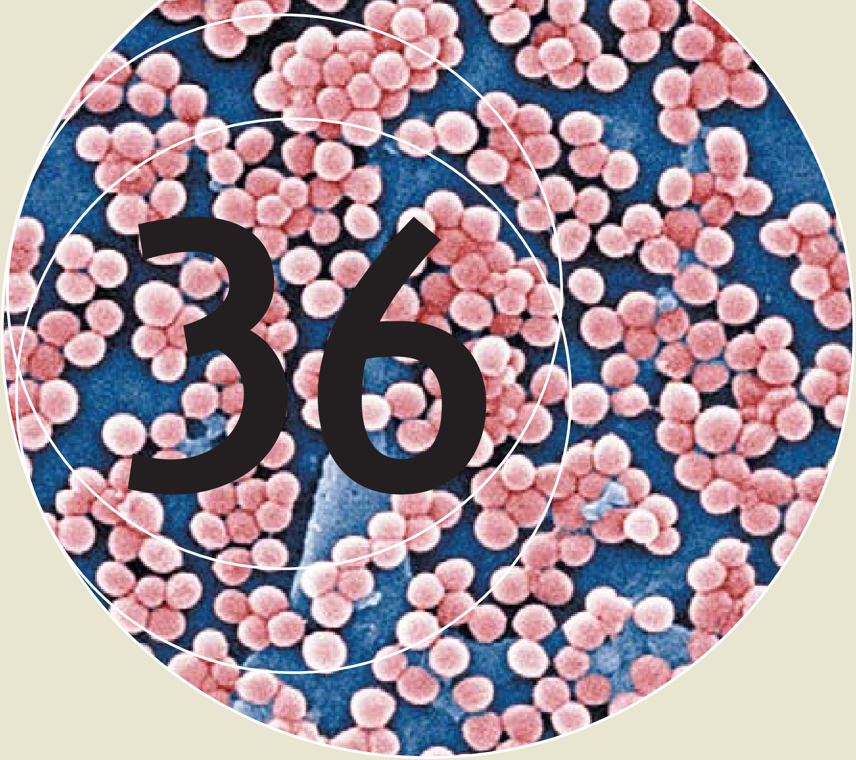

36

Food Preservation and Foodborne Microbial Diseases

Cells of the bacterium *Staphylococcus aureus* produce a toxin that causes severe intestinal distress. “Staph” food poisoning is a classic and common food-borne illness and is typically linked to contaminated foods left under conditions that allow for rapid growth of the organism.

Humans are constantly exposed to bacteria, fungi, and viruses in food as well as in air and water. The foods we eat, whether they are fresh, prepared, or even preserved, are seldom sterile and may be contaminated with spoilage microorganisms or occasionally with pathogens. On the other hand, microbial activity is important for the production of some foods. For example, cheese, buttermilk, sour cream, and yogurt are all produced by microbial fermentation. Sauerkraut is a fermented vegetable food. Certain sausages, pâtés, and liver spreads are produced by microbial fermentation. Cider vinegar is produced by the activities of the acetic acid bacteria, and alcoholic beverages are produced by fermentation. Some foods contain living microorganisms thought to confer health benefits. We discussed these foods, called probiotic foods, in the context of replacing or augmenting the normal microbial flora in the human gut (see the Chapter 27, Microbial Sidebar, "Probiotics").

Here we examine food preservation methods that limit unwanted microbial growth and food spoilage. We also look at microbial processes that aid in food preservation and, not incidentally, create a variety of fermented foods. Finally, we discuss microbial products and microorganisms that cause food poisoning and food infection.

I Food Preservation and Microbial Growth

Microorganisms are important spoilage agents in foods, causing food shortages and economic loss. Various methods, some utilizing desirable microbial growth, are used for controlling spoilage organisms.

36.1 Microbial Growth and Food Spoilage

Microorganisms, including a few human pathogens, colonize and grow on common foods. Foods provide a suitable medium for the growth of various microorganisms, and microbial growth often reduces food quality and availability.

Food Spoilage

Food spoilage is any change in the appearance, smell, or taste of a food product that makes it unacceptable to the consumer. Spoiled food may still be safe to eat, but is generally regarded as unpalatable and will not be purchased or readily consumed. Food spoilage causes losses to producers, distributors, and consumers in the form of reduced quality and quantity, and inevitably leads to higher prices.

Foods consist of organic materials that can be nutrients for the growth of chemoorganotrophic bacteria. The physical and chemical characteristics of the food determine its degree of susceptibility to microbial activity. With respect to spoilage, foods are classified into three major categories: (1) **perishable food**, including many fresh food items; (2) **semiperishable food**, such as potatoes and nuts; and (3) **stable** or **nonperishable food**, such as flour and sugar (**Table 36.1**).

Table 36.1 *Food classification by storage potential*

Food classification	*Examples*
Perishable	Meats, fish, poultry, eggs, milk, most fruits and vegetables
Semiperishable	Potatoes, some apples, and nuts
Nonperishable	Sugar, flour, rice, and dry beans

The three food categories differ greatly with regard to their *moisture content*, which is related to **water activity, a_w** (Section 5.16). Water activity is a measure of the availability of water for use in metabolic processes. Nonperishable foods have low water activity and can generally be stored for considerable lengths of time without spoilage. Perishable and semiperishable foods, by contrast, typically have higher water activities. Thus, these foods must be stored under conditions that inhibit microbial growth.

Fresh foods are spoiled by many different bacteria and fungi. The chemical properties of foods vary widely, and each food is characterized by the nutrients it contains as well as other factors such as acidity or alkalinity. As a result, each fresh food is typically colonized and spoiled by a relatively restricted group of microorganisms; the spoilage organisms are those that can gain access to the food and use the available nutrients (**Table 36.2**).

For example, enteric bacteria such as *Salmonella, Shigella*, and *Escherichia*, all potential pathogens sometimes found in the gut of animals, often contaminate meat. At slaughter, intestinal contents containing live bacteria may be accidentally spilled during removal of the intestines and result in contamination of the carcass. These organisms can also contaminate produce through fecal contamination of water supplies. Likewise, lactic acid bacteria, the most common microorganisms in dairy products, are the major spoilers of milk and milk products. *Pseudomonas* species are found in both soil and animals and cause the spoilage of fresh foods of all types.

Growth of Microorganisms in Foods

Microbial growth in foods follows the normal pattern for bacterial growth (Section 5.7). The length of the lag phase depends on the properties of the contaminating microorganism and the food substrate. The time required for the population density to reach a significant level in a given food product depends on both the size of the initial inoculum and the rate of growth during the exponential phase. The rate of growth during the exponential phase depends on the temperature, the nutrient value of the food, and the suitability of other growth conditions.

Throughout much of the exponential growth phase, microbial numbers in a food product may be so low that no measurable effect can be observed, with only the last few population doublings leading to observable spoilage. Thus, for much of the period of microbial growth in a food there is no visible or easily detectable change in food quality; spoilage is usually observed only when the microbial population density is high.

MiniQuiz

- List the major food groups as categorized by water availability.
- Identify factors that lead to growth of microorganisms in food.

Table 36.2 *Microbial spoilage of fresh food*[a]

Food product	*Type of microorganism*	*Common spoilage organisms, by genus*
Fruits and vegetables	Bacteria	*Erwinia*, *Pseudomonas, Corynebacterium* (mainly vegetable pathogens; rarely spoil fruit)
	Fungi	*Aspergillus, Botrytis, Geotrichum, Rhizopus, Penicillium, Cladosporium, Alternaria, Phytophthora*, various yeasts
Fresh meat, poultry, eggs, and seafood	Bacteria	*Acinetobacter, Aeromonas*, *Pseudomonas*, *Micrococcus, Achromobacter, Flavobacterium, Proteus, Salmonella, Escherichia, Campylobacter, Listeria*
	Fungi	*Cladosporium, Mucor, Rhizopus, Penicillium, Geotrichum, Sporotrichum, Candida, Torula, Rhodotorula*
Milk	Bacteria	*Streptococcus, Leuconostoc, Lactococcus, Lactobacillus, Pseudomonas, Proteus*
High-sugar foods	Bacteria	*Clostridium, Bacillus, Flavobacterium*
	Fungi	*Saccharomyces, Torula, Penicillium*

[a]The organisms listed are the most commonly observed spoilage agents of fresh, perishable foods. Many of these genera include species that are human pathogens.

36.2 Food Preservation

Food storage and preservation methods slow the growth of microorganisms that spoil food and cause foodborne disease.

Cold

A crucial factor affecting microbial growth is temperature (Section 5.12). In general, a lower storage temperature results in less microbial growth and slower spoilage. However, a number of psychrotolerant (cold-tolerant) microorganisms can grow, albeit slowly, at refrigerator temperatures (3–5°C). Therefore, storage of perishable food products for long periods of time (more than several days) is possible only at temperatures below freezing. Freezing and subsequent thawing, however, alter the physical structure, taste, and appearance of many foods such as leafy green vegetables like spinach and lettuce, making them unacceptable to the consumer. Freezing is widely used, however, for the preservation of solid foods such as meats and many fruits and vegetables. Freezers providing a temperature of −20°C are most commonly used. Storage for weeks or months is possible at −20°C, but microorganisms can still grow in pockets of liquid water trapped within the frozen food. For long-term storage, temperatures of −80°C [the temperature of solid carbon dioxide (CO_2), "dry ice"] are necessary. Because of the high equipment and energy costs necessary to maintain such low temperatures, ultracold freezing is not used for routine food storage.

Pickling and Acidity

Another factor affecting microbial growth in food is pH. Foods vary somewhat in pH, but most are neutral or acidic. Microorganisms differ in their ability to grow under acidic conditions, but conditions of pH 5 or less inhibit the growth of most spoilage organisms. Therefore, weak acids are often used for food preservation, a process called **pickling**. Vinegar, a dilute acetic acid fermentation product of the acetic acid bacteria, is usually added in the pickling process. Pickling methods usually mix the vinegar with large amounts of salt or sugar to decrease water availability (as discussed below) and further inhibit microbial growth. Common pickled foods include cucumbers (sweet, sour, and dill pickles), peppers, meats, fish, and fruits.

Drying and Dehydration

As we mentioned, water activity, or a_w, is a measure of the availability of water for use by microorganisms in metabolic processes. The a_w of pure water is 1.00; the molecules in pure water are loosely ordered and rearrange freely. When solute is added, the a_w decreases. As water molecules reorder around the solute, the free rearrangement of the solute-bound water molecules becomes energetically unfavorable. The microbial cells must then compete with solute for the reduced amount of free water. In general, bacteria are poor competitors for the remaining free water, but fungi are good competitors. In practice this means that high concentrations of solutes such as sugars or salts, which greatly reduce a_w, typically inhibit bacterial growth. For example, most bacteria are inhibited by a concentration of 7.5% sodium chloride (NaCl) (a_w of 0.957), with the exception of some gram-positive cocci, such as *Staphylococcus*. On the other hand, molds compete well for free water under conditions of low a_w and often grow well in high-sugar foods such as syrups.

Some commercially important foods are preserved by the addition of salt or sugar. Sugar is used mainly in fruits (jams, jellies, and preserves). Salted products are primarily meats and fish. Sausage and ham are preserved using various curing salts, including NaCl. Some meats also undergo a smoking process. Preserved meat products vary widely in a_w, depending on how much salt is added and how much the meat has been dried. Some cured meat products such as country ham or jerky can be kept at room temperature for extended periods of time. Others with higher a_w require refrigeration for long-term storage.

Microbial growth in foods can also be controlled by drying, which lowers water content and availability. Drying is used to preserve highly perishable foods such as meat, fish, milk, vegetables, fruit, and eggs. The least damaging physical method used to

Figure 36.1 **Spray dryer.** Industrial spray dryers are used to dry or concentrate large volumes of high-value liquid foods.

dry foods is the process of **lyophilization (freeze-drying)** in which foods are frozen and water is then removed under vacuum. This method is very expensive, however, and is used mainly for specialized applications such as preparation of military rations that may need to be stored for long periods even under wet or warm conditions.

Spray drying is the process of spraying, or atomizing, liquids such as milk in a heated atmosphere. The atomization produces small droplets, increasing the surface-to-volume ratio of the liquid, promoting rapid drying without destroying the food. This technology is widely used in the production of powdered milk, certain concentrated liquid dairy products such as evaporated milk, and concentrated food ingredients such as liquid flavorings (**Figure 36.1**).

