the cycle of mold growth. Most molded food should be thrown away. Some molds are used in making food such as Roquefort cheese, which owes its flavor to mold.

a. The structure of a mold cell is similar to that of the eukaryotic cell. Masses of mold cells appear as a long thread with a spore at one end. (Note: Mold and mildew are quite similar. Mildew is a grey, mold-like growth fungus organism. A fungus disease on some plants is also referred to as mildew and can be controlled by the application of a pesticide known as fungicide.)

C. Viruses are not complete cells though they consist of genetic material in a protective protein coat known as a ***capsid***. Viruses are known as particles–not as cells or organisms. A complete virus particle is known as a ***virion***. (Note: Viruses are contrasted with prions and viroids–virus-like structures that lack either protein or genetic material.) Most scientists feel that viruses are not living organisms, as they do not meet the criteria of being a living organism. Viruses are typically much smaller than bacteria though some may be as large. Size is typically 10 to 300 nanometers. Scanning and transmission electron microscopes are used to see the particles.

1. The structure of viruses requires methods of observation more powerful than the ordinary microscope. A virus has a capsid, which is an outer shell, and contains subunits of protein. Capsids protect the genetic material in a virion, determine if it is suitable for infection, and initiate an infection by attaching to and "opening" the target cell into which it injects genetic material.

2. Four morphological virus types have been found:

    - helical—the structure resembles a spiral staircase made up of small tubes; rod-shaped virions may be short and rigid or long and flexible; genetic material is inside the tubes; an example is the tobacco mosaic virus
    - icosahedral—the structure is ring-shaped capsomers (a capsomer is a morpholocial unit of a capsid) somewhat resembling a geodesic dome; this

structure is an efficient way of enclosing protein; an example is hepatitis B, which is comprised of 240 proteins assembled to form one capsid

- enveloped—the structure has an additional outer ring comprised of proteins coded with the viral genome; this ring provides added protection from enzymes and chemicals; an example is HIV
- complex—these viruses have additional structures over the others, such as a protein tail, outer wall, or central disk structure; examples are the poxviruses

Use TMD, TM–E, and TM–F to outline and present the content of this problem area. Use TM–G, TM–H, and TM–I to show illustrations of particle and cellular structures. Students may be referred to textbooks on microbiology or biology. In some cases, web sites may be useful such as the interactive Cells Alive site: www.cellsalive.com/cells. Students may be required to go to this site as homework or during supervised study in class and prepare drawings of microorganisms that may pose food safety issues.

Objective 3: Identify conditions for microbial growth and methods of assessing organisms present in food.

Anticipated Problem: What conditions are needed for the growth of microorganisms in food? How is the presence of microorganisms in food assessed?

III. ***Food microbiology*** is the study of microorganisms that affect food materials. The organisms may have a detrimental effect or they may be used to gain desired food products.

A. Microorganism growth is related to the habitat that is provided by food materials. ***Habitat*** is the place where organisms live, grow, and reproduce. Food materials provide an ideal source of food for the growth and reproduction of microorganisms. Unfortunately, microorganisms alter their habit causing changes in the food materials where they may exist.

1. Most microorganisms thrive in warm environments. Microorganisms on food materials that are not kept refrigerated or frozen provide near ideal conditions for their growth. Milk will spoil rapidly unless refrigerated to slow the rate of growth of microorganisms.
2. Most microorganisms need moisture to live and grow. Dry conditions usually bring their activity to a near halt. Most food materials, however, contain moisture that promotes microorganism growth.
3. Most microorganisms need nutrients to live and grow. Food materials are, of course, ideal in this regard. Protecting food materials from exposure to microorganisms helps prevent contamination. Packaging is often used for this purpose.
4. Microorganisms prefer a food pH range of 4.6 to 8.0. Acidic foods with a pH below 4.6 will discourage the growth of microorganisms.

B. The presence of microorganisms in food can be assessed in several ways. Certainly, growths on food or the presence of abnormal or undesirable odors and flavors is a sign of spoilage. Food scientists use additional methods for assessing the presence of microorganisms.

1. Specimens of growth on foods can be examined with a microscope. Molds are usually readily identified in this manner.
2. Cultures of small amounts of substances with organisms from the food materials can be made to determine the presences of microorganisms.
   a. The process used is known as cell culture. ***Cell culture*** is the process by which prokaryotic or eukaryotic cells are grown under controlled conditions. A culture is comprised of the microorganisms that grow in a culture medium.
   b. A ***culture medium*** is the solution containing all of the nutrients to support the growth of microorganisms. Of course, the medium must be incubated in an environment that promotes microorganism cell growth. The medium used by food scientists is typically agar.
   c. ***Agar*** is a culture medium that gels and stabilizes. It often involves extract from red algae or seaweeds. ***Nutrient agar*** is agar that has been enriched to promote the growth of bacteria, fungi, and other unicellular organisms or particles of viruses. An ***agar plate*** is a sterile petri dish that contains agar or nutrient agar. The preferred agar in some food and kitchen work is tryptic soy agar. It can be obtained as powder and prepared into the petri dishes or the dishes can be obtained ready-to-use. Agars are obtained for special purposes, as different microbes require suitable nutrient gels for growth.
   d. ***Inoculation*** is the process of transferring potential microorganisms to the agar. Microorganisms placed on a plate and held at the correct temperature will produce colonies of microorganisms in several hours. The inoculated agar plate is incubated at a temperature of 35°C (95°F). Many are incubated for 24 hours.
   e. After incubation, agar plates are observed for the growth of microorganisms. In some cases, large colonies will develop that can be seen without a microscope. In some cases, a hand lens may be satisfactory. In most cases, a microscope will be needed to closely examine the morphology of the organisms for proper identification.

Use TM–J to outline and present the content of the objective. Use TM–K to summarize protocol in making a culture. Use LS–A as a lab activity for students to culture samples of microorganisms obtained from food or surfaces where food is prepared. Students may be referred to textbooks on microbiology or biology for additional information.

Objective 4: Describe the role of temperature, water activity, pH, oxygen, redox, and antimicrobials on microbes.

Anticipated Problem: What is the role of temperature, water activity, pH, oxygen, redox, and antimicrobials on microorganisms?

IV. A major goal in the food industry is to control food spoilage by microorganisms. This is achieved in two major ways: preventing contamination and preventing the growth and reproduction of microorganisms.

