**Study Plan: Episode 9—Microbial Control**

1. **Read the UNIT OVERVIEW presented in this Study Guide.**
2. **View the video “Microbial Control.”**
3. **Read UNIT OBJECTIVES and KEY CONCEPTS sections of this study guide.**
4. **View the video a second time, this time taking notes. Pay particular attention to topics identified by the UNIT OBJECTIVES or KEY CONCEPTS as significant.**
5. **Read Chapter Eleven, pages 216 – 236.**
6. **Review, as needed, related topics. A review of the genetic transmission of antibiotic resistance factors may provide insight into the special problems faced by those responsible for microbial control in hospitals.**
7. **Return to the Unit Objectives and Key Concepts listed in this Study Guide. Do you feel you have achieved each objective? Review sections of the text or video pertinent to material you don’t feel you have mastered.**
8. **Test your mastery of the material by answering the Review Questions at the end of this Study Guide.**
9. **Check your answers against the answer key; review material relating to any questions you missed.**

**Unit Overview**

Man’s relationships with microbes are for the most part beneficial; however, in certain circumstances the control of microbial growth is important. Microbial growth can be inhibited or controlled by a variety of chemical and physical means. Lethal heat and barriers that inhibit the transfer of microbes are important physical control agents. Chemical controls include sterilants, disinfectants and antiseptics. Where environmental factors favor the growth of microbes or their presence represents a particular threat, a combination of these methods may be employed. The level of control indicated is determined by a number of interacting factors, chief among them the likelihood and consequences of infection. Microbial control is of particular importance in food handling, both in the home and in industry, and in the very special environment of hospitals. Humans currently enjoy a competitive edge in the battle against harmful microbes; measures such as adequate supplies of safe drinking water, public health and sanitation measures are powerful weapons for disease prevention, and antibiotics can treat bacterial infections if they occur. However, strong preventative measures and access to antibiotics is not universal and microbes still cause preventable diseases in many countries.

Although in the U.S. we can prevent and treat infectious diseases better today than ever before, microbes hold powerful weapons as well. Our relationship with microbes is an "adaptation arms race." Each “advance” by one side (microbial or human) represents a new challenge. Each advance demands a response; each response in turn demands a further change. Each species adapts to changes in its environment, and each species has some effect on its environment. The example given earlier of the introduction of the myxoma virus to control rabbits is an excellent illustration of the adaptation arms race.

Microbial adaptation to our use of antibiotics threatens our current ability to control disease. The search for novel and effective antimicrobial agents is critical and existing products must be used wisely to retard the development of resistant mutants. The increased speed of international travel and commerce can quickly expose populations located very far apart from one another to each others’ diseases and microbes. For example, people in the United States can easily be exposed to diseases cultured in villages on the other side of the globe, and vice-versa. Strong public health measures implemented on a global scale, widespread education of how diseases are transmitted, and good hygiene remain our best weapons against the epidemic spread of pathogenic microbes.

**Objectives**

* Distinguish between and provide examples of both physical and chemicals methods of controlling microbes.
* Define and explain the differences between an antiseptic and a disinfectant. Distinguish from a sterilant.
* Describe several food preservation and safety techniques.
* List and explain the four levels of isolation used in a hospital.
* List and describe the primary causes of nosocomial acquired infections.
* Compare and contrast antibiotics with chemical control methods (when used, selectivity, time to work, target sites)
* Explain how microbes become resistant to antibiotics and what can done to reduce/prevent the development of antibiotic resistance.

**Key Terms**

* Aminoglycoside
* Antibiotic
* Antiseptic
* Arsphenamine
* Autoclave
* Bordeaux mixture
* Broad-spectrum antibiotic
* Cephalosporin
* Chemical control methods
* Desiccation
* Detergent
* Disinfectant
* Endospore
* Ionizing radiation
* Lyophilization
* Narrow spectrum antibiotic
* Pasteurization
* Penicillin
* Penicillinase
* Physical control methods
* Prontosil
* Sterilization
* *Streptomyces*
* Sulfanilamide
* Triclosan
* UV radiation