Heating

Heat is used to reduce the bacterial load or even sterilize a food product; it is especially useful for the preservation of liquids and wet foods. *Pasteurization*, a process in which liquids are heated to a specified temperature for a precise time, was described in Section 26.1. Pasteurization does not sterilize liquids, but reduces the bacterial load of spoilage organisms and pathogens, significantly extending the shelf life of the liquid. Pasteurization can be done at 63°C (145°F) for 30 seconds or at 71°C (160°F) for 15 seconds. Typically, perishable liquids such as milk, fruit juices, and beer are pasteurized. Ultrahigh-temperature (UHT) processing, sometimes called ultrapasteurization, can be used to preserve the same liquids. UHT processing heats the liquid to 138°C (238°F) for 2 to 4 seconds. This treatment kills all microorganisms, extending the shelf life of liquids like milk to 6 months or longer without the need for refrigeration. UHT-processed milk is common in Europe, but is not easily found in the United States.

Canning is a process in which food is sealed in a container such as a can or glass jar and then heated. In theory, canning should sterilize the food product, but this requires processing at the correct temperature for the correct length of time. However, when properly sealed and heated, most canned food should remain stable and unspoiled indefinitely at any temperature.

The temperature–time relationships for canning depend on the type of food, its pH, the size of the container, and the consistency or density of the food. Because heat must completely penetrate the food within the container, effective heating times must be longer for large containers or very dense foods. Acidic foods can often be canned effectively by heating just to boiling, 100°C, whereas nonacidic foods must be heated to autoclave temperatures (121°C). For some foods in large containers, times of 20–50 minutes must be used. Heating times long enough to guarantee absolute sterility of every container would make most foods unpalatable and could also reduce nutritional value. Even properly canned foods, therefore, may not be sterile. The process used for commercial canning, called *retort canning*, employs equipment similar to an autoclave to apply steam under pressure (Section 26.1).

If live microorganisms remain in a can, growth of organisms can produce extensive amounts of gas and build pressure, resulting in bulges or, in extreme cases, explosion (**Figure 36.2**). The environment inside a can is anoxic, and some of the anaerobic bacteria that grow in canned foods are toxin producers of the genus *Clostridium* (Section 36.7). Food from a bulging can, therefore, should never be eaten. On the other hand, the lack of obvious gas production is not an absolute guarantee that canned food is safe to consume.

Aseptic Food Processing

Several foods in the United States and many more in Asia and Europe are now prepared and packaged under aseptic conditions. Foods processed and packaged aseptically can be stored at room temperature for months or longer without spoilage. Aseptic processing uses *flash heating*, a process using a rapid, short heating cycle, or sterilization by cooking. The processed foods are then packaged in aseptic containers, usually cardboard cartons lined with foil and plastic. The process may require "clean room" conditions similar to those in a hospital operating room. For example, incoming room air is filtered to limit contamination from spores and bacteria in the atmosphere. Special equipment is required to flash-heat and deliver the product aseptically into sterile packaging materials.

In the United States, fruit juices (juice boxes) and milk substitutes are often processed in this way. In many European countries, milk products are flash-heated to 133°C and packaged aseptically. Perishable food products prepared aseptically can be stored at room temperature for at least 6 months. This developing technology significantly increases product shelf life and eliminates the need for refrigeration for many products. However, the equipment and processing plants necessary for aseptic food processing are expensive.

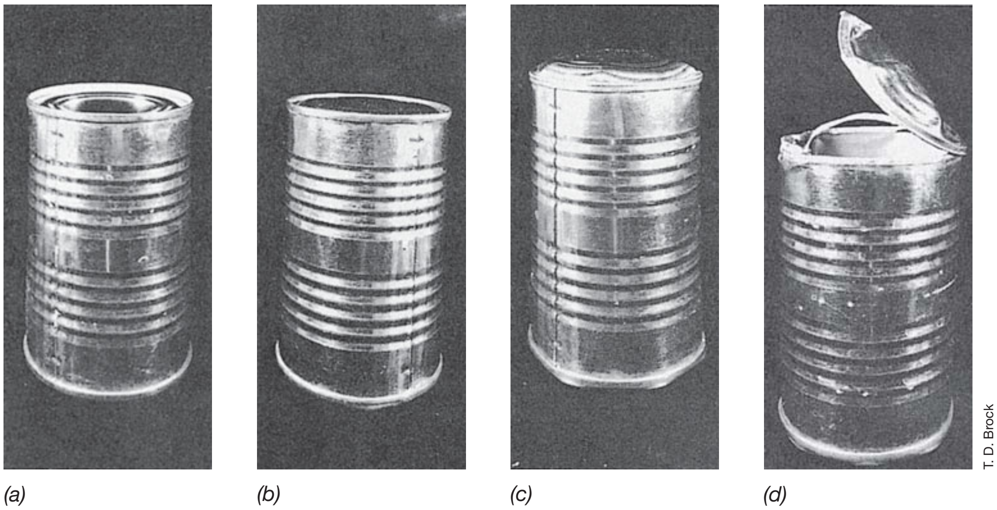

Figure 36.2 Changes in sealed tin cans as a result of microbial spoilage. *(a)* A normal can. The top of the can is pulled in slightly due to the negative pressure (vacuum) inside. *(b)* Swelling resulting from minimal gas production. The top of the can bulges slightly. *(c)* Severe swelling due to extensive gas production. *(d)* The can shown in part c was dropped, and the gas pressure resulted in a violent explosion, tearing the lid apart.

High-Pressure Processing

High-pressure processing (HPP) is a technology that uses very high hydrostatic pressure (up to 100,000 lb/in^2) to kill most pathogens and spoilage organisms in packaged foods. Applications include several food types. Fruits and vegetables such as avocado products, salsas, chopped onions, applesauce, ready-to-eat meats, and juices can all be processed in bulk or in consumer packaging. The packaged foods are loaded into a vessel that is flooded with water and placed under pressure (**Figure 36.3**). Pressure treatment kills most foodborne pathogens, but does not kill endospores; the products are not absolutely sterile, but shelf life is increased from days to months.

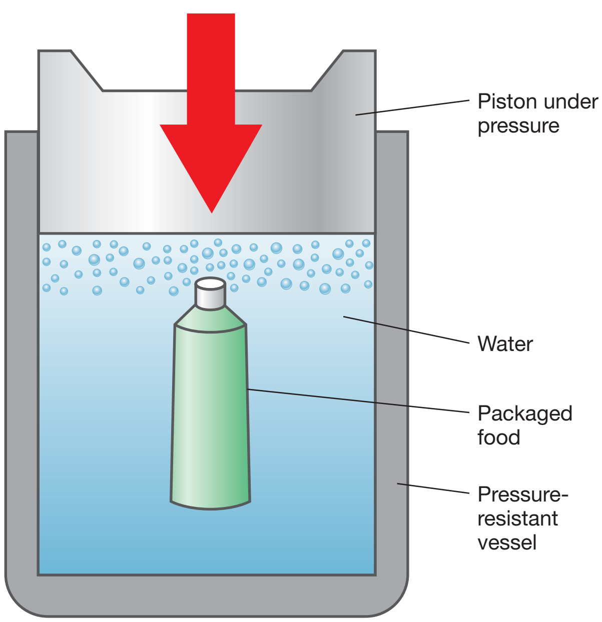

Figure 36.3 High-pressure processing (HPP) of food. Packaged foods are loaded into a vessel that is flooded with water and placed under very high hydrostatic pressure (up to 100,000 lb/in^2) to kill most pathogens and spoilage organisms in packaged foods.

Chemical Preservation

Over 3000 different compounds are used as food additives. These chemical additives are classified by the United States Food and Drug Administration as "generally recognized as safe" (GRAS) and find wide application in the food industry for enhancing or preserving texture, color, freshness, or flavor. A small number of these compounds are used to control microbial growth in food (**Table 36.3**). Many of these microbial growth inhibitors, such as sodium propionate and sodium benzoate, have been used for many years with no evidence of human toxicity. Others, such as nitrites (a carcinogen precursor) and ethylene oxide and propylene oxide (mutagens), are more controversial food additives because these compounds may adversely affect human health. The use of spoilage-retarding additives, however, significantly extends the useful shelf life of finished foods. Chemical food additives contribute significantly to an increase in quantity and in the perceived quality of available food items.

Table 36.3 *Chemical food preservatives*

Chemical	*Food*
Sodium or calcium propionate	Bread
Sodium benzoate	Carbonated beverages, fruit, fruit juices, pickles, margarine, preserves
Sorbic acid	Citrus products, cheese, pickles, salads
Sulfur dioxide, sulfites, bisulfites	Dried fruits and vegetables, wine
Formaldehyde (from food-smoking process)	Meat, fish
Ethylene and propylene oxides	Spices, dried fruits, nuts
Sodium nitrite	Smoked ham, bacon

Because of the time and cost required for testing of any chemical proposed as a food preservative or additive, it is unlikely that many new chemicals will be added to the list of safe and approved chemical food preservatives listed in Table 36.3.

Irradiation

Irradiation of food with ionizing radiation is an effective method for reducing contamination by bacteria, fungi, and even insects (Section 26.2). **Table 36.4** lists foods for which radiation treatment has been approved in the United States. Foods including herbs, spices, and grains are routinely irradiated. Fresh meats and fish can be irradiated to limit contamination by *Escherichia coli* O157:H7 (ground beef), *Campylobacter jejuni* (poultry), and *Vibrio* spp. (seafood). In an attempt to limit foodborne disease outbreaks in fresh produce, irradiation was approved in 2008 to control foodborne pathogens in iceberg lettuce and spinach. In many countries throughout the world, spices, seafood, vegetables, grains, potatoes, sterilized meals, and meats are irradiated.

For food irradiation, gamma rays generated from radioactive cobalt (^{60}Co) or cesium (^{136}Cs), or from high-energy electrons produced by linear accelerators, are used as radiation sources. Alternatively, beta rays can be generated from an electron gun, analogous to but significantly more powerful than the electron beam generated by the cathode ray gun formerly used in television sets. In addition, X-rays can be generated with electron beams focused on metal foil. X-rays have much greater penetrating power than beta rays and are therefore useful for treating large-volume food preparations. Beta ray and X-ray sources can be switched on and off at will and do not require a radioactive source.

Irradiated food products receive a controlled radiation dose. This dose varies considerably for each food category and purpose. For example, a dose of 44 kilograys (kGy) is used to sterilize meat products used on United States NASA space flights and is nearly ten times higher than the dose of 4.5 kGy used for control of pathogens in ground beef (Table 36.4). In the United States, a consumer product information label and the radura, the international symbol for radiation, must be affixed to foods that are irradiated in whole (**Figure 36.4**). Irradiated ingredients that are a major portion of a food product must be identified in the ingredients list, but the radura symbol does not need to be shown. Irradiated ingredients such as spices that are minor components of a finished food product do not have to be identified as irradiated.

Figure 36.4 The radura, the international symbol for radiation. Packaging of foods treated with radiation must be labeled with the radura, the international symbol for radiation, as well as the statement "treated by irradiation" or "treated with radiation."

Table 36.4 *Irradiated foods by category, dose, and purpose*

Food category	*Dose[a] (kGy)*	*Purpose*
Fresh meat: ground beef	4.50	Reduce bacterial pathogens
Herbs, spices, enzymes, and flavorings	30.00	Sterilize
Pork	1.00	Reduce *Trichinella spiralis* protist
Meats used in NASA[b] space flight program	44.0	Sterilize
Poultry	3.00	Reduce bacterial pathogens
Wheat flour	0.50	Inhibit mold
White flour	0.15	Inhibit mold

[a]The highest value for recommended doses is given. One kGy (kilogray) is 1000 grays. One gray, an SI unit, is 1 joule of radiation absorbed by 1 kilogram of matter and is also equivalent to 100 rad. For the radiation requirements necessary to kill specific microorganisms, refer to Table 26.1.
[b]National Aeronautics and Space Administration, USA.

MiniQuiz

- Identify food spoilage microorganisms that are also pathogens.
- Identify physical and chemical methods used for food preservation. How does each method limit growth of microorganisms?

36.3 Fermented Foods and Mushrooms

Many common foods and beverages are preserved, produced, or enhanced through the direct actions of microorganisms. Desirable microbial processes can produce significant alterations in raw foods; the product is called a *fermented food*. **Fermentation** is the anaerobic catabolism of organic compounds, generally carbohydrates, in the absence of an external electron acceptor (Section 4.8). Bacteria important in the fermented foods industry are the lactic acid bacteria, the acetic acid bacteria, and the propionic acid bacteria (**Table 36.5**). These bacteria do not grow below about pH 3.5, so food fermentation is a self-limiting process.