A. Temperature can be used to suppress or destroy the lives and growth of microorganisms. High temperatures destroy and rid food of microorganisms in which they are present. The temperature range of 4°C (40°F) to 60°C (140°F) is the danger zone. This is where many microorganisms can thrive, particularly within the mid-part of this range. Higher temperatures destroy organisms. Lower temperatures make microorganisms inactive and unable to reproduce. Heat during canning and cooking destroys microorganisms. Refrigeration and freezing reduce the growth of microorganisms.

B. Water activity refers to moisture levels in food materials. Some food products are dried, such as raisins and dates. Without a sufficient amount of moisture, microorganisms that cause food spoilage fail to survive, reproduce, and grow.

C. pH refers to the acidity or basicity of food materials. Most microorganisms thrive above 4.6 pH but a pH above 9.0 tends to reduce their activity. They prefer neutral conditions with a Ph of 7.0. Orange juice, vinegar, and similar liquids are acidic and do not support the growth of microorganisms. Pickled cucumbers, eggs, or meat products resist spoilage because microorganisms cannot flourish in the acidic solution.

D. Most microorganisms need oxygen to live, grow, and reproduce. Though some bacteria are anaerobic, many species of bacteria, molds, and fungi thrive in the presence of oxygen. Removing oxygen (and air contaminated with microorganisms) will reduce contamination and fail to support the growth of organisms that require oxygen.

E. Redox is a chemical process in which atoms have their oxidation number changed. The process involves both reduction and oxidation. In reduction, an electron is gained by the molecule, atom, or ion. This is opposite oxidation which is the loss of an electron by a molecule, atom, or ion. Some substances oxidize others; other substances are reducers.

F. Antimicrobials are sometimes used to reduce or eliminate populations of microorganisms. An ***antimicrobial*** is a substance that destroys or suppresses microbial organisms. Antimicrobials are often categorized by the species controlled such as antibacterials (or antibiotics in medicine) are used to control bacteria. Antifungals are used to control fungi. Prominent uses of antimicrobials are in cleaning surfaces where food products are produced or prepared for consumption. An example is the use of a bleach solution to wash or wipe surfaces in kitchens and canneries. Antimicrobial resistance is jeopardizing the use of antimicrobial products, such as those used in human medicine.

Use TM–L to outline and present the content of this lesson. Have students investigate methods of destroying or reducing microorganisms in food products and how these *methods are used to prolong the shelf life of foods, with examples being canning, freezing, drying, and packaging in containers that remove oxygen. Students may be referred to textbooks on microbiology or food science. Students may prepare a bleach solution to sterilize surfaces. (A mix is 20 ml of chlorine bleach with 1L of tap water. The solution is lightly mist sprayed or wiped on a surface with a cloth or sponge. Such surfaces include counter tops, sinks, refrigerator surfaces, microwave surfaces, and table tops. Always wear goggles, rubber/plastic gloves, and appropriate apron or lab coat. Note: A bleach solution may damage some surfaces if not wiped off immediately. Always use solutions within a few hours of their preparation.)*

Objective 5: Explain food spoilage and how it impacts shelf life.

Anticipated Problem: What is food spoilage and how does it impact shelf life?

V. ***Food safety*** is keeping food wholesome and free of organisms or substances that cause illness. Following proper practices in food production, processing, storage, and preparation helps assure safety.

A. ***Spoilage*** is food loss due to decay, improper processing or storage, and planning in terms of product acquisition and need. Spoilage is often due to bacterial action in food materials. In most cases, the food is unsafe for consumption. Consuming spoiled food may lead to food poisoning, serious illness, and death of the individual. Some foods can be observed for signs of spoilage. Food that has a foul odor, a non-normal color, and an unnatural consistency is likely spoiled–do not eat it! Of course, some microorganisms may be in food and not readily visible unless colonies have formed such as mold on bread. Laboratory testing may be needed.

B. ***Shelf life*** is the length of time that a food product is safe to consume. At the end of the time and if properly stored, the food is still safe to eat. Various methods of preservation and packaging are used to enhance shelf life. For example, canning is used to extend the shelf life of fresh peas, which normally is only a few days, for several months and/or years. Dates are often stamped on food product containers or labels. Milk containers, for example, often have dates stamped on them. This means that the product should be wholesome and useable if properly stored until the date that is stamped.

C. Food spoilage results from several actions. Preventing spoilage is a high priority in the food industry.

1. Preventing contamination by spoilage agents is important. ***Contamination*** is the accidental or purposeful introduction of substances that cause food spoilage. This includes microorganisms as well as chemical contamination. Preventing the introduction of microorganisms into food helps prevent later growth and activity of the microorganisms to cause spoilage.

a. Air movement should be controlled. Air carries molds, yeasts, and bacteria. Preventing air contact with food materials helps prevent contamination with food spoilage organisms. In some cases, filters can be used on fans to remove contaminants.

b. Surfaces of counters and tables and people working with food are sources of contamination. Keeping surfaces clean helps reduce the likelihood of contamination. Having people handling food wash their hands, wear clean clothing, and use hair nets and other practices helps reduce the likelihood of contamination.

c. Controlling the temperature at which food is stored will help reduce the likelihood of spoilage. Some foods and contaminants are very sensitive to temperature, such as the bacteria in milk. Other foods may become rancid or otherwise develop an off-flavor if stored improperly and/or too long.

d. Most microorganisms require moisture in order to live and grow. Any process that lowers moisture levels also helps protect food from spoilage.

2. Preventing the growth of microorganisms helps maintain food quality. This refers to establishing conditions under which microorganisms do not thrive.

a. Heat can be used to destroy microorganisms. Heat causes the death of microbes, depending on how it is used. Two concepts are involved: sterilization and pasteurization. ***Sterilization*** is the destruction of microorganisms that are present in food materials. None survive the sterilization process. Sterilization is the most effective approach in extending shelf life of a food product. ***Pasteurization*** is the use of heat to destroy selected pathogenic microorganisms that are present in food materials. Milk and fruit juices are often pasteurized. Different microorganisms have different temperature levels for destruction. Heating milk to 72.9°C (162°F) for 15 seconds destroys the pathogenic microorganisms typically found in milk. This, however, does not necessarily destroy other microorganisms that may be present.

b. Inhibiting the growth of microorganisms is another practice that can be used with food products. The microorganisms are not destroyed but the conditions under which they grow are made very unfavorable. Acid content of the food material is a factor as higher acidic foods are less likely to support the growth of microorganisms. In some cases, foods have the water removed. Dried foods inhibit the growth of microorganisms.