**Key Concepts**

**CONTROL MEASURE OVERVIEW**

* Microbes levels are controlled to prevent infection and disease.
* Both physical and chemical methods are used; what is used depends upon the situation and the level of risk.
* If an infection is not prevented, antibiotics can be used to treat bacterial infections (also antifungal, anti-virals, etc. if available)
* Sterilization is the destruction of all microbial life, including endospores (and the non-living viruses)
* Physical/chemical methods can damage (1) the cell wall or the cell membrane making the cell leaky, (2) the nucleic acids (DNA) so the cell cannot divide or make proteins or (3) destroy proteins (structural or enzymes) so the cell cannot function.
* In general physical and chemical control methods are fast-acting.
* Control of microorganisms is still a problem in some areas of the world. As many as 50,000 people die per day from diseases such as malaria, yellow fever, typhoid fever, tuberculosis, and cholera. This is due to:
  + poor hygiene
  + no source of clean water
  + lack of education on infectious disease
  + poor nutrition, which weakens the immune system

**PHYSICAL CONTROLS ON MICROORGANISMS**

* Heat kills microbes by denaturing proteins and diving off water that is needed for biological reactions; direct flame, hot air from ovens, and steam under pressure are the most common methods of applying heat. A protein is denatured when its shape is changed from its tertiary structure to secondary or primary structure. The proteins in eggs are visibly changed from their fluid state into a solid when they are cooked. The egg proteins are denatured by this change.
* Ultraviolet (UV) light damages DNA directly (be creating thymine dimers), killing microbes.
* Ionizing radiation kills cells indirectly; it ionizes molecules in the cell, creating ions that destroy the cell. X-rays and gamma rays are examples of ionizing radiation (Other sources will indicate that ionizing radiation can cause breaks in the DNA, which leads to the death of the cell).
* Filtration physically removes microbes from an aqueous solution.
* Drying controls microbes by removing water from the cell. Water is necessary to carry out reactions inside the cell.
* Increasing osmotic strength by adding salt or sugar (solutes) causes water to diffuse out of cells, effectively causing death by drying! Osmosis is the movement of water across a cell membrane from regions of high solute concentration to regions of low solute concentration. Here’s an example: When you make pickles, you place the cucumbers into a strong salt solution called brine. There’s more water inside the cells of the cucumber than there is outside, so water diffuses out into the brine. The cucumber is dehydrated, and, when stored in the brine, there are few bacteria that can tolerate such conditions.
* Cold is a microbiostatic measure; it **slows** microbial growth.
* Physical barriers, such as the gowns and glove worn in hospitals or sealed containers, control microbes by preventing their spread.

**CHEMICAL CONTROLS ON MICROORGANISMS**

* *Disinfectant* is the term used for germicides (germicides kill microorganisms) used on inanimate objects.
* *Antiseptic* is the term used for germicides applied to living tissue.
* Germicides are classified as having high, intermediate, or low germicidal activity. FYI:
  + Their effectiveness is defined by a ratio that compares the dilution that will kill in 10 minutes (but not 5 minutes) with that of phenol. Phenol was used in old formulations of Lysol®.
  + The effectiveness of a germicide against a specific organism is defined by measuring the zone of growth inhibition that forms around a disc saturated with the agent.
* The ideal chemical agent would have the following properties:
  + Ability to kill all microbes and their spores
  + Soluble in water
  + Stable on standing (while not in use)
  + Antimicrobial activity would not diminish over time
  + Non-toxic to humans and animals
  + Uniform in composition so that all ingredients are present at each application
  + Would not combine with organic matter other than microbes
  + Toxicity would be highest at room temperature or body temperature
  + Effectively penetrate surfaces
  + Would not damage metals (corrode or cause rust) or fabrics (stain or disintegrate)
  + Readily available in sufficient quantities and at reasonable cost
* Examples of chemical agents:
  + Phenols and phenolics denature proteins; phenolics also act on lipids. The throat spray called Chloroseptic® is phenol-based.
  + Alcohols denature proteins and disrupt the cytoplasmic membrane. This is why hand sanitizers are effective.
  + Halogens such as bromine, chorine, and iodine deactivate enzymes by oxidizing "functional" groups. Iodine is an antiseptic; chlorine and bromine are disinfectants (i.e. in bleach or bromine tablets for spas).
  + Hydrogen peroxide is an oxidizing agent; it is a weak antiseptic used to clean wounds and as a mouthwash. It is also used to disinfect fragile medical instruments and contact lenses.
  + Salts of heavy metals react with sulfhydryl groups to denature proteins. Merthiolate and Mercurochrome are skin disinfectants that contain mercury.
  + *Surfactants* are compounds with hydrophilic (water-loving) and hydrophobic (water-hating) parts that penetrate oily substances in water and form an emulsion. Soaps and detergents are surfactants that control microorganisms by washing them away.
  + Quaternary ammonium salts are powerful surfactant germicides. They kill all classes of cellular microorganisms and viruses that have membranes.
  + Alkylating agents attach short carbon chains to proteins. Formaldehyde, formalin, and glutaraldehyde are examples. Most of these substances also can attach short carbon chains to DNA, and are proven or suspected carcinogens (cancer-causing).
  + Ethylene oxide is a gas used to sterilize heat-sensitive materials and unwieldy objects. It is extremely toxic to humans and explosive in the pure state.