One of the most common fermented foods is yeast bread, in which the fermentation of simple sugars and grain carbohydrates by the yeast *Saccharomyces cerevisiae* (Table 36.5) produces

Table 36.5 *Fermented foods and fermentation microorganisms*[a]

Food category	*Primary fermenting microorganism*
Dairy foods	
Cheeses	*Lactococcus*
	Lactobacillus
	Streptococcus thermophilus
Fermented milk products	
Buttermilk	*Lactococcus*
Sour cream	*Lactococcus*
Yogurt	*Lactobacillus*
	Streptococcus thermophilus
Alcoholic beverages	*Zymomonas*
	Saccharomyces[b]
Yeast breads	*Saccharomyces cerevisiae*[c]
Meat products	
Dry sausages (pepperoni, salami) and semidry sausages (summer sausage, bologna)	*Pediococcus*
	Lactobacillus
	Micrococcus
	Staphylococcus
Vegetables	
Cabbage (sauerkraut)	*Leuconostoc*
	Lactobacillus
Cucumbers (pickles)	Lactic acid bacteria
Soy sauce	*Aspergillus*[d]
	Tetragenococcus halophilus
	Yeasts

[a]Unless otherwise noted, these are all species of *Firmicutes* except for *Micrococcus*, which is a genus of *Actinobacteria*. *Zymomonas* is a genus of *Alphaproteobacteria*.
[b]Yeast. Various *Saccharomyces* species are used in alcohol fermentations.
[c]Baker's yeast.
[d]A mold.

carbon dioxide (CO_2), raising the bread and producing the holes in the finished loaf (**Figure 36.5**). Other *Saccharomyces*-fermented food products include wine, beer, and whiskey. Production of these beverages is often carried out at industrial scales (Sections 15.7 and 15.8).

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Figure 36.5 Fermented foods. Bread, sausage meats, cheeses, many dairy products, and fermented and pickled vegetables are food products that are produced or enhanced by fermentation reactions catalyzed by microorganisms.

Dairy Products

Fermented dairy products were originally developed to preserve milk, an economically important fresh food that normally undergoes rapid spoilage. Dairy products include cheese and other fermented milk products such as yogurt, buttermilk, and sour cream (Figure 36.5). Milk contains the disaccharide lactose. Lactose can be hydrolyzed by the enzyme lactase into glucose and galactose. These monosaccharides are fermented to the final product of lactic acid by the lactic acid bacteria (Table 36.5). This fermentation reaction produces a significant decrease in pH from the neutral or slightly basic pH of raw milk to a pH of less than 5.3 in cheeses and less than 4.6 in other fermented milk products.

Starter cultures of lactic acid bacteria are introduced into raw milk, and fermentation proceeds for a time depending on the desired product. For some cheeses, a second inoculum may be introduced to produce a second fermentation. For example, following lactic acid fermentation, Swiss-style (Emmentaler) cheeses are reinoculated with *Propionibacterium*. The secondary fermentation catabolizes lactic acid to propionic acid, acetic acid, and CO_2. The CO_2 produces the large holes that characterize Swiss-style cheeses. Secondary fermentations with *Lactobacillus* and the mold *Penicillium roqueforti* produce the blue veins and distinctive taste and aroma of blue cheese. Each cheese type is produced under carefully controlled conditions. Time of fermentation, temperature, the extent of aging, and the types of fermenting microorganisms are rigidly controlled to ensure a distinctive and reproducible product.

Meat Products

Fermented meat products fall into several categories. *Sausages* are generally made from pork, beef, or poultry. The most common are the dry sausages, such as salami and pepperoni, and the semidry sausages such as bolognas and summer sausages (Figure 36.5).

Sausages are made using a uniformly blended mixture of meat, salt, and seasonings. A starter culture of lactic acid bacteria is added, and fermentation reduces the pH of the mixture to below 5. After fermentation, sausages are often smoked and dried to a moisture content of about 30%. Dry sausages can be held at room temperature for extended periods of time. Semidry sausages such as summer sausage have a final moisture content of about 50% and are less resistant to spoilage, so they are generally refrigerated.

Fish, often mixed with rice, shrimp, and spices, are also fermented to make fish pastes and fish-flavored products.

Vegetables and Vegetable Products

The variety of specialty fermented vegetable foods is practically endless. The most economically important fermented vegetable foods are sauerkraut (fermented cabbage) and some types of pickles (fermented cucumbers). Peppers, olives, onions, tomatoes, and many fruits are also fermented.

Vegetables are often fermented in salt brine to enhance preservation and flavor. The salt also helps prevent the growth of unwanted organisms, the desired fermentative organisms being salt-tolerant. Fermentation may also improve digestibility by breaking down plant tissues. For example, fermented legume products (peas, beans, and lentils) have a marked reduction in the flatulence-producing oligosaccharides that characterize fresh legumes.

Soy Sauce

Soy sauce is a complex fermentation product made by fermentation of soybeans and wheat. A culture of the fungus *Aspergillus* (Table 36.5) is spread on a cooked wheat–soybean mixture, where it grows for 2–3 days. This preparation, known as koji, is then mixed with brine [17–19% sodium chloride (NaCl)] and fermentation proceeds for 2–4 months or more in large vats (**Figure 36.6**). The *Aspergillus* and various microorganisms, including *Lactobacillus* and *Pediococcus* and several other fungi, produce fermentation products from the brined koji that contribute to the desirable characteristics of the final product. After fermentation, the liquid sauce is filtered, pasteurized, and bottled as soy sauce.

Vinegar

Vinegar is produced by the conversion of ethyl alcohol to acetic acid by the acetic acid bacteria. Key genera of acetic acid bacteria include *Acetobacter* and *Gluconobacter* (Section 17.8). Vinegar is produced from dilute ethanol solutions; the usual starting material is wine, fermented rice, or alcoholic apple juice (hard cider). Vinegar can also be produced from a mixture of pure alcohol in water, in which case it is called distilled vinegar, the term "distilled" referring to the alcohol from which the product is made rather than the vinegar itself. Vinegar is used as a flavoring agent in salads and other foods, and because of its acidity, it is also used in pickling. Foods pickled with high concentrations of vinegar can be stored unrefrigerated for years.

Figure 36.6 Soy sauce fermentation. The vats, each about 1 m deep and 4 m in diameter, contain koji, a mixture of wheat and soybeans inoculated with *Aspergillus* mixed in salt brine. Fermentation proceeds for up to 1 year in the vats. The liquid is then filtered, pasteurized, and bottled as soy sauce.

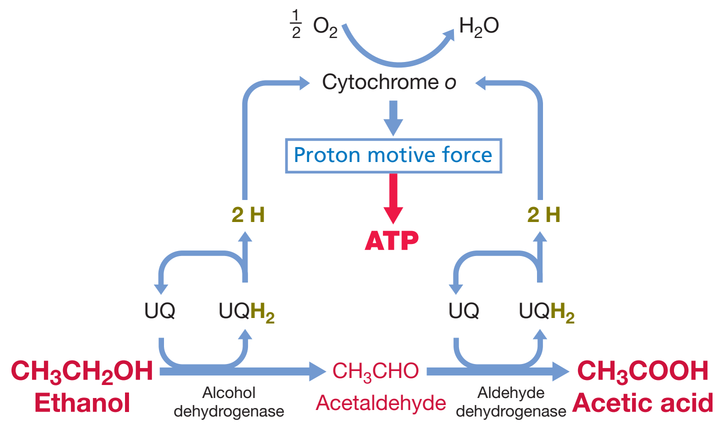

Figure 36.7 Vinegar production. The key process in vinegar production is the oxidation of ethanol to acetic acid. UQ, ubiquinone.

Acetic acid bacteria are strictly aerobic, but unlike most other aerobes, some species such as *Gluconobacter* do not oxidize organic electron donors completely to CO_2 and water (H_2O) (**Figure 36.7**). If ethyl alcohol is the electron donor, they oxidize it to acetic acid, which then accumulates in the medium. Acetic acid bacteria are acid-tolerant and are not killed by the low pH products that they generate. Because this process is aerobic, the demand for oxygen during growth is very high; the production of vinegar requires sufficient aeration of the medium.

Three processes are in use for vinegar production. The *open-vat method*, which is the original process, is still used in France where it was first developed. Wine is placed in shallow vats to facilitate exposure to the air. At the surface of the liquid, the acetic acid bacteria develop as a slimy layer. This process is not very efficient because the bacteria contact both the air and substrate only at the surface.

The second process is the *trickle method*. Alcoholic liquid is trickled over loosely packed beechwood twigs or shavings, arranged in a vat or column. The bacteria grow on the surface of the wood shavings, using the trickling liquid as a substrate. A stream of air enters at the bottom and passes upward, facilitating maximum contact between the bacteria, air, and substrate. The vat is called a *vinegar generator* (**Figure 36.8**), and the process is operated in a continuous fashion. The wood shavings in a vinegar generator are the support on which the bacteria grow to produce a biofilm, so they are not consumed and can last from 5 to 30 years, depending on the kind of alcoholic liquid used in the process.

Finally, the *bubble method* of vinegar production incorporates industrial incubation techniques to introduce and mix air into a fermenter containing the alcoholic substrate and inoculated with acetic acid bacteria. The bubble method is highly efficient; 90–98% of the alcohol is converted to acetic acid.

Acetic acid can easily be made chemically from alcohol, but the microbial product, *vinegar*, is a distinctive material. The flavor of vinegar is affected by other substances present in the starting material and produced during fermentation. For this reason, the microbial methods for vinegar production, especially the vinegar generator method, have not been supplanted by chemical processes.

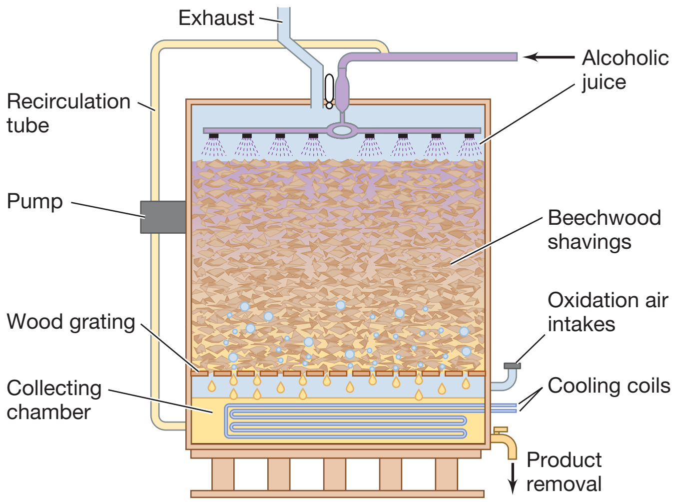

Figure 36.8 A vinegar generator. Alcoholic juice is trickled through wood shavings as air is passed upward. Acetic acid bacteria colonize the wood shavings, forming a biofilm that oxidizes alcohol to acetic acid. The dilute acetic acid pools in the collecting chamber, from where it is recycled through the generator. The pooled product is drained off when the acetic acid content reaches 4%, the minimum concentration to be labeled as *vinegar*.

Mushrooms

Several kinds of fungi are sources of human food, of which the most important are the mushrooms. Mushrooms are a group of filamentous fungi that form large, edible fruiting bodies (**Figure 36.9**). The fruiting body is called the mushroom and is formed through the association of a large number of individual fungal hyphae to form a mycelium.

The mushroom commercially grown in most parts of the world is the basidiomycete *Agaricus bisporus*, and it is generally cultivated in "mushroom farms." The organism is grown in special beds, usually in buildings where temperature and humidity are carefully controlled and exposure to light is severely limited. Beds are prepared by mixing soil with a material rich in organic matter, such as horse manure, and the beds are then inoculated with a pure culture of the mushroom fungus that has been grown in large bottles on an organic-rich medium.

In the mushroom bed, the mycelium grows and spreads through the substrate, and after several weeks it is ready for the next step, the induction of mushroom formation. This is accomplished by adding a layer of soil to the surface of the bed. The appearance of mushrooms on the surface of the bed is called a *flush* (Figure 36.9*a*), and for freshness the mushrooms must be collected immediately upon flushing. After collection they are packaged and kept cool until brought to market.

Another cultured mushroom is the shiitake, *Lentinus edulus*. Shiitake mushrooms are cellulose-digesting fungi that grow on hardwood trees. They are cultivated on small logs (Figure 36.9*b*). The logs are hydrated by soaking in water. Plugs of mushroom culture are inoculated into small holes drilled in the logs. The fungus grows through the log, and after about a year forms a flush of fruiting bodies, the edible mushroom. Shiitake mushrooms are considered to have a superior taste to *Agaricus bisporus*, and therefore are more expensive.

MiniQuiz

- Identify important dairy, meat, and vegetable food products that are produced or enhanced by microbial fermentation or growth.
- Identify the microbial group or groups that are most important for food fermentations.
- Identify edible fungi.