Use TM–M and TM–N to outline and present the content of this objective. Refer students to food science and biotechnology books for additional information. Students may also demonstrate pasteurization in a laboratory activity.

Objective 6: Describe how food is contaminated leading to food poisoning and food-borne infections.

Anticipated Problem: What causes food poisoning and food-borne infections and how can they be prevented?

VI. Foods sometimes pose threats to human well-being. These threats are often in two forms: food poisoning and food infections. It is sometimes difficult to separate these two forms;

however, food infections are included in the broad meaning of food poisoning. (Note: Some people have allergies to certain foods such as peanuts, milk, and eggs. The consumption of such foods may cause severe allergic reactions though the food product is wholesome.)

A. ***Food poisoning*** is an acute gastrointestinal or neurologic disorder caused by bacteria and other microorganisms, viruses, parasites, or harmful chemicals that are in foods. (Note: Parasites are not covered in this lesson. One of the most common is amebiasis, which is caused by the one-celled parasite, *Entamoeba histolytica*. This illness is most common in developing countries and among people who travel to these countries. Poor sanitary conditions contribute to the presence of these organisms. Infected food and water are common sources.)

   1. Bacteria contribute to food poisoning in three ways:
      - infect the individual following consumption of contaminated food
      - produce a toxin in food before it is consumed
      - produce a toxin in the gastrointestinal tract after the food has been consumed
   2. A ***food-borne infection*** is an illness caused by ingesting a microorganism in a contaminated food. This is probably the easiest microorganism-induced food-borne illness to get. These illnesses are in two groups: those in which the food is the carrier and those in which the food is a substrate and a carrier. A ***substrate*** is the substance in food from which the microorganisms derive nutrients. Most food materials have an abundance of substrates for microorganisms, such as mold, bacteria, yeasts, and viruses. Several infectious bacteria are associated with food poisoning. Other organisms and viruses may also cause food illnesses.
      a. Some food toxins are said to be preformed. This means that they are in food before preparation. Being resistant to heat, some of the bacteria may remain active in foods after they have been cooked. Two examples are included here.
         (1) *Staphylococcus aureus* is commonly called a staph infection. *S. aureus* is caused by a spherical-shaped bacterium that frequently lives on the skin and in the nostrils of a healthy person. It is a Gram-positive that appears as grape-like clusters when viewed with a microscope. Large, round golden-yellow colonies may be formed. There are two subspecies: 1—*S. aureus anaerobius* is an anaerobic organism and not commonly encountered and 2—*S. aureus aureus* is an aerobic organism that is most often found.
         (2) *Clostridium botulinum* is the source of a potent toxin known as botulin. It can cause serious nerve damage and paralysis leading to respiratory and musculoskeletal paralysis. All forms of botulin can be fatal and should be treated as medical emergencies. On average, 110 cases occur in the United States each year. Three forms of botulism occur: foodborne, infant, and wound.
            (a) Foodborne botulism results from consuming foods that are contaminated with the botulinum toxin. Slightly over one-fifth of all botulin cases are foodborne.

(b) Infant botulism results from consuming spores of the botulinum bacteria. These spores grow in the intestines and release the botulin toxin. Nearly three-fourths of all botulin cases are of this type.

(c) Wound botulism results from toxin produced by a wound that has been infected with *C. botulinum*. It is a rare type that can largely be prevented through sanitation and proper wound care.

b. Poisons are sometimes produced in the gastrointestinal tract. These are produced by organisms that survive cooking and are spore-forming bacteria.

(1) *Clostridium perfringens* is a Gram-positive, anaerobic, rod-shaped bacterium that forms spores. It is a normal part of the environment in rotting vegetation, soil, and insects, among others, as well as the human intestines. This is the third most common cause of food poisoning in the United States. When cultured, *C. perfringens* produces flat, spreading, translucent colonies with irregular margins. Anaerobic conditions are needed. A Nagler agar plat is used. The medium should contain 5–10 percent egg yolk. Half of the culture plate may be inoculated with antitoxin to serve as a control in identification.

(2) *Campylobacter jejuni* is the most common cause of human diarrhea. Though severely debilitating, it is rarely life-threatening. It has been linked to other disease, such as the neurodegenerative disease commonly known as GBS. *C. jejuni* is caused by a curved, rod-shaped, gram negative bacterium. It is commonly found in animal feces, particularly chickens. Contaminated drinking water, unpasteurized milk, incorrectly prepared poultry and meat, and lack of sanitation in the food preparation area lead to unsafe food.

(3) *Escherichia coli* is one of the main organisms that live in the intestines. In an average day, a human excretes 100 billion to 10 trillion *E. coli* organisms in feces. The organism is an aerobic species that is Gram-negative, non-spore-forming, and rod-shaped. The bacteria ferment lactose resulting in gas production within 48 hours at 35°C (95°F). The gas is released by flatulence. The presence of *E. coli* in water, on plants, and in food is an indication of fecal contamination. Sanitation is important in keeping *E. coli* out of food. Crop harvesters, food merchandisers, and food preparers should wash their hands thoroughly after using a toilet and keep all surfaces clean. Wearing clean, non-contaminated clothing is also important. There are many strains of *E. coli*, with some being very useful in genetic engineering research. New strains that cause human illness regularly arise through evolution, with *E. coli* O157:H7 being most virulent. Those normally present in the human body usually pose no problem. New strains are more than likely the cause of human illness.

(4) Salmonellosis is an infection caused by *Salmonella* bacteria. There are several species within the genus that are known as potential sources of disease. Individuals with salmonellosis typically have diarrhea, fever, vomiting, and abdominal pain within 6 to 72 hours after being infected. Most individuals recover well without medical treatment. Dehydration may result thereby

creating the need for hospitalization to receive intravenous fluids. The most common Salmonellosis is caused by non-typhoidal *Salmonella*. It is contracted by eating raw or undercooked eggs, poultry meat and beef, and other foods which have been contaminated during preparation. Pet turtles, lizards, and other reptiles carry Salmonella bacteria on their skin; therefore, exercise caution in handling such animals and always wash hands thoroughly afterward.

(5) Other food-borne illnesses include:

- Listerosis, which is caused by *Listeria monocytogenes*, and is rarely a human health problem.
- *Vibrio parahaemolyticus* is a Gram-negative bacterium species found in saltwater. Ingestion usually occurs in contaminated seafood via fecal-oral contact.
- Yersiniosis is an infectious disease caused by the *Yersinia enterocolitica* bacterium. It is most common in young children and manifests itself as fever, abdominal pain, and diarrhea. Pigs are a major source of the illness though rodents, rabbits, sheep, cattle, horses, dogs, and cats may also be a source of *Y. enterocolitica*.