**PRESERVING FOOD**

* Temperature is the environmental factor most often used to preserve food.
* Refrigeration inhibits the growth of most microorganisms (i.e., bacteriostatic).
* Canning — steam under pressure — is an effective method of food preservation that is considered to be the oldest and most reliable method used. Two factors, time and temperature, determine safe heat treatments for canning. The environment inside a pressure canner must reach 121°C (destroys endospores such as those of *Clostridium botulinum* that causes botulism). Canned goods are essentially sterile and they are sealed to prevent contamination. The reason the quality of canned goods is often less desirable than fresh foods is that the process used is designed to kill 1012 (1,000,000,000,000) *Clostridium* spores – that’s a lot!
* Pasteurization is a heat treatment that kills pathogens in milk and lowers spoilage microorganism numbers, but does not sterilize foods. It is used to promote safety and maintain high food quality. This method is used to destroy pathogens such as *Escherichia*, *Brucella*, *Mycobacterium*. *Campylobacter*, *Salmonella*, *Listeria*, *Staphylococcus*, *Coxiella*, *Streptococcus*, *Yersinia*, and others that might be found in the milk.
  + Holding method is a pasteurization technique that utilizes lower temperature (62.9o C) for 30 minutes. In high-throughput situations this is not practical.
  + High temperature short time (HTST) pasteurization (a.k.a. Flash method) is a pasteurization method that utilizes high temperature (71.6oC) and 15 seconds to 30 seconds.
  + Ultra-high flash method pasteurization utilizes higher temperatures (82oC) for 3 seconds.
* Low pH prevents the growth of most microorganisms. Adding vinegar is one method of lowering pH. *Clostridium botulinum* spores will not germinate below a pH of 4.6. If the organism does not germinate it will not produce its deadly toxin.
* Reducing “available water” controls microbial growth. Drying and salting have been used for centuries to preserve meats, fish, and fruits. High sugar content operates on the same principle; it is used to preserve fruits.

**HOSPITAL CONTROL MEASURES**

* Epidemiology is the study of the transmission of disease.
* Hospital epidemiologists study the transmission of nosocomial infections, establish, and monitor hospital infection control programs. Various levels of control are implemented within a hospital, including routine floor mopping to sterilization of surgical implements.
* Nosocomial (hospital acquired) infections are due to three factors:
  + patients are susceptible due to lowered immune systems,
  + microbes are prevalent in the hospital: more antibiotic-resistant strains and patients with infections are in the hospital
  + invasive procedures used in hospital provide access (i.e. catheters, incisions, IV, etc.)
* Four levels of isolation are used to control transmission to/from patients:
  + standard precautions (handwashing) to reduce the typically encountered microbes
  + contact precautions (add gloves and gowns) to reduce or eliminate microbes that are spread through direct contact
  + droplet precautions (add mask) are for microbes that are spread by rather large droplets (relatively speaking)
  + airborne precautions (add special masks and negative pressure rooms) are used for microbes that behave as though they can fly – measles, chickenpox, and tuberculosis; high efficiency filters are used fro this
  + Surgical asepsis (asepsis = absence of microbes) involves attire, aggressive handwashing, and sterile field.
  + If infections do result, antimicrobials are used.
  + Bacterial strains highly resistant to commonly used antibiotics and germicides are a consequence of the highly aggressive microbial growth control practices required in hospitals.

**DRUG RESISTANCE**

* A microorganism is drug-resistant if it can grow in the presence of a drug. Natural drug resistance is an intrinsic (or built-in) property of a microbial species such as being gram-negative vs. gram-positive.
* Microbial drug resistance can be acquired through genetic change (mutations) or by gene sharing.
* The three mechanisms of acquired drug resistance are:
  + The microorganism produces an enzyme that destroys the drug.
  + There is a change in the target the drug attacks.
  + The drug is unable to get into the cell (change in cell membrane permeability).
* Since the first antimicrobials were antibacterial; the term antibiotic refers to antibacterial drugs
* Narrow-spectrum antimicrobial drugs affect only a single microbial group; broad-spectrum drugs affect more than one microbial group. Broad spectrum can harm more of a person’s own normal flora.
* Acquired drug resistance can be slowed by limiting nonmedical uses of antibiotics (i.e., in farm animal food) and inappropriate uses (i.e., taking antibiotics for viral infections), using narrow spectrum when possible, and using as prescribed (don't stop just because you feel better because this allows the ones already somewhat resistant to thrive).