II Foodborne Disease, Microbial Sampling, and Epidemiology

If food is not decontaminated or preserved, pathogens may grow in it and cause foodborne diseases with significant morbidity and mortality. Like waterborne diseases, foodborne illnesses are common-source diseases. A single contaminated food source from

(a) (b)

Figure 36.9 Mushroom production. *(a)* A flush of the mushroom *Agaricus bisporus*. *(b)* The shiitake mushroom *Lentinus edulus*.

a food-processing plant or a restaurant may affect a large number of people. In 2010, chicken feed contaminated with *Salmonella* used at two egg production farms in Iowa infected eggs distributed nationally, and caused over 1500 infections. Each year in the United States, there are an estimated 25,000 foodborne disease outbreaks. As many as 76 million Americans are affected, an estimated 13 million acquire significant illnesses, 325,000 are hospitalized, and 5000 people die from foodborne diseases each year. Most outbreaks are due to improper food handling and preparation by consumers and affect small numbers of individuals, usually in the home. Occasional outbreaks affect large numbers of individuals because they are caused by breakdowns in safe food handling and preparation at food-processing and distribution plants. Most foodborne illnesses are unreported because the connection between food and illness is not made.

Foodborne illness is largely preventable; appropriate monitoring of food sources and disease outbreaks provides the basis for protecting consumers. The food industry and the government set standards and monitor food sources to control and prevent foodborne disease.

36.4 Foodborne Disease and Microbial Sampling

The most prevalent foodborne diseases in the United States are classified as *food poisonings* (*FP*) or *food infections* (*FI*); some diseases fall into both categories. **Table 36.6** lists the microorganisms that cause these diseases. Special microbial sampling techniques are necessary to isolate and identify the pathogens and toxins responsible for foodborne diseases, and a variety of growth-dependent, immunological, and molecular techniques are used. Foodborne illnesses and outbreaks are reported to the Centers for Disease Control and Prevention through PulseNet and FoodNet reporting systems.

Foodborne Diseases

Food poisoning, also called **food intoxication**, is disease that results from ingestion of foods containing preformed microbial toxins. The microorganisms that produced the toxins do not have to grow in the host and are often not alive at the time the contaminated food is consumed; the ingestion and action of a bioactive toxin causes the illness. We previously discussed some of these toxins, notably the exotoxin of *Clostridium botulinum*

Table 36.6 *Annual foodborne disease estimates for the United States*[a]

Organism	*Disease*[b]	*Number per year*	*Foods*
Bacteria			
Bacillus cereus	FP and FI	27,000	Rice and starchy foods, high-sugar foods, meats, gravies, pudding, dry milk
Campylobacter jejuni	FI	1,963,000	Poultry, dairy
Clostridium perfringens	FP and FI	248,000	Meat and vegetables held at improper storage temperature
Escherichia coli O157:H7	FI	63,000	Meat, especially ground meat, raw vegetables
Other enteropathogenic *Escherichia coli*	FI	110,000	Meat, especially ground meat, raw vegetables
Listeria monocytogenes	FI	2,500	Refrigerated "ready to eat" foods
Salmonella spp.	FI	1,340,000	Poultry, meat, dairy, eggs
Staphylococcus aureus	FP	185,000	Meat, desserts
Streptococcus spp.	FI	50,000	Dairy, meat
Yersinia enterocolitica	FI	87,000	Pork, milk
All other bacteria	FP and FI	102,000	
Total bacteria		***4,177,500***	
Protists			
Cryptosporidium parvum	FI	30,000	Raw and undercooked meat
Cyclospora cayetanensis	FI	16,000	Fresh produce
Giardia intestinalis	FI	200,000	Contaminated or infected meat
Toxoplasma gondii	FI	113,000	Raw and undercooked meat
Total protists		***359,000***	
Viruses			
Noroviruses	FI	9,200,000	Shellfish, many other foods
All other viruses	FI	82,000	
Total viruses		***9,282,000***	
Total annual foodborne diseases		***13,818,500***	

[a]Estimates are based on data provided by the Centers for Disease Control and Prevention, Atlanta, Georgia, USA, and are typical of recent years.
[b]FP, food poisoning: FI, food infection.

(Figure 27.22) and the superantigen toxins of *Staphylococcus* and *Streptococcus* (Section 28.10). **Food infection** is ingestion of food containing sufficient numbers of viable pathogens to cause infection and disease in the host. We discuss major foodborne infections in Sections 36.8–36.12.

Microbial Sampling for Foodborne Disease

Along with nonpathogenic microorganisms that cause spoilage, pathogenic microorganisms may be present in fresh foods. Rapid diagnostic methods that do not require pathogen growth or culture have been developed to detect important food pathogens such as *Escherichia coli* O157:H7, *Salmonella, Staphylococcus*, and *Clostridium botulinum*. Molecular and immunology-based tests are used to identify both toxin and pathogen contamination of foods and other products such as drugs and cosmetics. We discussed the use of nucleic acid probes and the polymerase chain reaction for the detection of specific pathogens, including foodborne pathogens, in Sections 31.12 and 31.13. The presence of a foodborne pathogen or toxin is not sufficient to link a particular food to a specific foodborne disease outbreak; the suspect pathogen or toxin must be isolated and identified to establish its role in a foodborne illness.

Isolation and growth of pathogens from nonliquid foods usually require preliminary treatment to suspend microorganisms embedded or entrapped within the food. A standard method uses a specialized blender called a *stomacher* (**Figure 36.10**). The stomacher processes a wide variety of solid and semisolid samples such as fresh and processed meat, dry fruits, cereals, grains, seeds, cheese, cosmetics, and for biomedical applications, pharmaceutical products and tissue samples. The sample is sealed in a sterile bag. Paddles in the stomacher crush, blend, and homogenize the samples under conditions that prevent contamination by other organisms. Although a traditional blender could also be used to process samples, the sealed bag stomacher system prevents contamination from outside sources, eliminates cleanup between each sample run, and eliminates generation of aerosols. The homogenized samples can then be analyzed in various ways.

Foods sampled for microorganisms or toxins should be examined as soon after processing as possible; if examination cannot begin within 1 hour of sampling, the food should be refrigerated. Frozen food should be thawed in the original container in a refrigerator and examined or cultured as soon as thawing is complete. In addition to identifying pathogens in food, disease investigators must obtain foodborne pathogens from the disease outbreak patients to establish a cause-and-effect relationship between the pathogen and the illness. In many cases, fecal samples can be cultured to recover suspected foodborne pathogens.

Food or patient samples can be inoculated onto enriched media, followed by transfer to differential or selective media for isolation and identification, as described for the isolation of human pathogens (Section 31.2). Final identification of foodborne pathogens is based on growth characteristics and biochemical reaction patterns. The use of molecular and genetic methods such as the polymerase chain reaction, enzyme immunoassays, nucleic acid probes, nucleic acid sequencing, pulsed-field gel electrophoresis (PFGE), and ribotyping may be used to identify specific organisms.

Figure 36.10 A stomacher. Paddles in this specialized blender homogenize the solid food sample in a sealed, sterile bag. The sample is suspended in a sterile solution.

MiniQuiz

- Distinguish between food infection and food poisoning.
- Describe microbial sampling procedures for solid foods such as meat.

36.5 Foodborne Disease Epidemiology

There are often clusters of cases of a foodborne disease in a particular place because microorganisms from a single common contaminated food, such as salads or hamburgers served from a home, school cafeteria, college dining hall, restaurant, or mess hall, are ingested by many individuals. In addition, central processing plants and central food distribution centers provide opportunities for contaminated foods to cause multiple disease outbreaks in far-flung locations, as when contaminated spinach grown in California caused outbreaks across the United States. We shall see how the food epidemiologist tracks outbreaks and determines their source, often down to the field, processing plant, or point-of-preparation facility in which the food was contaminated.

Spinach and *Escherichia coli* O157:H7

In 2006 an outbreak of illness associated with *Escherichia coli* O157:H7 occurred in the United States and was linked to the consumption of ready-to-eat packaged fresh spinach. The outbreak was quickly traced to a food-processing facility in California. First linked to the spinach product in September, the outbreak caused at least 199 infections. Of these, 102 individuals were hospitalized and 31 developed hemolytic uremic syndrome. At least three deaths were attributed to the outbreak.

Figure 36.11 ***Escherichia coli*** **O157:H7.** The cell, about 1 μm in diameter, as it appears in a colorized transmission electron micrograph showing peritrichous flagella.

The remarkably short duration and rapid end to this epidemic—the first case was confirmed in late August and the last reported in early October—is a testament to efficiency and cooperation among public health facilities across the country. We discussed surveillance networks for infectious disease information in Chapter 32. In this case, two of these networks—FoodNet and PulseNet—were used to define the source and stop the outbreak. The contaminated spinach was distributed nationwide from the California processing plant, but most disease cases were not in the West. The two states affected most were Wisconsin, with 49 cases, and Ohio, with 25 cases; there were only 2 cases in California.

Because *E. coli* O157:H7 (**Figure 36.11**) has been well studied, public health officials were able to identify the strain found in the bagged spinach and determine its origin. They conclusively linked the outbreak to the bagged spinach, traced it back to the processing plant, and eventually traced it to an agricultural field in the vicinity of the processing plant. DNA from the organisms isolated from regional outbreaks was typed using pulsed-field gel electrophoresis (PFGE), a form of gel electrophoresis that better distinguishes between large molecules and is used in pathogen identification. The patterns obtained were then compared; the results showed that the same strain was responsible for the disease in various parts of the country. The common thread in the geographically isolated outbreaks was consumption of the suspected lots of bagged spinach originating from a single California facility.

The precise source of the outbreak, although it has been traced to a field near the processing plant, remains unknown. Feral pigs and domestic cattle are present in the vicinity of the identified field, and contaminated wells or surface waters used for irrigation may have introduced the pathogen into the fields and eventually into the spinach. The original source was almost certainly animal in origin, as *E. coli* is an enteric organism found naturally only in the intestine of animals.

The spinach epidemic, although serious and even deadly for some, was discovered, contained, and stopped very quickly. However, this incident also shows how centralized food-processing facilities can quickly spread disease to large and distant populations. Food hygiene standards and surveillance must be maintained at the highest possible level in central food-processing and distribution facilities.

Food Disease Reporting

In the United States foodborne outbreaks are reportable to the Centers for Disease Control and Prevention through *FoodNet*. Identification of particular organisms responsible for foodborne disease outbreaks is particularly important. A reporting system called *PulseNet International* is an international molecular subtyping network for foodborne disease surveillance. It consists of national and regional PulseNet organizations from the United States, Canada, Europe, Asia, Latin America, the Caribbean, and the Middle East. The organization collects and shares molecular subtyping data from PFGE DNA fingerprints of organisms implicated in foodborne disease outbreaks. Tracking the characteristics of foodborne illnesses and identifying the causal agents often allows epidemiologists to pinpoint the original source of contaminated food, as we discussed above.

MiniQuiz

- Identify the potential for a foodborne disease outbreak from a single contamination event at a centralized food-processing facility.
- Describe tracking of a foodborne disease outbreak.

III Food Poisoning

Food poisoning can be caused by various bacteria and fungi. Here we consider *Staphylococcus* and *Clostridium*, the two genera responsible for the highest numbers of microbial food poisoning cases.

36.6 Staphylococcal Food Poisoning

Food poisoning is often caused by staphylococcal enterotoxin (SE) produced by the bacterium *Staphylococcus aureus*. Staphylococci are small, gram-positive cocci (**Figure 36.12**; Section 18.1) and, as we discussed in Section 33.9, they are normal members of the flora of the skin and upper respiratory tract of at least 20–30% of all humans, and are often opportunistic pathogens. *S. aureus* is frequently associated with food poisoning because it can grow in many common foods, and some strains produce several heat-stable enterotoxins. SE consumed in food produces gastroenteritis characterized by nausea, vomiting, and diarrhea, usually within 1–6 hours.

Epidemiology

Each year there are an estimated 185,000 cases of staphylococcal food poisoning in the United States (Table 36.6). The foods most commonly responsible are custard- and cream-filled baked goods, poultry, eggs, raw and processed meat, puddings, and creamy salad dressings. Salads prepared with mayonnaise-based dressings such as those containing shellfish, chicken, pasta, tuna, potato, egg, or meat are also commonly implicated. If these foods are refrigerated immediately after preparation, they usually remain

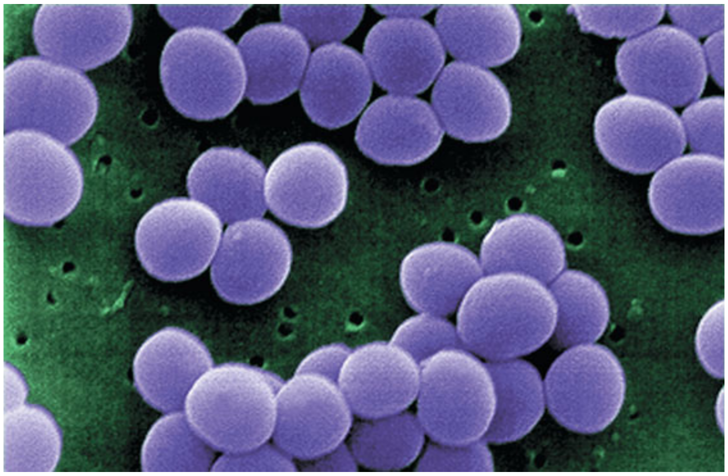

Figure 36.12 ***Staphylococcus aureus*****.** In this colorized scanning electron micrograph, the individual gram-positive cocci are about 0.8 μm in diameter. Staphylococci divide in multiple planes, producing the appearance that gives the genus its name (from the Greek *staphyle*, bunch of grapes).

safe because *S. aureus* grows poorly at low temperatures. However, foods kept at room temperature in kitchens or outdoors at picnics can support rapid bacterial growth and enterotoxin production if contaminated with *S. aureus*. Even if the toxin-containing foods are heated before eating, the heat-stable toxin may remain active. Some SEs are stable for over 16 hours at 60°C, a temperature that would kill *S. aureus*. Live *S. aureus* need not be present in foods causing illness: The illness is solely due to the preformed SE.