B. A non-bacterial source of food poisoning is the *Norwalk virus*. It was identified in 1972 after an outbreak in Norwalk, Ohio. The illness symptoms include stomach pain, diarrhea, and vomiting in humans. It is sometimes called stomach flu. The virus responsible is characterized as having small round structures. Genomic study has found that this virus is in the Caliciviridae family. It is transmitted via fecal to mouth contact with contaminated food and water. Contaminated ice machines have been found as a transfer source. It may also be transferred from one person to another. Diagnosis of the Norwalk virus is routinely done with RT–PCR assays, with real-time RT–PCR tests now available. (Note: RT–PCR stands for reverse transcription-polymerase chain reaction. It is used to detect and quantify mRNA in small samples.)

C. Agents causing food poisoning and food-borne infection are controlled through sanitation and proper handling, including preparation, of food products. Applying all methods of preventing spoilage in food is essential. Here are some approaches to follow in preventing microbial food-borne illnesses:

- store food at the proper temperature
- cook food until the proper internal temperature is reached
- separate cooked and uncooked food
- properly wash hands and utensils
- marinate in the refrigerator
- consume only good food

D. Food can sometimes be contaminated with chemicals that cause food poisoning. These are non-living substances that enter food. Pesticides, petroleum fuels, cleaning agents, and other substances may be present in the food production and storage environment. Care should be used to prevent chemical contamination.

E. Another threat to food safety is bioterrorism. This involves the deliberate contamination of food to make it unsafe to eat. Steps are being taken to assure food supplies that are safe from bioterriorism.

F. Food quality assurance programs are voluntarily being used by producers of some food products. These programs provide guidance to assure that the food that is produced is safe to eat. Such regulations are more prominent in the pork and beef production industries that elsewhere. Vegetable producers follow practices to avoid contact with growing crops by manure and other products that may contain organisms that cause food poisoning.

G. Individuals who have diarrhea, vomiting, fever, and other symptoms may wish to have an accurate diagnosis. Having these symptoms does not make it possible to know if they were caused by bacteria, parasites, viruses, or chemicals. A ***stool culture*** is done to identify the cause of the infection and involves analyzing a sample of feces. This will require collecting a feces sample in a special container for such and taking it to a physician. All instructions on the container or from the physician should be followed. Properly store and transport to the physician. Laboratory analysis will be done to determine the cause of the illness. This may involve using chemicals as well as culturing samples using petri culture dishes similar and including Gram staining.

Use TM–O and TM–P to outline and present the content of this objective. Use TM–Q to show line art examples of bacteria organisms that may be present in food and on kitchen surfaces. Refer students to food science and microbiology books for additional information. Students may also demonstrate pasteurization in a laboratory activity. Hand washing and other sanitation practices may be investigated by students, reported, and demonstrated in class.

Review/Summary. Reviewing and summarizing the content may be combined, depending on student needs and situations. Use the objectives for the lesson to guide the review and summary process. Call on individual students to discuss the content that goes with each objective. Re-teach content as needed to assure that students have mastered the objectives.

Application. Supervised experience, agriscience activities, and other approaches may be used for student application of the content. In some cases, students will be able to apply the content in other courses in which they are enrolled or as independent study. Students may be asked to apply the safety information their personal food preparation plans in their homes or places of work.

Evaluation. Evaluation should concentrate on achievement of the objectives by the students. Questioning students throughout the teaching process will allow assessment of learner understanding. A sample written test is attached.

Answers to the Sample Test:

Part One: Matching

1. e
2. f
3. d
4. a
5. c
6. b

Part Two: Fill-in-the-Blank

1. Contamination
2. Spoilage
3. antimicrobial
4. virion
5. morphology
6. Gram staining

Part Three: Multiple Choice

1. b
2. c
3. a
4. d
5. a

Part Four: Short Answer

1. Prokaryotic cell lacks a well-defined nuclei and membrane-bound organelles. A eukaryotic cell is more complex and has a nuclei and genetic material in membrane-bound organelles.
2. Food microbiology is the study of microorganisms that affect food materials. It includes those that cause food spoilage as well as those that causing food poisoning.
3. With the modern system, organisms are classified using a seven tier system: kingdom, phylum or division, class, order, family, genus, and species. In addition, the species of some organisms are classified into strains.
4. Habitat conditions that promote the growth of food spoilage organisms include: warm environments, moisture, and nutrients.
5. Temperature and pH can be used to suppress the growth of organisms in food. Temperatures of 40°F or below and those above 140°F prevent organism growth or destroy organisms. Most organisms survive and/or thrive in the temperature range of 40–140°F. Organisms prefer substances with a pH near neutral. Most thrive between a pH of 4.6 and 9.0.

6. Actions to prevent food spoilage include: prevent contamination, control air movement, keep surfaces clean where food is handled and prepared, control the temperature, and lower the moisture level.
7. Food poisoning is an acute gastrointestinal or neurologic disorder caused by bacteria or other substances, viruses, or harmful chemicals in food. Kinds of food poisoning include: staph infections, botulism, *C. jejuni*, *E. coli*, salmonellosis, listerosis, *V. parahaemolyticus*, and yersiniosis. The Norwalk virus may also be listed. (Students only need to name any two to the kinds of food poisoning.)
8. Practices to prevent food-borne illness when handling food include: store food at the proper temperature, cook food until done, separate cooked and uncooked food, properly wash hands and utensils, marinate foods in a refrigerator, and consume only good food.

Test

Name ________________________________

FOOD MICROORGANISMS AND SAFETY

Part One: Matching

Instructions: Match the term with the correct definition.

a. agar
b. habitat
c. shelf life
d. inoculation
e. binomial name
f. taxonomy

_______1. The scientific name of an organism involving two words, genus and species.

_______2. The classification of living things.

_______3. The process of transferring potential microorganisms to a culture medium.

_______4. A culture medium.

_______5. The length of time that a food product is safe to consume.

_______6. A place where organisms live and grow; an environment in which organisms thrive.

Part Two: Fill-in-the-Blank

Instructions: Complete the following statements.

1. ____ is the accidental or purposeful introduction of substances that cause food spoilage.
2. ____ is a condition in which food is unsafe to consume due to contamination, decay, improper storage, or improper preparation.
3. A substance that destroys or suppresses microorganisms is an ____.
4. A complete virus particle is known as a ____.
5. ____ is the study of the form and structure of organisms.
6. ____ ____ is a method of differentiating between bacterial species based on the chemical and physical properties of cell walls.