**ANTIMICROBIAL DRUGS**

* Antimicrobial drugs are more specific than chemical control agents and target specific sites, i.e., one enzymes vs. all proteins
* Since antimicrobial drugs are taken internally they need to target microbes and not our cells
  + the principle of selectivity
* Antibiotics take longer to work than chemical control methods since they are selective and we are putting them in our bodies.
* The search for new antimicrobials is constant. As microbes develop resistance to our antimicrobials we need to find new ones.
* Most antimicrobial available target bacteria. Few examples include:
  + The cell wall of bacteria is an appropriate target for antimicrobial drugs because animal cells lack a cell wall. Penicillins, cephalosporins, vancomycin, and bacitracin target cell wall synthesis.
  + Drugs targeting the cell membrane must be used with extreme caution; eukaryotic cells have membranes, too! Polymyxin B and amphotericin B are examples of drugs that target the cell membrane.
  + Bacterial ribosomes are a safe target for antibiotics because they differ from eukaryotic ribosomes. Examples are tetracycline and erythromycin.
* Sulfonamides (sulfa drugs) inhibit the synthesis of folic acid, which the bacteria need to make DNA. These agents are safe for clinical use because animals do not synthesize folic acid (they require it preformed - in the diet).
* There are few anti-viral and anti-fungal compounds because the viruses use or own cells so we have trouble targeting them. Likewise, the fungi are eukaryotic and share many similarities with us, which makes it difficult to treat.

**Review Questions**

**Multiple Choice**

1. Select the term that involves a treatment to destroy all microbial life.
2. antisepsis
3. decontamination
4. disinfection
5. sterilization
6. Moist heat treatment kills cells by
7. altering osmotic conditions.
8. blocking UV light.
9. dehydrating cells.
10. denaturing proteins.
11. A pressure canner normally maintains a temperature of degrees C at 15 pounds per square inch pressure.
12. 85
13. 100
14. 121
15. 155
16. Nosocomial Infectious diseases happen
17. because sick people have weakened immune systems and are most susceptible to disease
18. because not all staff follow infection control procedures
19. because a source of microbes exists because people with infectious diseases come to the hospital
20. because medical procedures provides access for microbes to enter patients bodies
21. all of the above
22. no more often than in the community
23. Penicillins are safe for clinical use because
24. Eukaryotic cells do not have a nucleus.
25. Eukaryotic cells do not have peptidoglycan cell walls.
26. Eukaryotic cells do not have ribosomes.
27. Eukaryotic cells do not have a cell membrane.

1. Which kind of bacteria can grow in a refrigerator?
2. barophile
3. mesophile
4. psychrophile
5. thermophile

**Fill In**

1. \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ is a treatment designed to destroy all microbial life.
2. \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ is a germicide prepared for use on living tissue.
3. \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ is a germicide prepared for use on inanimate objects and surfaces.

**True/False**

1. A hospital epidemiologist studies the transmission of nosocomial infections.
2. Penicillin effectively treats viral infections.
3. Broad-spectrum antibiotics can significantly alter the normal bacterial communities of the body.
4. There is a rising tide of antibiotic-resistant microbes.
5. Hand washing is a good thing, but of minimal importance in the transmission of disease.
6. Pasteurization is a process used to sterilize milk.
7. Fungal infections are difficult to treat because fungi are eukaryotic cells.
8. Moist heat or steam under pressure is only used in hospitals.
9. Salting meats is an effective means of protecting them from degradation by microbes.

**Discussion Questions**

1. Roman army surgeons dressed wounds with honey. What beneficial effect do you think this would have and why?

2. Why does soap and water continue to be an effective antimicrobial agent? Why don’t we see “soap resistant” microbes?

**Answers**

**Multiple Choice**

1. D 2. D 3. C 4. E 5. B 6. C

**Fill In**

1. Sterilization 2. Antiseptic 3. Disinfectant

**True/False**

* 1. T 2. F 3. T 4. T 5. F 6. F. 7. T 8. F 9. T

**Discussion**

* 1. The high osmotic pressure of the honey killed microorganisms, protecting the wounds against infection. High osmotic pressure reduces available water and kills microbes by “drying” them!
  2. Soap does not kill microbes — it simply rinses them away. Because microbes are not killed, microbial populations are not placed under the heavy selective pressure that contributes to the evolution of antibiotic resistant microbes.