Staphylococcal Enterotoxins

S. aureus strains produce up to 20 different but related SEs. Most strains of *S. aureus* produce only one or two of these toxins, and some strains are nonproducers. However, any one of the toxins can cause staphylococcal food poisoning. These enterotoxins are further classified as *superantigens*. Superantigens stimulate large numbers of T cells, which in turn release intercellular mediators called *cytokines*. In the intestine, superantigens activate a general inflammatory response that causes gastroenteritis and significant fluid loss due to diarrhea and vomiting (Section 28.10).

The *S. aureus* enterotoxins are called SEA, SEB, SEC, and SED and are encoded by the genes *SEA*, *SEB*, *SEC*, and *SED*. *SEB* and *SEC* are on the bacterial chromosome, *SEA* is on a lysogenic bacteriophage, and *SED* is on a plasmid. The *S. aureus* SE genes are genetically related. The phage- and plasmid-encoded genes are movable genetic elements that can transfer toxin production to nontoxigenic strains of *Staphylocccus* by horizontal gene transfer (Section 12.11).

Diagnosis, Treatment, and Prevention

Certain assays detect SEs in food, and other assays detect *S. aureus* exonuclease, an enzyme that degrades DNA, as a metabolite in food. These qualitative tests confirm that *S. aureus* is or has been present. To obtain quantitative data and determine the extent of bacterial contamination, bacterial plate counts are required. For staphylococcal counts, a high-salt medium (either sodium chloride or lithium chloride at a final concentration of 7.5%) is used. Compared to most bacteria present in foods, staphylococci thrive in habitats with a high salt content and low water activity.

The symptoms of *S. aureus* food poisoning can be quite severe, but are typically self-limiting, usually resolving within 48 hours as the toxin passes from the body. Severe cases may require treatment for dehydration. Treatment with antibiotics is not useful because staphylococcal food poisoning is caused by a preformed toxin, not an active bacterial infection. Staphylococcal food poisoning can be prevented by proper sanitation and hygiene in food production, food preparation, and food storage. As a rule, foods susceptible to colonization by *S. aureus* and kept for several hours at temperatures above 4°C should be discarded rather than eaten.

MiniQuiz

- Identify the symptoms and mechanism of staphylococcal food poisoning.
- Why does antibiotic treatment not affect the outcome or the severity of disease in patients with staphylococcal food poisoning?

36.7 Clostridial Food Poisoning

Clostridium perfringens and *Clostridium botulinum* cause serious food poisoning. Members of the genus *Clostridium* are anaerobic endospore-forming rods (Section 18.2). Canning and cooking procedures kill living organisms but do not necessarily kill all endospores. Under appropriate anaerobic conditions, the endospores in food can germinate and produce toxin.

Clostridium perfringens Food Poisoning

C. perfringens is an anaerobic, gram-positive, endospore-forming rod commonly found in soil (**Figure 36.13**). *C. perfringens* is also found in sewage, primarily because it lives in small numbers in the intestinal tract of many humans and animals. *C. perfringens*

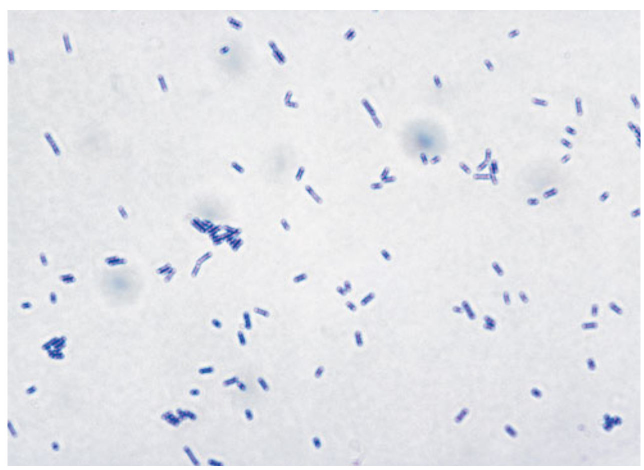

Figure 36.13 ***Clostridium perfringens*****.** The Gram stain shows individual gram-positive rods about 1 μm in diameter.

is the most often reported cause of food poisoning in the United States, with an estimated 248,000 annual cases (Table 36.6).

Perfringens food poisoning requires the ingestion of a large dose of *C. perfringens* ($>10^8$ cells) in contaminated cooked or uncooked foods, usually high-protein foods such as meat, poultry, and fish. Large numbers of *C. perfringens* can grow in meat dishes cooked in bulk where heat penetration is often insufficient. Surviving *C. perfringens* endospores germinate under anoxic conditions, as in sealed containers such as jars or cans. The *C. perfringens* grows quickly in the food, especially if left to cool at 20–40°C for short time periods. However, the toxin is not yet present at this stage.

Ingested with contaminated food, the living *C. perfringens* begin to sporulate and produce toxin in the consumer's intestine (Table 27.4). The perfringens enterotoxin alters the permeability of the intestinal epithelium, leading to nausea, diarrhea, and intestinal cramps, usually with no fever. The onset of perfringens food poisoning begins about 7–15 hours after consumption of the contaminated food, but usually resolves within 24 hours. Fatalities are rare.

Diagnosis, Treatment, and Prevention

Diagnosis of perfringens food poisoning is made by isolation of *C. perfringens* from the feces or, more reliably, by a direct enzyme immunoassay to detect *C. perfringens* enterotoxin in feces. Because *C. perfringens* food poisoning is self-limiting, antibiotic treatment is not indicated. Supportive therapy—fluids and electrolyte replacement—may be used in serious cases. Prevention of perfringens food poisoning requires preventing contamination of raw and cooked foods and proper heating of all foods during cooking and canning. Cooked foods should be refrigerated as soon as possible to rapidly lower temperatures and inhibit *C. perfringens* growth.

Botulism

Botulism is a severe, often fatal, food poisoning caused by the consumption of food containing the exotoxin produced by *C. botulinum*. This bacterium normally inhabits soil or water, but its endospores may contaminate raw foods. If the foods are properly processed so that the *C. botulinum* endospores are removed or killed, no problem arises; however, if viable endospores remain in the food, they may germinate and produce botulinum toxin. Ingesting even a small amount of this neurotoxin can be dangerous.

We discussed the nature and activity of botulinum toxin in Section 27.10 (Figure 27.22). Botulinum toxin is a neurotoxin that causes flaccid paralysis, usually affecting the autonomic nerves that control body functions such as respiration and heartbeat. At least seven distinct botulinum toxins are known. Because the toxins are destroyed by heat (80°C for 10 minutes), thoroughly cooked food, even though contaminated with toxin, is totally harmless.

Most cases of foodborne botulism are caused by eating processed foods contaminated with *C. botulinum* endospores. Typically, such foods are consumed without cooking after processing. For example, nonacid, home-canned vegetables such as corn and beans are often used without cooking when making cold salads. Smoked and fresh fish, vacuum-packed in plastic, are also often

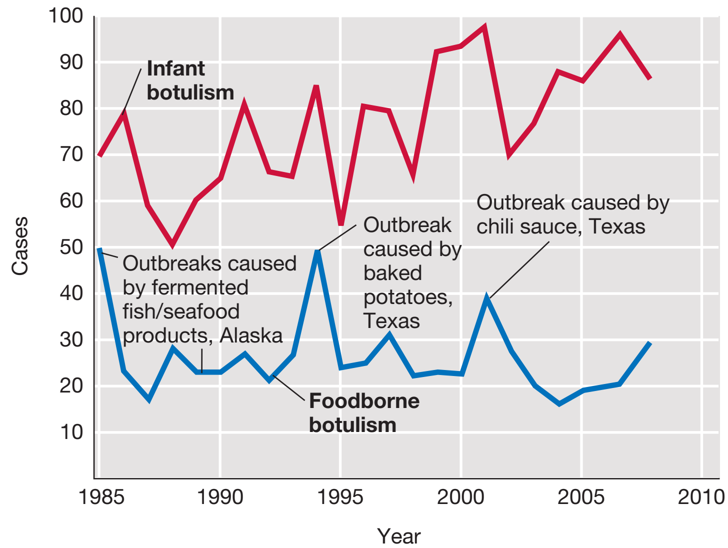

Figure 36.14 Botulism in the United States. Both foodborne and infant botulism are shown. In years with high numbers of cases, major outbreaks that account for the increase are indicated. Data are from the Centers for Disease Control and Prevention, Atlanta, Georgia, USA.

eaten without cooking. Under such conditions, viable *C. botulinum* endospores may germinate, and the vegetative cells may produce sufficient toxin to cause severe food poisoning. An average of 25 cases of foodborne botulism, about 18% of all botulism, occurred annually in the United States between 2000 and 2007 (**Figure 36.14**).

The majority of botulism cases occur following *infection* with *C. botulinum*. For example, infant botulism occurs after newborns ingest endospores of *C. botulinum* (Figure 36.14). In most cases, the source cannot be identified because *C. botulinum* endospores are widespread. If the infant's normal flora is not developed or if the infant is undergoing antibiotic therapy, ingested endospores can germinate in the infant's intestine, triggering *C. botulinum* growth and toxin production. Most cases of infant botulism occur between the first week of life and 2 months of age, rarely occurring in children older than 6 months, presumably because the normal intestinal flora is more developed. Over 60% of all botulism cases in the United States are in infants. An average of 86 cases of infant botulism occurred annually in the United States from 2000 to 2007 (Figure 36.14). Wound botulism can also occur from infection, presumably from endospores in contaminating material introduced via a parenteral route. Wound botulism is most commonly associated with illicit injectable drug use; in the United States an average of about 29 cases occurred annually from 2000 to 2007.

All forms of botulism are quite rare, with at most six cases occurring per 10 million individuals per year in the United States. Botulism, however, is a very serious disease because of the high mortality associated with the disease; about 16% of all foodborne cases are fatal. Death occurs from respiratory paralysis or cardiac arrest due to the paralyzing action of the botulinum neurotoxin.

Diagnosis, Treatment, and Prevention

Botulism is diagnosed when botulinum toxin is found in patient serum or when toxin or live *C. botulinum* is found in food the patient has ingested. Laboratory findings are coupled with clinical observations, including neurological signs of localized paralysis (impaired vision and speech) beginning 18–24 hours after ingestion of contaminated food. Treatment is by administration of botulinum antitoxin if the diagnosis is early, and mechanical ventilation for flaccid respiratory paralysis. In infant botulism, *C. botulinum* and toxin are often found in bowel contents. Infant botulism is usually self-limiting, and most infants recover with only supportive therapy, such as assisted ventilation. Antitoxin administration is not recommended. Respiratory failure causes occasional deaths.

Prevention of botulism requires careful control of canning and preservation methods. Susceptible foods should be heated to destroy endospores; boiling for 20 minutes destroys the toxin. Home-prepared foods are the most common source of foodborne botulism outbreaks.

MiniQuiz

- Describe the events that lead to *Clostridium perfringens* food poisoning. What is the likely outcome of the poisoning?
- Describe the development of botulism in adults and infants. What is the likely outcome of botulism?

IV Food Infection

Food infection results from ingestion of food containing sufficient numbers of viable pathogens to cause infection and disease in the host. Food infection is very common (Table 36.6), and we begin with a common bacterial cause, *Salmonella*. Many food infection agents can also cause waterborne diseases.

36.8 Salmonellosis

Salmonellosis is a gastrointestinal disease typically caused by foodborne *Salmonella* infection. Symptoms begin after the pathogen colonizes the intestinal epithelium. *Salmonella* are gram-negative, facultatively aerobic, motile rods related to *Escherichia coli* and other enteric bacteria (Section 17.11). *Salmonella* normally inhabits the animal intestine and is thus found in sewage.