▶ Part Three: Multiple Choice

Instructions: Circle the letter of the correct answer.

_______1. A subspecies within a species is a ____.

a. kingdom
b. strain
c. genus
d. order

_______2. A biological particle that is not considered to be an organism is a ____.

a. protozoan
b. bacterium
c. virus
d. fungi

_______3. A complete virus particle is known as a ____.

a. virion
b. fungus
c. cell
d. petri

_______4. An organism that grows threadlike structures is said to be ____.

a. round
b. circular
c. flat
d. filamentous

_______5. Agar that has been enriched to serve as a medium for a specific organism is said to be ____.

a. nutrient agar
b. liquid agar
c. petroleum
d. ready-to-use

▶ Part Four: Short Answer

Instructions: Answer the following questions.

1. What is the distinction between prokaryotic and eukaryotic cells? Draw an example of each cell and label the major parts.

2. What is food microbiology? Why is it used?

3. How are organisms classified using the modern system? What divisions or stages are used in the classification system?

4. What habitat conditions promote the growth of food spoilage organisms?

5. What are the roles of temperature and pH on food spoilage organisms?

6. What actions can be taken to prevent food spoilage?

7. What is food poisoning? What are two kinds of food poisoning?

8. What are some practices to prevent microbial food-borne illnesses when handling and preparing food?

MICROORGANISM CLASSIFICATION

- Taxonomy—classification of living things
  - grouping organisms based on similarities and differences, including DNA study
  - hierarchical categories are used
- Seven stage system:
  - kingdom
  - phylum or division
  - class
  - order
  - family
  - genus
  - species
- Species may be divided into subspecies or strains

SAMPLE CLASSIFICATION AND NAME OF *E. COLI*

- Kingdom: Monera
- Phylum: Proteobacteria
- Class: Gamma Proteobacteria
- Order: Enterobacteriales
- Family: Enterobacteriaceae
- Genus: *Escherichia*
- Species: *coli*
- Binomial name: *Escherichia coli*

BACTERIA ORGANISM CLASSIFICATION

- Bacteria—one celled organisms in the kingdom Monera
  - prokaryotic cells
  - perform many useful roles in the environment
  - some species cause food spoilage and disease
- Gram staining—method of differentiating between species
  - Gram-positive
  - Gram-negative
- Classes based on response to gaseous oxygen
  - aerobic
  - anaerobic
  - facultative

TM–D

FUNGI ORGANISMS

- Fungi—four phyla within kingdom Fungi
  - eukaryotic cells
- Molds—easiest fungi to identify
- Yeasts—more difficult to identify

TM–E

THE VIRUSES

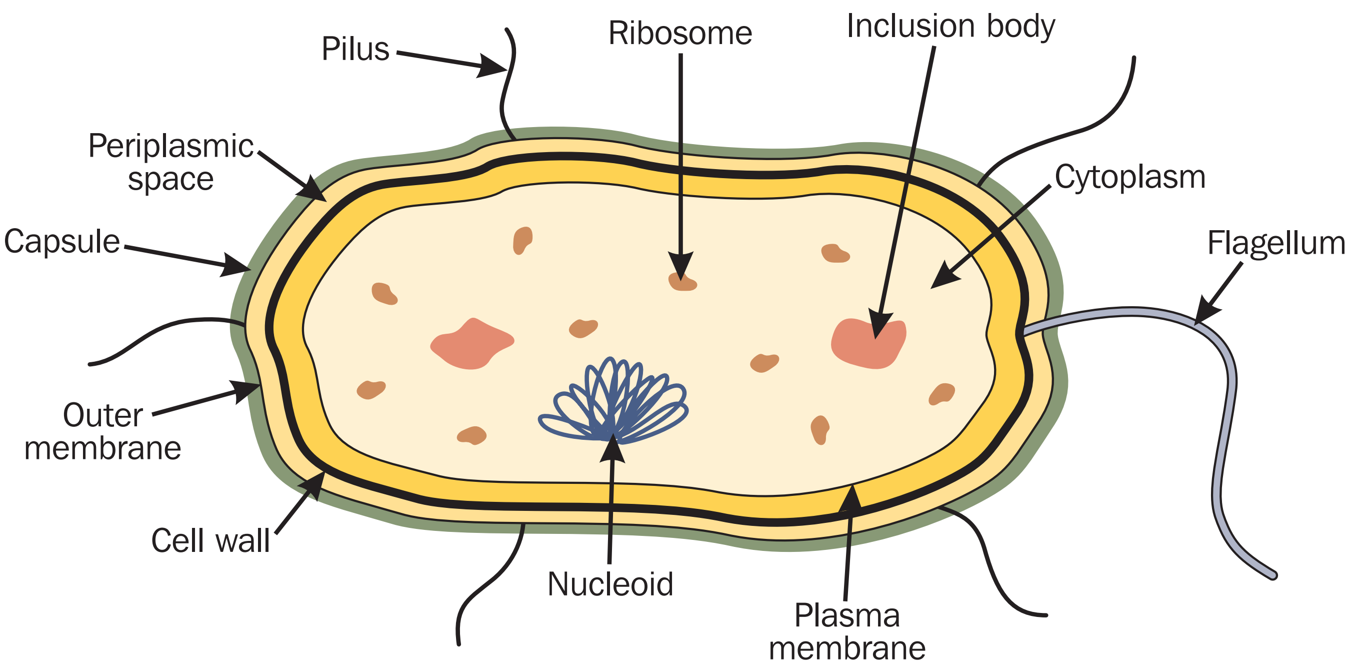
- Virus—a biological particle of genetic material and protein; not a living organism
- Classification:
  - based on numbered genogroups
  - order, family, subfamily, genus, and species may be used
  - no kingdom

TM–F

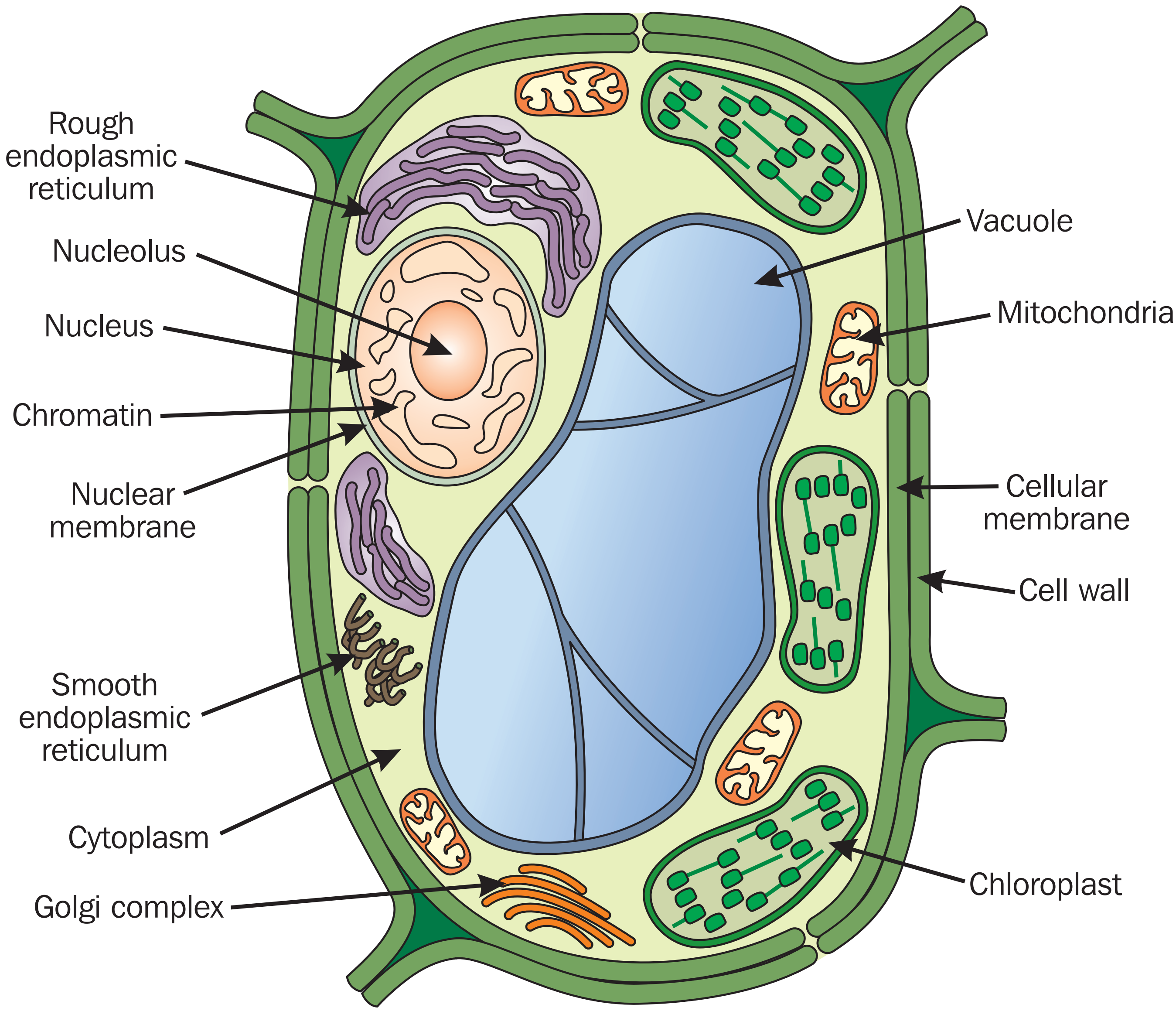
CELL STRUCTURE

- Morphology—study of form and structure of organisms
- Prokaryotic cell—lacks well-defined nuclei and membrane-bound organelles; bacteria
- Eukaryotic cell—complex cells with genetic material organized into membrane-bound organelles; yeasts, molds and other species

PROKARYOTIC CELL (BACTERIA)