The nomenclature of the *Salmonella* spp. is based on taxonomic schemes that differentiate strains by virtue of biochemical, serological, and molecular (nucleic acid–based) characteristics. The accepted species name for the pathogenic members of the genus is *Salmonella enterica*. Based on nucleic acid analyses, there are seven evolutionary groups or subspecies of *S. enterica*. Most human pathogens fall into group I, designated as a single subspecies, *S. enterica* subspecies *enterica*. Finally, each subspecies may be divided into *serovars* (serological variations, also called *serotypes*). Thus, the organism formally named *Salmonella enterica* subspecies *enterica* serovar Typhi is usually called *Salmonella enterica* serovar Typhi and is often abbreviated to *Salmonella Typhi. S. enterica* ser. Typhi causes the serious human disease typhoid fever but is rare in the United States. Most of the 500 or so annual foodborne cases caused by *S. enterica* ser. Typhi are acquired outside the United States.

A number of other *S. enterica* serovars also cause foodborne gastroenteritis. In all, over 1400 *Salmonella* serovars cause disease in humans. *S. enterica* serovars Typhimurium and Enteritidis are the most common agents of foodborne salmonellosis in humans.

Epidemiology and Pathogenesis

The incidence of salmonellosis has been steady over the last decade, with about 40,000–45,000 documented cases each year (**Figure 36.15**). However, less than 4% of salmonellosis cases are probably reported, so the incidence of salmonellosis is probably over 1 million cases every year (Table 36.6).

The ultimate sources of the foodborne salmonellas are the intestinal tracts of humans and other warm-blooded animals, and there are several routes by which these bacteria may enter the food supply. The bacteria may reach food through fecal contamination from food handlers. Food production animals such as chickens, pigs, and cattle may harbor *Salmonella* serovars that are pathogenic to humans, and the bacteria may be carried through to finished fresh foods such as eggs, meat, and dairy products. *Salmonella* food infections are often traced to products such as custards, cream cakes, meringues, pies, and eggnog made with uncooked eggs. Other foods commonly implicated in salmonellosis outbreaks are meats and meat products such as meat pies, cured but uncooked sausages and meats, poultry, milk, and milk products.

The most common salmonellosis is enterocolitis. Ingestion of food containing viable *Salmonella* results in colonization of the small and large intestine. Onset of the disease occurs 8–48 hours after ingestion. Symptoms include the sudden onset of headache,

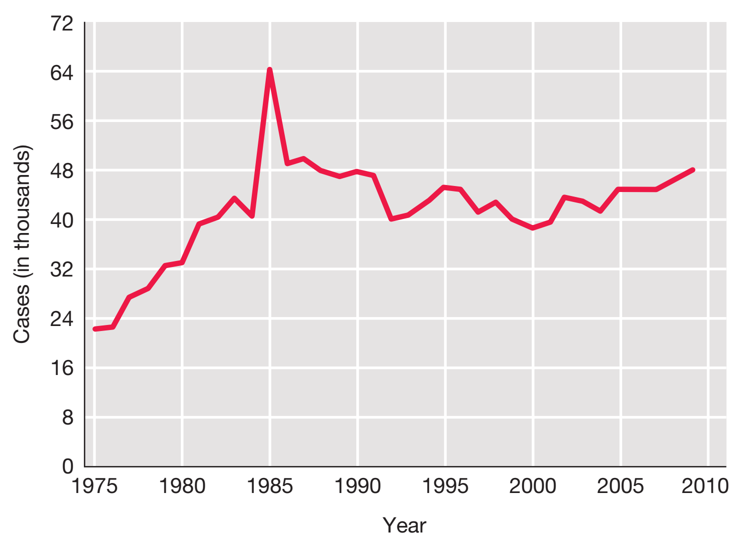

Figure 36.15 **Reported cases of salmonellosis in the United States.** Most cases of salmonellosis are foodborne. The total number of reported cases in 2007 was 47,995. The high incidence in 1985 was caused by contamination of pasteurized milk that was mixed with raw (unprocessed) milk in a dairy processing plant in Illinois. Data are from the Centers for Disease Control and Prevention, Atlanta, Georgia, USA.

chills, vomiting, and diarrhea, followed by a fever that lasts a few days. The disease normally resolves without intervention in 2–5 days. After recovery, however, patients may shed *Salmonella* in feces for several weeks. Some patients recover and remain asymptomatic, but shed organisms for months or even years; they are chronic carriers (Section 32.3). A few serovars of *Salmonella* may also cause septicemia (a blood infection) and enteric or typhoid fever, a disease characterized by systemic infection and high fever lasting several weeks. Mortality can approach 15% in untreated typhoid fever.

The pathogenesis of *Salmonella* infections starts with uptake of the organisms from the gut. *Salmonella* ingested in food or water invades phagocytes and grows as an intracellular pathogen, spreading to adjacent cells as host cells die. After invasion, pathogenic *Salmonella* uses a combination of endotoxins, enterotoxins, and cytotoxins to damage and kill host cells (see the Chapter 27 Microbial Sidebar "Virulence in *Salmonella*"), leading to the classic symptoms of salmonellosis.

Diagnosis, Treatment, and Prevention

Foodborne salmonellosis is diagnosed from observation of clinical symptoms, history of recent food consumption, and culturing the organism from feces. Selective and differential media are used to identify *Salmonella* and discriminate it from other gram-negative rods (Table 31.2). Tests for the presence of *Salmonella* are commonly used on animal food products, such as raw meat, poultry, eggs, and powdered milk. *Salmonella* has also been found, however, in nonmeat and nondairy food, including produce (cantaloupes and tomatoes) and peanut butter. Tests for *Salmonella* in food include several rapid tests, but even rapid tests rely on culture-based enrichment procedures to increase *Salmonella* numbers to testable levels. The established standard used by PulseNet for epidemiological investigations is pulsed-field gel electrophoresis (PFGE; Section 36.5). This molecular typing technique can discriminate between various *Salmonella* serovars.

For enterocolitis, treatment is usually unnecessary, and antibiotic treatment does not shorten the course of the disease or eliminate the carrier state. Antibiotic treatment, however, significantly reduces the length and severity of septicemia and typhoid fever. Mortality due to typhoid fever can be reduced to less than 1% with appropriate antibiotic therapy. Multi-drug-resistant *Salmonella* are a significant clinical problem.

Properly cooked foods heated to at least 70°C are generally safe if consumed immediately, held at 50°C, or stored immediately at 4°C. Any foods that become contaminated by an infected food handler can support the growth of *Salmonella* if the foods are held for long periods of time, especially without heating or refrigeration. *Salmonella* infections are more common in summer than in winter, probably because warm environmental conditions generally favor the growth of microorganisms in foods.

Although local laws and enforcement vary, because of the lengthy carrier state, infected individuals are often banned from work as food handlers until their feces are negative for *Salmonella* in three successive cultures.

MiniQuiz

- Describe salmonellosis food infection. How does it differ from food poisoning?
- How might *Salmonella* contamination of food production animals be contained?

36.9 Pathogenic *Escherichia coli*

Most strains of *Escherichia coli* are common members of the enteric microflora in the human colon and are not pathogenic. A few strains, however, are potential foodborne pathogens. There are about 200 known pathogenic *E. coli* strains, all of which act on the intestine. Several are characterized by their production of potent enterotoxins and may cause life-threatening diarrheal disease and urinary tract infections. The pathogenic strains are divided based on the type of toxin they produce and the specific diseases they cause.

Shiga Toxin–Producing *Escherichia coli* (STEC)

Shiga toxin–producing *Escherichia coli* (STEC) produce *verotoxin*, an enterotoxin similar to the Shiga toxin produced by *Shigella dysenteriae* (Table 27.4). Formerly known as enterohemorrhagic *E. coli* (EHEC), the most widely distributed STEC is *E. coli* O157:H7 (Figure 36.11). Up to 90% of all STEC infections are caused by *E. coli* O157:H7. After a person ingests food or water containing STEC, the bacteria grow in the small intestine and produce verotoxin. Verotoxin causes both hemorrhagic (bloody) diarrhea and kidney failure. *E. coli* O157:H7 causes an estimated 60,000 infections and 50 deaths from foodborne disease in the United States each year (Table 36.6). STEC strains are the leading cause of hemolytic uremic syndrome and kidney failure, with 292 cases reported in 2007, about half in children under 5 years of age.

About 40% of STEC infections are caused by the consumption of contaminated uncooked or undercooked meat, particularly mass-processed ground beef. *E. coli* O157:H7 is a member of the normal microbiome in healthy cattle; it can enter the human food chain if meat is contaminated with intestinal contents during slaughter and processing. In several major outbreaks in the United States caused by *E. coli* O157:H7, infected ground beef from regional distribution centers was the source of contamination. Infected meat products caused disease in several states. Another outbreak was caused by processed and cured, but uncooked beef in ready-to-eat sausages. The source of contamination was the beef, and the *E. coli* O157:H7 probably originated from slaughtered beef carcasses.

In 2003, the Food Safety and Inspection Service of the United States Department of Agriculture reported 20 positive results of 6584 samples (0.03%) of ground beef analyzed for *E. coli* O157:H7. *E. coli* O157:H7 has also been implicated in food infection outbreaks from dairy products, fresh fruit, and raw vegetables. Contamination of the fresh foods by fecal material, typically from cattle carrying the *E. coli* O157:H7 strain, has been implicated in several of these cases, as we discussed in Section 36.5.

Because *E. coli* O157:H7 grows in the intestines and is found in fecal material, it is also a potential source of waterborne gastrointestinal disease. Several outbreaks have also occurred in day-care facilities, where the presumed route of exposure is oral–fecal contamination.

Other Pathogenic *Escherichia coli*

Children in developing countries often contract diarrheal disease caused by *E. coli*. *E. coli* can also be the cause of "traveler's diarrhea," a common enteric infection causing watery diarrhea in travelers to developing countries. The primary causal agents are the enterotoxigenic *E. coli* (ETEC). The ETEC strains usually produce one of two heat-labile, diarrhea-producing enterotoxins. In studies of United States citizens traveling in Mexico, the infection rate with ETEC is often greater than 50%. The prime vehicles are foods such as fresh vegetables (for example, lettuce in salads) and water. The very high infection rate in travelers is due to contamination of local public water supplies. The local population is usually resistant to the infecting strains, presumably because they have acquired resistance to the endemic ETEC strains. Secretory IgA antibodies in the bowel prevent colonization of the pathogen in local residents, but the organism readily infects the nonimmune travelers and causes disease.

Enteropathogenic *E. coli* (EPEC) strains cause diarrheal diseases in infants and small children but do not cause invasive disease or produce toxins. Enteroinvasive *E. coli* (EIEC) strains cause invasive disease in the colon, producing watery, sometimes bloody diarrhea. The EIEC strains are taken up by phagocytes, but escape lysis in the phagolysosomes, grow in the cytoplasm, and move into other cells in much the same way as pathogenic *Salmonella* strains. This invasive disease causes diarrhea and is common in developing countries.

Diagnosis and Treatment

Illness from *E. coli* O157:H7 and other STEC strains is a reportable infectious disease in the United States. The general pattern established for diagnosis, treatment, and prevention of infection by *E. coli* O157:H7 reflects current procedures used for all of the pathogenic *E. coli* strains. Laboratory diagnosis requires culture from the feces and identification of the O (lipopolysaccharide) and H (flagellar) antigens and toxins by serology. Identification of strains is also done using DNA analyses such as restriction fragment length polymorphism and PFGE. *E. coli* O157:H7 outbreaks are reported through FoodNet and PulseNet to the Centers for Disease Control and Prevention.

Treatment of *E. coli* O157:H7 and other STEC infections includes supportive care and monitoring of renal function, blood hemoglobin, and platelets. Antibiotics may be harmful because they may cause the release of large amounts of verotoxin from dying *E. coli* cells. For other pathogenic *E. coli* infections, treatment usually includes supportive therapy and, for severe cases and invasive disease, antimicrobial drugs to shorten and eliminate infection.

Prevention

The most effective way to prevent infection with foodborne STEC is to make sure that meat is cooked thoroughly, which means that it should appear gray or brown and juices should be clear. As we discussed above (Section 36.2), the United States has approved the irradiation of ground meat as an acceptable means of eliminating or reducing food infection bacteria, largely because *E. coli* O157:H7 has been implicated in several foodborne epidemics. To process foods such as ground beef, large-scale production plants may mix and grind meat from hundreds or even thousands of animals together; the grinding process could distribute the pathogens from a single infected animal throughout the meat. Short of cooking, penetrating radiation is considered the only effective means to ensure decontamination.

In general, proper food handling, water purification, and appropriate hygiene prevent the spread of pathogenic *E. coli*. Raw foods should be washed thoroughly. Traveler's diarrhea can be prevented by avoiding consumption of local water and uncooked foods.

MiniQuiz

- Describe the pathology of *Escherichia coli* food infections due to STEC, ETEC, EPEC, and EIEC strains.
- Why is *E. coli* O157:H7 considered a dangerous and reportable pathogen?