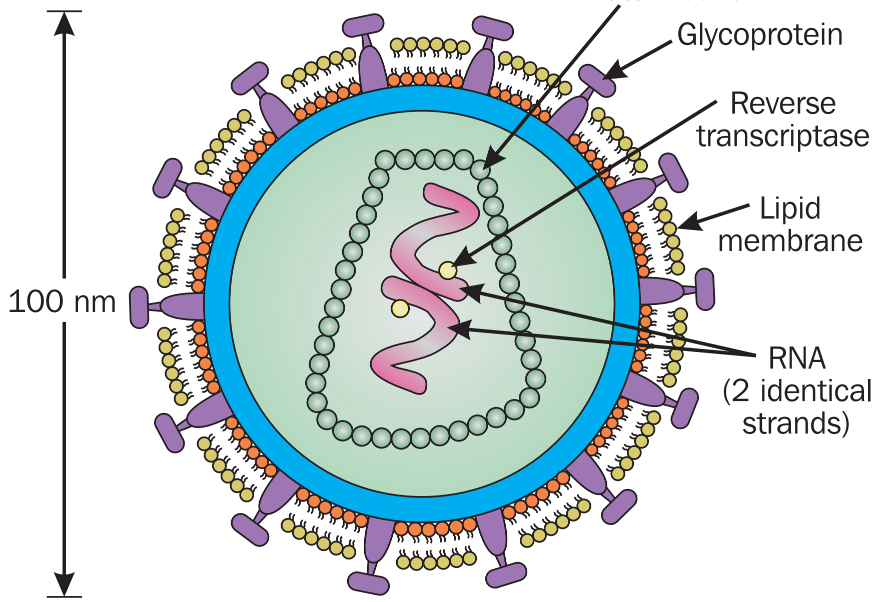


EUKARYOTIC CELL

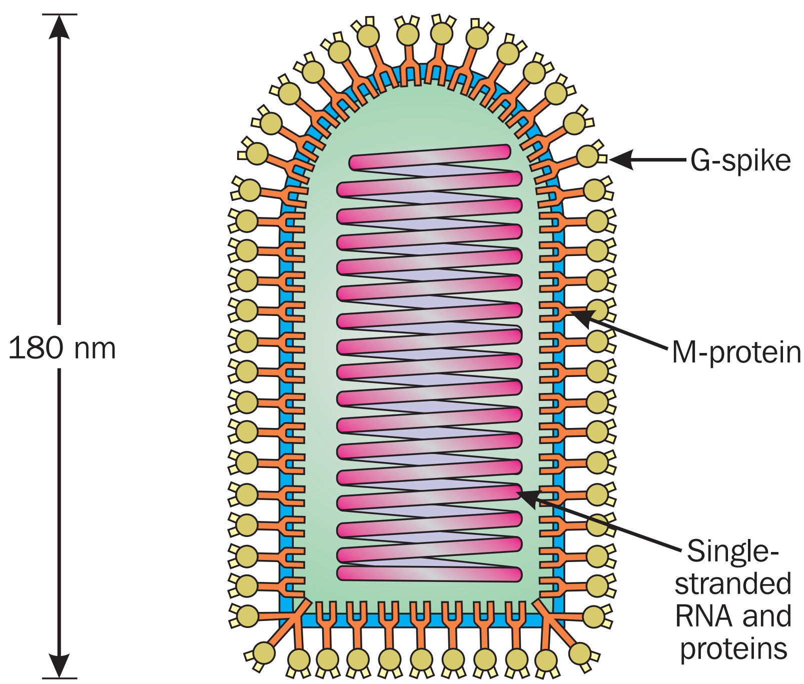


VIRUS STRUCTURE

HIV



Rabies Virus



MICROBIOLOGY AND FOOD

- Food microbiology—study of organisms that affect food
- Habitat—place where organisms live, grow, and reproduce
  - warm
  - moist
  - food nutrients
  - pH (prefer 4.6–8.0)

TM–K

CULTURES

- Cell culture—process of cells are grown under controlled conditions
- Culture medium—nutrient-rich substance that promotes cell growth
- Agar—culture medium that gels and stabilizes
- Nutrient agar—enriched agar to promote growth
- Agar plate—sterile petri dish holding agar
- Inoculation—transfer of potential microorganisms to agar
- Incubation—time for growth in good environment
- Identification—microscope, staining, genomic analysis, etc.

SUPPRESSION OF MICROBES

- Temperature—organisms prefer 40–140°F; below 40° and above 140° are unfavorable
- Water—required by organisms; reduce moisture in some foods to prevent spoilage
- pH—acidity and basicity of food affects microbes; below 4.6 pH and above 8.0 pH suppresses microbes
- Oxygen—most bacteria, molds, and fungi thrive with oxygen
- Redox—chemical process of electron gain; changing oxidation number
- Antimicrobial—a substance that destroys or suppresses microbes

FOOD SAFETY AND SPOILAGE

- Food safety—keeping food safe to eat
- Spoilage—food loss due to decay and improper handling; unsafe to eat
- Shelf life—length of time a food product is safe to eat; storage time

FOOD SPOILAGE

- Contamination—accidental or purposeful introduction of substances into food
- Air movement—air carries microbes
- Surfaces—keep surfaces clean; disinfect
- People—wash hands and wear clean clothing
- Temperature—control to prevent microbe growth and manage chemical processes in food
  - sterilization—destroy organisms with heat
  - pasteurization—destroy selected organisms with heat
- Moisture—reduce moisture levels

TM–O

HUMAN HEALTH THREATS

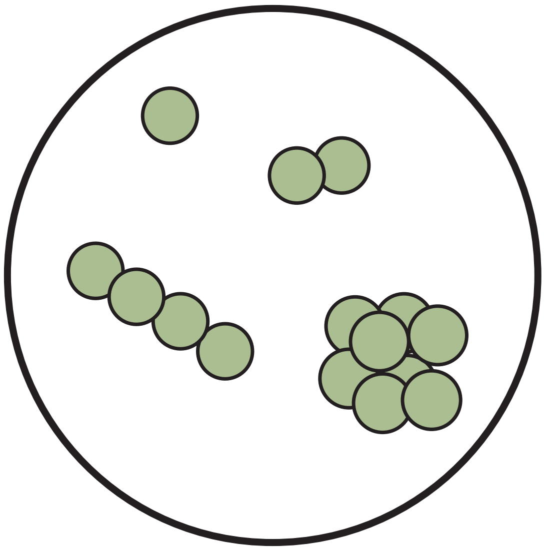
- Food poisoning—acute gastrointestinal or neurologic disorder
- Food-borne infection—illness caused by ingestion of food contaminated with microbes
- Examples:
  - preformed food toxins—*Staphylococcus aureus*, *Clostridium botulinum*
  - gastrointestinal poisons—*Clostridium perfringens*, *Campylobacter jejuni*, *Escherichia coli*, Salmonella spp, *Liseria monocytogenes*, *Vibrio parahaemolyticus*, and *Yersinia enterocolitica*

HOME FOOD PRACTICES

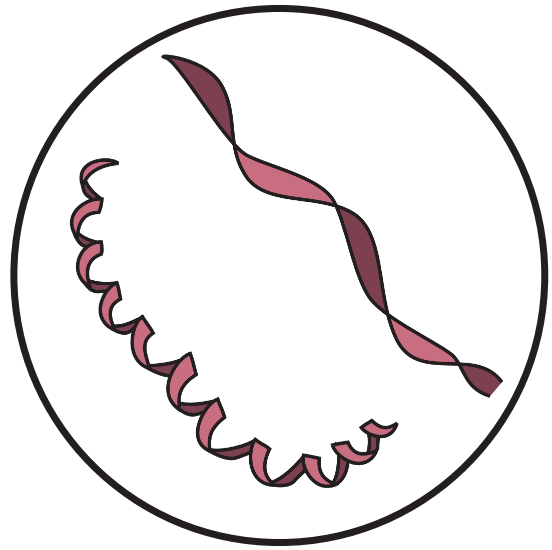
- Properly store food
- Cook until done internally
- Separate cooked and uncooked food (avoid cross-contamination)
- Properly wash hands, utensils, and surfaces
- Marinate in refrigerator
- Consume only food known to be wholesome

TM–Q

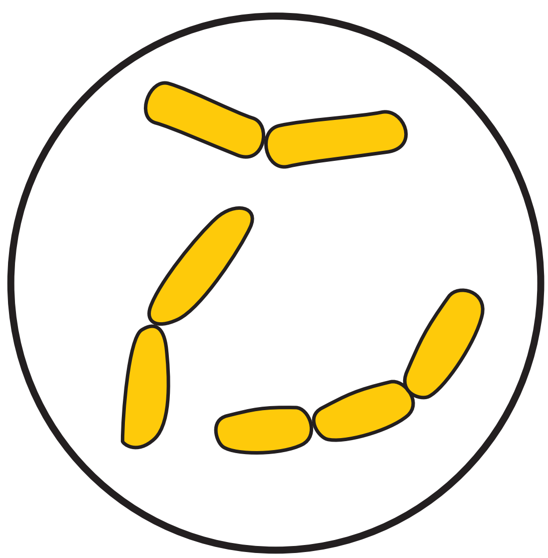
FOOD AND KITCHEN BACTERIA FORMS



Cocci



Spirilla



Bacilli

MAKING A CELL CULTURE

Problem:

The purpose of this activity is for students to make cell cultures using common sources of microorganisms.