36.10 *Campylobacter*

Species of *Campylobacter* are the most common reported cause of bacterial foodborne infections in the United States. Cells of *Campylobacter* species are gram-negative, motile, curved rods to spiral-shaped bacteria that grow at reduced oxygen tension as microaerophiles (Section 17.19). Several pathogenic species, *Campylobacter jejuni* (**Figure 36.16**), *C. coli*, and *C. fetus*, are recognized. *C. jejuni* and *C. coli* account for almost 2 million annual cases of bacterial diarrhea (Table 36.6). *C. fetus* is a major cause of sterility and spontaneous abortion in cattle and sheep.

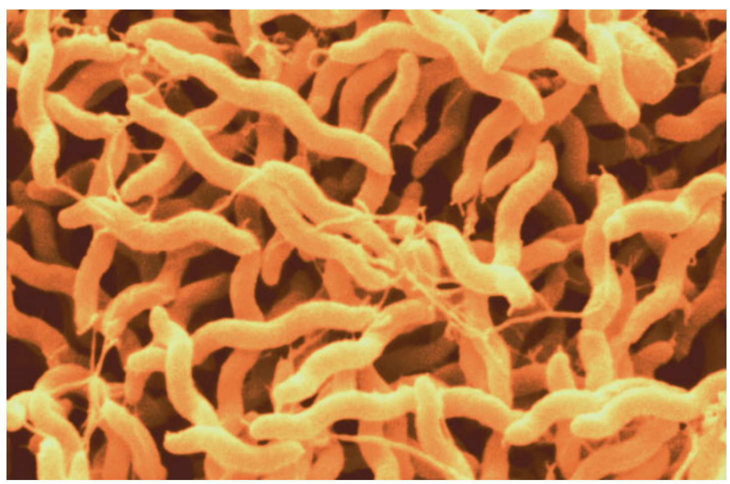

Figure 36.16 ***Campylobacter jejuni*.** The gram-negative curved rods shown in this colorized scanning electron micrograph are about 1 μm in diameter.

Epidemiology and Pathology

Campylobacter is transmitted to humans via contaminated food, most frequently in poultry, pork, raw shellfish, or in surface waters. *C. jejuni* is a normal resident in the intestinal tract of poultry; virtually all chickens and turkeys are normally colonized with this organism. According to the United States Department of Agriculture, up to 90% of turkey and chicken carcasses and over 30% of hog carcasses may be contaminated with *Campylobacter*. Beef, on the other hand, is rarely a vehicle for this pathogen. *Campylobacter* species also infect domestic animals such as dogs, causing a milder form of diarrhea than that observed in humans. *Campylobacter* infections in infants are frequently traced to infected domestic animals, especially dogs.

After a person ingests cells of *Campylobacter*, the organism multiplies in the small intestine, invades the epithelium, and causes inflammation. Because *C. jejuni* is sensitive to gastric acid, cell numbers as high as 10^4 may be required to initiate infection. However, this number may be reduced to less than 500 if the bacteria are ingested in food, or are ingested by a person taking medication to reduce stomach acid production. *Campylobacter* infection causes a high fever (usually greater than 104°F or 40°C), headache, malaise, nausea, abdominal cramps, and profuse diarrhea with watery, frequently bloody, stools. The disease subsides in about 7–10 days. Spontaneous recovery from *Campylobacter* infections is often complete, but relapses occur in up to 25% of cases.

Diagnosis, Treatment, and Prevention

Diagnosis of *Campylobacter* food infection requires isolation of the organism from stool samples and identification by growth-dependent tests, immunological assays, or molecular tests. Serious *C. jejuni* infections are often seen in infants. In these cases, diagnosis is important; selective media and specific immunological methods have been developed for positive identification of this organism. Erythromycin and quinolone treatment may be useful early in severe diarrheal disease. Adequate personal hygiene, proper washing of uncooked poultry (and any kitchenware coming in contact with uncooked poultry), and thorough cooking of meat eliminate *Campylobacter* contamination.

As with other foodborne infections, epidemiologic investigations are based on PFGE analysis of recovered organisms. Data shared on PulseNet are used to track the spread of *Campylobacter* and determine its origin.

MiniQuiz

- Describe the pathology of *Campylobacter* food infection. What is the likely outcome?
- How might *Campylobacter* contamination of food production animals be controlled?

36.11 Listeriosis

Listeria monocytogenes causes **listeriosis**, a gastrointestinal food infection that may lead to bacteremia and meningitis. *L. monocytogenes* is a short, gram-positive, nonsporulating coccobacillus that is acid-, salt- and cold-tolerant and facultatively aerobic (**Figure 36.17**; Section 18.1).

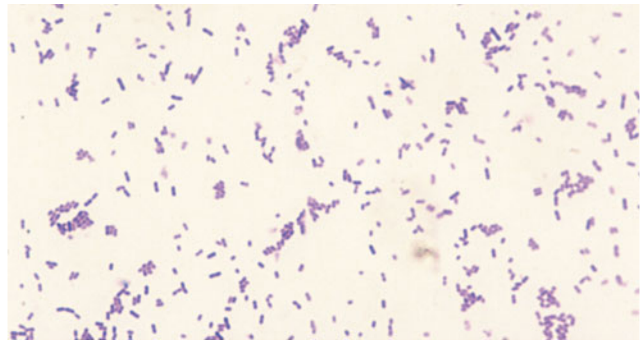

Figure 36.17 ***Listeria monocytogenes*.** This Gram stain shows gram-positive coccobacilli, about 0.5 μm in diameter.

Epidemiology and Pathology

L. monocytogenes is found widely in soil and water; virtually no food source is safe from possible *L. monocytogenes* contamination. Food can become contaminated at any stage during food production or processing. Food preservation by refrigeration, which ordinarily slows microbial growth, is ineffective in limiting growth of this psychrotolerant organism. Ready-to-eat meats, fresh soft cheeses, unpasteurized dairy products, and inadequately pasteurized milk are the major food vehicles for this pathogen, even when foods are properly stored at refrigerator temperature (4°C).

L. monocytogenes is an intracellular pathogen. It enters the body through the gastrointestinal tract in contaminated food. Phagocytes take up the pathogen in a phagolysosome. This triggers production of listeriolysin O, which lyses the phagolysosome and releases *L. monocytogenes* into the cytoplasm. Here it multiplies and produces ActA, a surface protein that induces host cell actin polymerization, which moves the pathogen to the cytoplasmic membrane. At the cytoplasmic membrane, the complex pushes out, forming protrusions called filopods. The filopods are then ingested by surrounding cells and the cycle starts again. This mechanism allows *L. monocytogenes* to move from cell to cell without exposure to antibodies, complement, or neutrophils. Specific immunity to *L. monocytogenes* is through cell-mediated T_H1 inflammatory cells (Section 29.6). Particularly susceptible populations include the elderly, pregnant women, newborns, and immunosuppressed individuals [for example, transplant patients undergoing steroid therapy and acquired immunodeficiency syndrome (AIDS) patients].

Although exposure to *L. monocytogenes* is undoubtedly very common, there are only about 2500 estimated cases of clinical listeriosis each year, and fewer than 1000 are reported. Nearly all diagnosed cases require hospitalization. Acute listeriosis is rare and is characterized by septicemia, often leading to meningitis, with a mortality rate of 20% or higher. About 30–40 listeriosis deaths are reported annually in the United States.

Diagnosis, Treatment, and Prevention

Listeriosis is diagnosed by culturing *L. monocytogenes* from the blood or spinal fluid. *L. monocytogenes* can be identified in food by direct culture or by molecular methods such as ribotyping and the polymerase chain reaction. Clinical isolates are analyzed by PFGE to determine molecular subtypes. The subtype patterns are reported to PulseNet at the Centers for Disease Control and Prevention. Intravenous antibiotic treatment with penicillin, ampicillin, or trimethoprim plus sulfamethoxazole is recommended for invasive disease.

Prevention measures include recalling contaminated food and taking steps to limit *L. monocytogenes* contamination at the food-processing site. Because *L. monocytogenes* is susceptible to heat and radiation, raw food and food-handling equipment can be readily decontaminated. However, without pasteurizing or cooking the finished food product, the risk of contamination cannot be eliminated because of the widespread distribution of the pathogen.

Individuals who are immunocompromised should avoid unpasteurized dairy products and ready-to-eat processed meats. Pregnant women should also avoid foods that may transmit *L. monocytogenes* because spontaneous abortion is a frequent outcome of listeriosis.

MiniQuiz

- What is the likely outcome of *Listeria monocytogenes* exposure in normal individuals?
- What populations are most susceptible to serious disease from *L. monocytogenes* infection? Why?

36.12 Other Foodborne Infectious Diseases

Over 200 other microorganisms, viruses, and other infectious agents such as prions contribute to foodborne diseases, and we consider a few of them here.

Bacteria

Table 36.6 lists several bacteria that cause human foodborne disease that we have not covered in this chapter. *Yersinia enterocolitica* is commonly found in the intestines of domestic animals and causes foodborne infections due to contaminated meat and dairy products. The most serious consequence of *Y. enterocolitica* infection is enteric fever, a severe life-threatening infection. *Bacillus cereus* produces two enterotoxins that cause diarrhea and vomiting. The organism grows in foods such as rice, pasta, meats, or sauces that are cooked and left at room temperature to cool slowly. Endospores of this gram-positive rod germinate and toxin is produced. Reheating may kill the *B. cereus*, but the toxin may remain active. *B. cereus* may also cause a food infection similar to that caused by *Clostridium perfringens*. *Shigella* species can cause severe invasive gastroenteritis called *shigellosis*. About 20,000 cases of shigellosis are reported each year in the United States, with up to 150 million cases worldwide. Most *Shigella* infections are the result of fecal–oral contamination, but food and water are occasional vehicles. Several members of the *Vibrio* genus cause food poisoning in persons who consume contaminated shellfish.

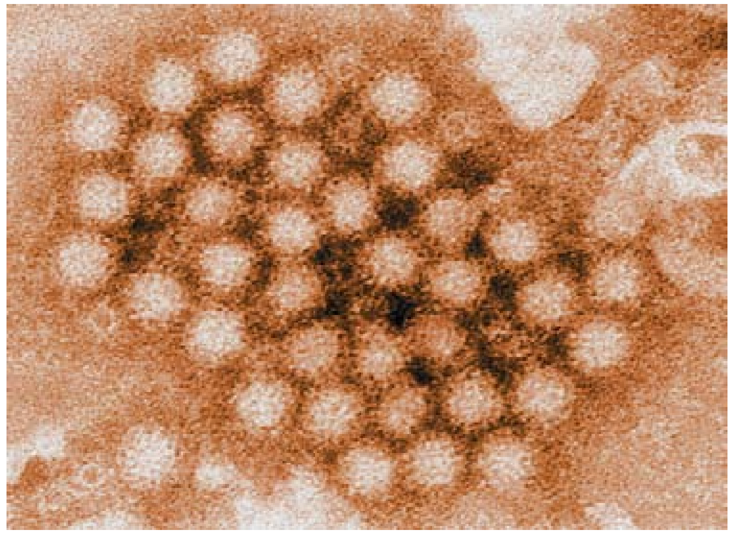

Figure 36.18 Human norovirus. The virus was isolated from a patient with diarrhea. Individual norovirus particles have an indistinct rough outer edge and are about 27 nm in diameter.

Viruses

The largest number of annual foodborne infections is thought to be caused by viruses. In general, viral foodborne illness consists of gastroenteritis characterized by diarrhea, often accompanied by nausea and vomiting. Recovery is spontaneous and rapid, usually within 24–48 hours ("24-hour bug"). Noroviruses (**Figure 36.18**) are responsible for most of these mild foodborne infections in the United States (Table 36.6), accounting for over 9 million of the estimated 13 million annual cases of foodborne disease. Rotavirus, astrovirus, and hepatitis A collectively cause 100,000 cases of foodborne disease each year. These viruses inhabit the gut and are often transmitted to food or water with fecal matter. As with many foodborne infections, proper food handling, handwashing, and a source of clean water to prepare fresh foods are essential to prevent infection.

Protists

Important foodborne protist diseases are listed in Table 36.6. Protists including *Giardia intestinalis, Cryptosporidium parvum* (Figures 35.16 and 35.17), and *Cyclospora cayetanensis* (**Figure 36.19*a***) can be spread in foods contaminated by fecal matter in untreated water used to wash, irrigate, or spray crops. Fresh foods such as fruits are often implicated as the source of these protists. We discussed giardiasis and cryptosporidiosis as waterborne diseases (Section 35.6). Cyclosporiasis is an acute gastroenteritis and is an important emerging disease. In the United States, most cases are acquired by eating fresh produce imported from other countries.

Toxoplasma gondii is a protist spread through cat feces, but is also found in raw or undercooked meat. In most individuals, toxoplasmosis is a mild, self-limiting gastroenteritis. However,

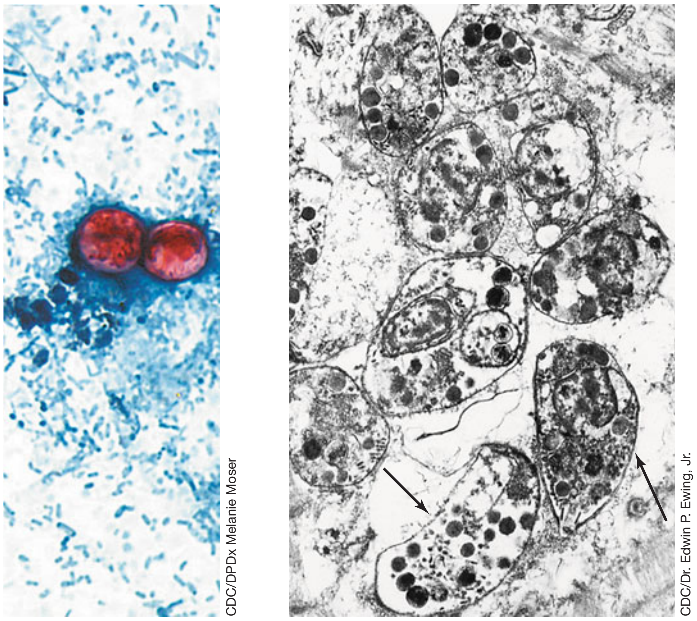

Figure 36.19 Protists transmitted in food. *(a) Cyclospora cayetanensis* oocysts in a stool sample from an affected patient. The oocysts, stained red with safranin, are about 8–10 μm in diameter. *(b)* Tachyzoites of *Toxoplasma gondii*, an intracellular parasite. In this transmission electron micrograph, the tachyzoites (arrows) are in a cystlike structure in a cardiac myocyte. Tachyzoites are generally elongated to crescent in form, about 4–7 μm long by 2–4 μm wide.

prenatal infection of the fetus can lead to serious acute toxoplasmosis resulting in tissue involvement, cyst formation, and complications such as myocarditis, blindness, and stillbirth. Immunocompromised individuals such as people with acquired immunodeficiency syndrome (AIDS) may develop acute toxoplasmosis. *T. gondii* grows intracellularly and forms structures called *tachyzoites* (Figure 36.19*b*) that eventually lyse the cell and infect nearby cells, resulting in tissue destruction. Tachyzoites can cross the placenta and infect the fetus. Toxoplasma infections in compromised hosts can be treated with the antiprotist drug pyrimethamine.

Prions, BSE, and nvCJD

Prions are proteins, presumably of host origin, that adopt novel conformations, inhibiting normal protein function and causing degeneration of neural tissue (Section 9.15). Human prion diseases are characterized by neurological symptoms including progressive depression, loss of motor coordination, and dementia.

A foodborne prion disease in humans known as new variant Creutzfeldt–Jakob Disease (nvCJD) has been linked to consumption of meat products from cattle afflicted with *bovine spongiform encephalopathy* (*BSE*), a prion disease commonly called "mad cow disease." A slow-acting degenerative nervous system disorder, nvCJD has a latent period that may extend for years after exposure to the BSE prion. Nearly 200 people in Great Britain and other European countries have acquired nvCJD. However, nvCJD linked to domestic meat consumption has not been observed in the United States. BSE prions consumed in meat products from affected cattle trigger human protein analogs to assume an altered conformation, resulting in protein dysfunction and disease (Figure 9.28). The terminal stages of both BSE and nvCJD are characterized by large vacuoles in brain tissue, giving the brain a "spongy" appearance, from which BSE derives its name (**Figure 36.20**).

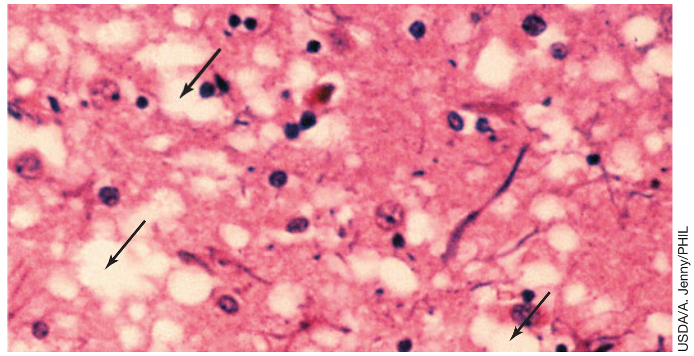

Figure 36.20 A brain section from a cow with bovine spongiform encephalopathy (BSE). The vacuoles, appearing as holes (arrows), give the brains of infected animals a distinct spongelike appearance.

In the United Kingdom and Europe, about 180,000 cattle were diagnosed with BSE and destroyed in the 1990s. Brains of slaughtered animals are routinely tested for BSE in the United States, and several cattle with BSE have been found in Canadian and U.S. herds. In Europe and North America, all cattle known or suspected to have BSE have been destroyed. Bans on cattle feeds containing cattle meat and bone meal appear to have stopped the development of new cases of BSE in Europe and have kept the incidence of this disease very low in North America. The infecting prions were probably transferred to food production animals through meat and bone meal feed derived from infected cattle or other animals not approved for human consumption.

Diagnosis of BSE is done by testing using a prion-susceptible mouse strain or by immunohistochemical or micrographic analysis of biopsied neural tissue (Figure 36.20).

MiniQuiz

- Identify the viruses, bacteria, and protists most likely to cause foodborne illnesses.
- How might prion contamination of food production animals be prevented in the United States?

Big Ideas

36.1
The growth of contaminating microorganisms causes most food spoilage. The potential for microbial food spoilage depends on the nutrient value and water content of the food. Microbial spoilage limits the shelf life of perishable and semiperishable foods. Some food spoilage microorganisms are also pathogens.

36.2
Microbial growth in foods must be limited to reduce spoilage and prevent disease. The growth of microorganisms in perishable foods can be controlled by refrigeration, freezing, canning, pickling, dehydration, aseptic processing, chemical preservation, and irradiation.

36.3
Microbial fermentation is used for preparing, preserving, and enhancing foods including breads, dairy products, meats, fruits, and vegetables.

36.4
Foodborne diseases include food poisoning and food infection. Food poisoning results from the action of microbial toxins, and food infections are due to the growth of microorganisms in the body. Specialized techniques are used to sample and identify microorganisms that cause foodborne disease outbreaks.

36.5
Tracking of foodborne disease outbreaks uses microbiological information disseminated via database-sharing networks. Identification of common characteristics of foodborne pathogens from seemingly isolated foodborne outbreaks can pinpoint the origin of foodborne contamination and track the spread of the disease.

36.6
Staphylococcal food poisoning results from the ingestion of a preformed staphylococcal enterotoxin, a superantigen produced by *Staphylococcus aureus* as it grows in food. In some cases, *S. aureus* cannot be cultured from toxin-containing food. Proper food preparation, handling, and storage can prevent staphylococcal food poisoning.

36.7
Clostridium food poisoning results from ingestion of toxins produced by microbial growth in foods or from microbial growth followed by toxin production in the body. Perfringens food poisoning is quite common and is usually a self-limiting gastrointestinal disease. Botulism is a rare but serious disease, with significant mortality.

36.8
More than 1 million cases of salmonellosis occur every year in the United States. Infection results from ingestion of *Salmonella* introduced into food from food production animals or food handlers.

36.9
Pathogenic *Escherichia coli* cause many food infections. Contamination of foods from fecal material spreads strains pathogenic to humans. Good hygiene practices and specific antibacterial measures such as irradiation of ground beef can control these pathogens.

36.10
Campylobacter infection is the most prevalent foodborne bacterial infection in the United States. Though usually self-limiting, this disease affects nearly 2 million people per year.

36.11
Listeria monocytogenes is an environmentally ubiquitous microorganism. In healthy individuals, *Listeria* seldom causes infection. However, in immunocompromised individuals, *Listeria* can cause serious disease and even death.

36.12
Over 200 different infectious agents cause foodborne disease. Viruses cause the most foodborne illnesses. Bacteria, protists, and prions also cause significant foodborne illness.

Review of Key Terms

Botulism food poisoning due to ingestion of food containing botulinum toxin produced by *Clostridium botulinum*

Canning sealing food in a container and heating to destroy living organisms and endospores

Fermentation the anaerobic catabolism of organic compounds, generally carbohydrates, in the absence of an external electron acceptor

Food infection a microbial infection resulting from the ingestion of pathogen-contaminated food followed by growth of the pathogen in the host

Food poisoning (food intoxication) a disease caused by the ingestion of food that contains preformed microbial toxins

Food spoilage a change in the appearance, smell, or taste of a food that makes it unacceptable to the consumer

Irradiation the exposure of food to ionizing radiation for the purpose of inhibiting growth of microorganisms and insect pests or to retard ripening

Listeriosis a gastrointestinal food infection caused by *Listeria monocytogenes* that may lead to bacteremia and meningitis

Lyophilization (freeze-drying) the removal of all water from frozen food under vacuum

Nonperishable (stable) food food of low water activity that has an extended shelf life and is resistant to spoilage by microorganisms

Perishable food fresh food generally of high water activity that has a very short shelf life due to potential for spoilage by growth of microorganisms

Pickling acidifying food to prevent microbial growth and spoilage

Salmonellosis enterocolitis or other gastrointestinal disease caused by any of over 1400 variants of *Salmonella* spp.

Semiperishable food food of intermediate water activity that has a limited shelf life due to potential for spoilage by growth of microorganisms

Water activity (a_w) a measure of the availability of water for use in metabolic processes

Review Questions

1. Identify and define the three major categories of food perishability (Section 36.1).
2. Identify the major methods used to preserve food. Provide an example of a food preserved by each method (Section 36.2).
3. Identify the major categories of fermented foods (Section 36.3).
4. Distinguish between food infection and food poisoning (Section 36.4).
5. Identify the organizations that track foodborne disease in the United States and internationally (Section 36.5).
6. Outline the pathogenesis of staphylococcal food poisoning. Suggest methods for prevention of this disease (Section 36.6).
7. Identify the two major types of clostridial food poisoning. Which is most prevalent? Which is most dangerous? Why (Section 36.7)?
8. What are the possible sources of *Salmonella* spp. that cause food infections (Section 36.8)?
9. What measures can control contamination by *Escherichia coli* O157:H7 and its growth in ground meat in food-processing and preparation settings (Section 36.9)?
10. *Campylobacter* causes more foodborne infections than any other bacterium. Identify at least one reason why this is true (Section 36.10).
11. Identify the food sources of *Listeria monocytogenes* infections. Identify the individuals who are at high risk for listeriosis (Section 36.11).
12. Why are viral agents so commonly associated with foodborne disease (Section 36.12)?

Application Questions

1. Identify optimum storage conditions for perishable, semiperishable, and nonperishable food products. Consider economic factors such as the cost of preservatives, storage space, and the intrinsic value of the food item.
2. For a food of your choice, devise a way to preserve the food by lowering the water activity without drying.
3. Perfringens food poisoning involves ingestion of *Clostridium perfringens* followed by growth and sporulation in the intestine of the host. Sporulation triggers toxin production. Is this disease truly a food poisoning, or might it be classified as a food infection? Explain.
4. Improperly handled potato salads are often the source of staphylococcal food poisoning or salmonellosis. Explain the means by which a potato salad could become contaminated with either *Staphylococcus aureus* or *Salmonella* spp.
5. *Clostridium botulinum* requires an anoxic environment for production of botulinum toxin. Identify methods of food preservation that create the anoxic environment necessary for growth of *C. botulinum*. Conversely, identify methods of food preservation that create an aerobic environment and prevent the growth of *C. botulinum*. What other factors influence the growth of *C. botulinum*?
6. Indicate the precautions necessary to prevent infection with pathogenic *Escherichia coli*. Concentrate on *E. coli* O157:H7 and safe food handling, cooking, and consumption practices.
7. Devise a plan to eliminate *Campylobacter* from a poultry flock or from the finished poultry product. Explain the benefits of *Campylobacter*-free poultry and explain the problems that your plan might encounter.
8. Listeriosis normally occurs only when there is a breakdown in T_H1 cell–mediated immunity. Indicate why this is so. Devise a vaccine to protect against listeriosis. Would your vaccine be an inactivated bacterial strain or product, or would it be an attenuated organism? Explain. Would your vaccine be of use in the listeriosis-prone population?
9. Indicate reasons for the high incidence of viral foodborne disease, especially with noroviruses. Devise a plan to eliminate noroviruses from the food supply.
10. Indicate the problems inherent in tracking a latent infectious agent such as the BSE prion. Can prion diseases be eliminated? If so, how?

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