Safety:

All students and the instructor should wear safety goggles, rubber/plastic gloves, and clothing–typically a lab coat. Wastes materials should be properly disposed of within guidelines of the school.

Background:

Read the Student Worksheet on Making a Cell Culture. Obtain and organize sufficient supplies for your class. Advance planning will be needed to be sure the needed petri dishes and agar are on hand.

Discuss the proposed activity with students. Have them read the procedure to be followed and discuss the activities. Be sure to discuss the importance of preventing contamination of the swab as well as the culture medium. This includes allowing organisms floating in the air to contact the agar as well as touching the end of the swab used to collect the sample.

If a sample is to be collected from a dry surface, wet the swab in boiled or sterile water and remove excess water from the tip by pressing it against the side of the bottle. Using a swab that is too wet will flow into the control side of the petri dish and contaminate the control area of the dish.

If a sample is liquid (such as milk or dishwater), dip the swab into the liquid and press excess liquid out against the inside of the container.

Students may work together as pairs. In addition, to labeling each half of the petri dishes, students may also add their names in small print to avoid later confusion.

After incubation, have students observe and describe what they see. All petri dishes may be lined up for class observation. Use an overhead projector or a video camera to show four petri dishes at one time. Discuss. Individual students may be asked to describe what they have observed.

Name ____________________________________

MAKING A CELL CULTURE

Purpose:

The purpose of this activity is to determine the presence of microorganisms on foods or surfaces by making a cell culture.

Materials Needed:

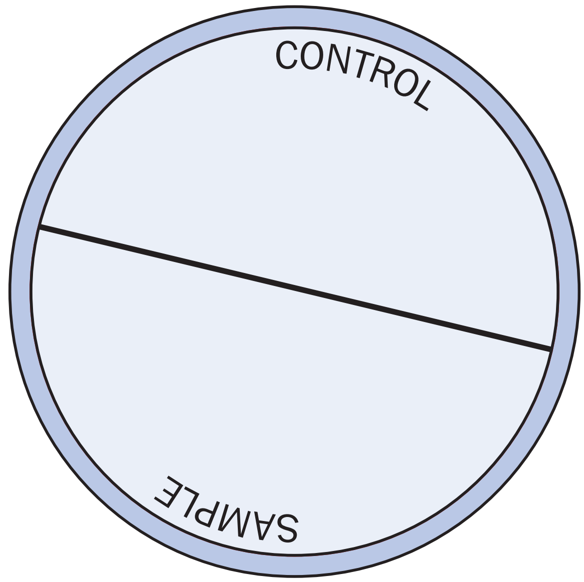
- safety goggles and gloves
- sterile petri dish with tryptic soy agar
- sterile cotton swab
- sterile water (or boiled)
- parafilm or masking tape
- incubator chamber (35°C or 95°F)
- food materials or contaminated surfaces for use in collecting samples (examples: spilled egg white, moist kitchen sponge, milk that has set out at room temperature, bottom of a sink, drawer in a refrigerator–any kitchen surface.)
- felt-tip marker

Safety:

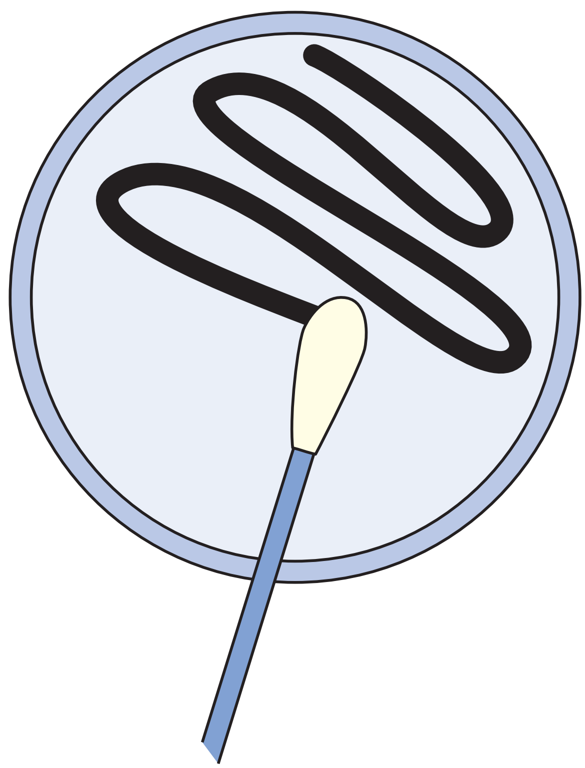
The substances and organisms used in this activity may be hazardous. Use precautions by wearing goggles and gloves. Properly dispose of wastes.

Procedures:

1. Put on safety equipment: goggles, gloves, and lab coat.
2. Obtain the materials needed for the making the cell culture.
3. Keep the petri dish with agar tightly covered until ready to inoculate the medium. (This is to prevent organisms that float in the air from contaminating your culture.)
4. Draw a line midway across the bottom of the petri dish to divide it into approximately equal halves. Label one side as "control;" label the other whatever it is you will use as the source of your sample that is cultured.



5. Position the petri dish close to the source of your sample.
6. Remove the wrapper from the sterile swab. Hold it by one end using care to keep the other end/tip sanitary. Quickly touch the substance or surface with a stroke of the swab once using a back-and-forth motion.
7. Open the petri dish just enough to get the end of the swab inside and quickly swab the half being cultured using two or three back-and-forth motions.



8. Quickly cover the petri dish. Use Parafilm around the edge or short pieces of masking tape on opposite sides to hold the cover in place. Do not block your view of the agar surface.
9. Place the inoculated petri dish upside down (label side up) in an incubator at 35°C (95°F). (If an incubator is not available, place in a warm location that approximates the desired temperature–maybe under a heat lamp. Caution: Avoid overheating and creating a fire.
10. Allow the dish to incubate 24–36 hours. (Longer may be needed if a cooler temperature is used.)

11. Remove the dish from the incubator and observe the presence of changes. Are there growths? Compare the “control” half with the “treatment” half. Use a hand lens. (Compare what you observe the drawings of bacteria in textbooks or on TM–Q.)
12. Prepare a slide for microscope examination.
13. Identify the organisms observed. Sketch what you see below on this page.

14. Write your overall assessment here: