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Foodborne Infections and Infestations

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Introduction

The microbiological safety of foods is a major concern to consumers and to the food industry. During the last decade, food safety received considerable attention due to the emergence of several new foodborne pathogens, and the involvement of foods that traditionally have been considered safe in many foodborne disease outbreaks. Further, increased globalization of the food supply and consumer demands for preservative-free convenience foods and ready-to-eat meals highlight the relevance of the microbial safety of foods. A recently published study by the U.S. Centers for Disease Control and Prevention reported an estimated 76 million cases of foodborne illness which resulted in 325,000 hospitalizations and 5000 deaths in the United States annually.[1] Besides the public health impact, outbreaks of foodborne illness impose major economic losses to both the food industry and society. The various microbiological hazards associated with foods can be classified as bacterial, viral, fungal, and parasitic.

Bacterial Foodborne Pathogens (Table 55.1)

Bacteria are a major agent of microbial foodborne illnesses. Bacterial foodborne illnesses can be classified into foodborne infections resulting from ingestion of foods containing viable cells of bacterial pathogens, and foodborne intoxications, which result from consumption of foods containing preformed toxins produced by toxigenic bacteria. The various bacterial pathogens associated with foodborne diseases are discussed below.

Escherichia coli O157:H7

Enterohemorrhagic *Escherichia coli* O157:H7 emerged in 1982 as a food-borne pathogen and is now recognized as a major public health concern in the United States. Many food-

TABLE 55.1

Bacterial Foodborne Pathogens

Microorganism	Biochemical and Growth Characteristics	Sources/ Reservoirs	Vehicles	Estimated No. of Foodborne Cases Annually in USA[1]	Incubation Period, Symptoms and Duration	Detection Methods	Control/Prevention
Escherichia coli O157:H7	Gram negative, facultative anaerobe, nonspore-forming, optimum growth at 37°-40°C, inability to grow at ≥ 44.5°C in presence of selective agents, inability to ferment sorbitol within 24 h, does not produce β-glucuronid-ase, acid tolerance	Cattle, humans	Raw or undercooked beef, unpasteurized milk and apple juice, lettuce, alfalfa sprouts, water	62,500	3 to 9 days Severe abdominal cramps, watery diarrhea that can become bloody, absence of fever, kidney failure, seizures, coma Duration is days to weeks	Cultural methods followed by confirmatory biochemical tests[102,103] Latex agglutination assay[104, 105] ELISA[106, 107] PCR[108]	Adequate cooking of beef; pasteurization of milk and apple juice; use of potable water for drinking; avoid eating alfalfa and vegetable sprouts; good personal hygiene
Salmonella spp. (non typhoid)	Gram negative, facultative anaerobe, oxidase negative, catalase positive, nonsporeforming, growth at 5°-47°C, optimum growth at 37°C, metabolize nutrients by respiratory and fermentative pathways	Cattle, swine, poultry, humans	Raw or undercooked meat, poultry, eggs, and milk and untreated water	1,340,000	6 to 72 h up to 4 days. Abdominal cramps, diarrhea, fever, chills, headache and vomiting. Duration is few days to one week, occasionally up to 3 weeks.	Cultural methods followed by confirmatory biochemical tests[109, 110] Latex agglutination assay[111] ELISA[112] PCR[113, 114]	Adequate cooking of food; avoid cross-contamina-tion of raw foods of animal origin with cooked or ready to eat foods; avoid eating raw or undercooked foods of animal origin; use of potable water; good personal hygiene
Salmonella enterica serovar Typhi	Gram negative, facultative anaerobe, ferment D-xylose	Humans	Raw milk, shellfish, raw salads, under cooked foods	660 (> 70% of cases acquired abroad)	7 to 28 days Remittent fever with stepwise increments over a period of days, high temperature of 103 to 104°F, abdominal pain, diarrhea, and headache Duration is up to 3 weeks	Biochemical tests[115] Latex test[116] ELISA[117] PCR[118]	Good personal hygiene and food handling practices; proper sewage systems; effective surveillance of known carriers

Campylobacter jejuni and *coli*	Gram negative, microaerophilic, nonsporeforming, optimal growth at 42°C, CO_2 is required for good growth, growth optimal in 3-6% O_2, sensitive to dehydration, survives best at refrigeration temperature	Poultry Swine Cattle Sheep Wild birds	Raw or undercooked chicken, pork, and beef, and unpasteurized milk	1,960,000	1 to 11 days, usually 2 to 5 days Abdominal pain, diarrhea, malaise, headache, fever Duration is up to 10 days	Cultural methods followed by confirmatory biochemical tests[31, 119] Immunoassay[120] PCR[121, 122]	Adequate cooking of meat; avoid cross-contamination of raw foods of animal origin with cooked or ready to eat foods; pasteurization of milk
Shigella spp.	Gram negative, facultative anaerobe, nonspore-forming, does not ferment lactose, growth at 10°-45°C, optimal growth at 37°C	Humans	Raw foods and water contaminated with human feces; prepared salads	89,600	1 to 7 days Severe abdominal and rectal pain, bloody diarrhea with mucus, fever Dehydration Duration is few days to few weeks	Cultural methods followed by confirmatory biochemical tests[123] ELISA[124] PCR[125]	Good personal hygiene, including adequate cooking of food, drinking potable water
Yersinia enterocolitica	Gram negative, facultative anaerobe, nonspore-forming, growth at 0°-44°C, optimal growth at ca. 29°C, growth at pH 4.6-9.0, growth in presence of 5% NaCl but not 7% NaCl	Swine is principal reservoir of pathogenic strains	Undercooked or raw pork, especially tongue	86,700	1 to 11 days, usually 24 to 36 h Severe abdominal pain, nausea, diarrhea, fever, sometimes vomiting Duration is usually 2-3 days but may continue for up to 3 weeks	Cultural methods followed by confirmatory biochemical tests[126] PCR[127]	Adequate cooking of pork, disinfection of drinking water, control of *Y. enterocolitica* in pigs, prevent cross-contamination of pig viscera, feces, and hair with food and water
Vibrio cholerae	Gram negative, facultative anaerobe, nonsporeforming, growth at 18°-42°C with optimal growth at 37°C, growth is stimulated in presence of 3% NaCl, pH range for growth is 6-11	Humans, marine waters, especially brackish water and estuaries	Undercooked or raw seafoods; vegetables fertilized with contaminated human feces or irrigated with contaminated water; water	49	1 to 3 days Profuse watery diarrhea, which can lead to severe dehydration, abdominal pain, vomiting Duration is up to 7 days	Cultural methods followed by confirmatory biochemical tests[128, 129] ELISA[130] PCR[131, 132]	Safe disposal of human sewage, disinfection of drinking water, avoid eating raw seafood, adequate cooking of food
Vibrio parahaemolyticus	Gram negative, facultative anaerobe, nonspore-forming, growth in presence of 8% NaCl, optimal growth at 37°C with rapid generation time (ca. 10 minutes), growth at 10°C, sensitive to storage at refrigeration temperature	Coastal seawater, estuarine brackish waters above 15°C, marine fish, shellfish	Raw or undercooked fish and seafoods	5100	9 to 25 hours, up to 3 days, Profuse watery diarrhea, abdominal pain, vomiting, fever Duration is up to 8 days	Cultural methods followed by confirmatory biochemical tests[128, 129] ELISA[133] PCR[134]	Adequate cooking of seafood, rapid chilling of seafoods, prevent cross-contamination from raw seafoods to other foods and preparation surfaces

TABLE 55.1 *(Continued)*

Bacterial Foodborne Pathogens

Microorganism	Biochemical and Growth Characteristics	Sources/ Reservoirs	Vehicles	Estimated No. of Foodborne Cases Annually in USA[1]	Incubation Period, Symptoms and Duration	Detection Methods	Control/Prevention
Vibrio vulnificus	Gram negative, nonsporeforming, optimal growth at 37°C	Coastal and estuarine waters	Raw seafood, especially raw oysters	47	12 h to 3 days, Profuse diarrhea with blood in feces, fulminating septicemia, hypotension Duration is days to weeks	Cultural methods followed by confirmatory biochemical tests[128,129] ELISA[135] PCR[136]	Avoid eating raw seafood, especially raw oysters when have a history of liver disease or alcoholism
Aeromonas hydrophila	Gram negative, facultative anaerobe, nonsporeforming, oxidase positive, some strains are psychrotrophic (4°C) optimum growth at ca. 28°C	Aquatic environment, freshwater fish (especially Salmonids)	Untreated water	Very few	24 to 48 h Abdominal pain, vomiting, watery stools, mild fever Duration is days to weeks	Cultural methods followed by confirmatory biochemical tests[137, 138, 139] PCR[140, 141]	Avoid consumption of raw seafoods, avoid long-term storage of refrigerated foods, adequate cooking of foods, disinfection of drinking water
Plesiomonas shigelloides	Gram negative, facultative anaerobe, nonsporeforming, oxidase positive, some strains are psychrotrophic	Fresh and estuarine waters, fish, and shellfish	Fish, shellfish, oysters, shrimp and untreated water	Very few	1 to 2 days Abdominal pain, nausea, vomiting, diarrhea, chills, headache Duration is days to weeks	Cultural methods followed by confirmatory biochemical tests[137, 138]	Avoid consumption of raw seafoods, disinfection of drinking water
Listeria monocytogenes	Gram positive, facultative anaerobe, nonsporeforming, growth at 2°-45°C, optimal growth at 30°-35°C, growth in presence of 10% NaCl	Soil, sewage, vegetation, water, and feces of humans and animals	Raw milk, soft cheese, pâte, ready-to-eat cooked meat products (poultry, hot dogs) and cooked seafoods (smoked fish), and raw vegetables	2490	Few days to several weeks Flu-like symptoms such as fever, chills, headache Abdominal pain and diarrhea are present in some cases In pregnant women, spontaneous abortion and stillbirth Duration is days to weeks	Cultural methods followed by confirmatory biochemical tests[142, 143, 144] Immunoassay[145] PCR[146]	Proper sanitation of food processing equipment and environments, adequate cooking of meat and meat products, prevent recontamination of cooked products, proper reheating of cooked food, avoid drinking raw milk, avoid certain high risk foods (e.g., soft cheeses and pâtes) by pregnant women and immunocompromised individuals

Staphylococcus aureus (staphylococcal enterotoxin)	Gram positive, facultative anaerobe, nonsporeforming, coagulase positive, growth at 7°-48°C, optimal growth at ca. 37°C, toxin production at a_w of 0.86, toxin is heat stable (can withstand boiling for 1 h)	Humans (nose, throat and skin) and animals	Ham, chicken and egg salads, cream-filled pastries	185,000	2 to 6 h Abdominal cramps, nausea, vomiting, diarrhea, headache, chills, and dizziness Duration is up to 2 days	Cultural methods followed by confirmatory biochemical tests[147, 148] PCR[149, 150] Detection of toxin by microslide gel double diffusion test[151]	Good personal hygiene in food preparation and handling, adequate cooking of foods, proper refrigeration of cooked foods
Clostridium botulinum (botulinum neurotoxin)	Gram positive, obligate anaerobe, sporeforming, produce seven potent neurotoxins A-G (only A, B, E and rarely F associated with human illness), proteolytic strains grow at 10°-50°C, nonproteolytic strains can grow at 3.3°C, spores are resistant to normal cooking temperatures, and survive freezing and drying	Soil, dust, vegetation, animals, birds, insects, and marine and fresh water sediments and the intestinal tracts of fish (type E)	Beef, pork, fish, vegetables, and honey (infant botulism)	58	12 to 36 h, can range from few h to 8 days Very severe life threatening intoxication, headache, fixed and dilated pupils, vertigo, blurred or double vision, lack of muscle coordination, dry mouth, difficulty in breathing Gastrointestinal symptoms include abdominal pain, nausea, vomiting, constipation Duration is days to months (8 months)	Cultural methods followed by confirmatory biochemical tests[152] PCR[153, 154] Detection of toxin by mouse bioassay[155]	Boiling of foods will destroy toxin, adequate heat processing of home-canned foods, proper refrigeration of vacuum-packaged fresh or lightly cooked/smoked foods, acid-preserved foods should be below pH 4.6, discard swollen cans, avoid feeding honey to infants
Clostridium perfringens	Gram positive, anaerobe, sporeforming, optimum growth at 37°-47°C, grows slowly below 20°C	Soil, sewage, dust, vegetation, feces of humans and animals	Cooked meat and poultry, especially roast beef, turkey and gravies	249,000	8 to 24 h Abdominal pain and diarrhea Duration is 1 to 2 days	Cultural methods followed by confirmatory biochemical tests[68] Latex agglutination test[156] Colony hybridization assay[157] PCR[156]	Adequate cooking of foods; cooked food should be rapidly cooled (<5°C) or held hot (>60°C); proper refrigeration and adequate reheating of stored cooked foods
Bacillus cereus	Gram positive, facultative anaerobe, sporeforming, some strains can grow at 4°-6°C; optimum growth at 28°-37°C	Widely distributed in nature, soil, dust, vegetation	Cereals, fried rice, potatoes, cooked meat products, milk and dairy products, spices, dried foods	27,000	*Diarrheal syndrome* (toxic infection): 8 to 16 h Abdominal pain, watery diarrhea Duration is 24 to 36 h *Emetic syndrome* (preformed, heat stable toxin): 1 to 5 h Nausea, vomiting, malaise, sometimes diarrhea Duration is 24 to 36 h	Cultural methods followed by confirmatory biochemical tests[158] ELISA[159] PCR[160]	Adequate cooking of foods; cooked foods should be rapidly cooled (<5°C) or held hot (60°C); avoid leaving cooked foods at room temperature for long time

TABLE 55.1 *(Continued)*

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Microorganism	Biochemical and Growth Characteristics	Sources/ Reservoirs	Vehicles	Estimated No. of Foodborne Cases Annually in USA[1]	Incubation Period, Symptoms and Duration	Detection Methods	Control/Prevention
Brucella spp.	Gram negative, aerobe, nonsporeforming, optimal growth at 37°C	Cattle, sheep, pig, goat	Raw milk and products made from unpasteurized milk	780	Acute form: 3 to 21 days, infrequently months Pyrexia, profuse sweats, chills, constipation, weakness, malaise, body aches, joint pains, weight loss, anorexia Chronic form: several months Long history of fever, inertia, recurrent depression, sexual impotence, insomnia Duration is weeks	Cultural methods[161] ELISA[162, 163] PCR[163]	Vaccination of livestock against *Brucella* spp., avoid contact with infected animals, eradication of diseased animals; pasteurization of milk; avoid eating unpasteurized dairy products
Helicobacter pylori	Gram negative, microaerophile to anaerobe	Humans, cats	Untreated water, foodborne transmission of disease has not been proven	Unknown	Gastritis, dyspepsia, peptic ulcer, gastric carcinoma	Cultural methods[78] ELISA[76] PCR[77]	Avoid contact with infected animals, use of chlorinated water for cooking and drinking

associated outbreaks are reported each year, with 340 outbreak-associated confirmed cases reported in 1997.[2] A wide variety of foods, including undercooked ground beef, raw milk, roast beef, venison jerky, salami, yogurt, lettuce, unpasteurized apple juice, cantaloupe, alfalfa sprouts, and coleslaw, have been implicated as vehicles of *E. coli* O157:H7 infection.[3] In addition, outbreaks involving person-to-person and waterborne transmission have been reported.[3] Cattle have been identified as an important reservoir of *E. coli* O157:H7,[4,5] with undercooked ground beef being a major vehicle of foodborne outbreaks.[6] A survey performed by the National Animal Monitoring System of the U.S. Department of Agriculture revealed that 1.6% of feedlot cattle shed *E. coli* O157:H7 and 0.4% shed *E. coli* O157 nonmotile bacteria in their feces.[7] This is likely an underestimate of the actual percentage of *E. coli* O157:H7 harbored by cattle. The use of more sensitive isolation procedures will likely identify considerably higher carriage rates. *E. coli* O157:H7 localizes in cattle primarily in the rumen and colon, and is shed in feces.[8] *E. coli* O157:H7 can survive in bovine feces for many months,[9] hence potentially contaminating cattle, food, water, and the environment. During slaughter and subsequent processing operations, contamination of carcasses with *E. coli* O157:H7 from the digesta or feces of cattle can occur. Moreover, fruits and vegetables grown on soil fertilized with cattle manure or irrigated with water contaminated with cattle manure has the potential of being a vehicle of *E. coli* O157:H7.

Acidification is commonly used in food processing to control growth and survival of spoilage-causing and pathogenic microorganisms in foods. The U.S. Food and Drug Administration does not regard foods with pH ≤ 4.6 (high-acid foods) to be microbiologically hazardous. However, *E. coli* O157:H7 has been associated with outbreaks attributed to high-acid foods, including apple juice, mayonnaise, fermented sausage, and yogurt,[10] raising concerns about the safety of these foods. Several studies have revealed that many strains of *E. coli* O157:H7 are highly tolerant to acidic conditions, being able to survive for extended periods of time in synthetic gastric juice and in highly acidic foods.[10,11] Further, exposure of *E. coli* O157:H7 to mild or moderate acidic environments can induce an acid tolerance response, which enables the pathogen to survive extreme acidic conditions. For example, acid-adapted cells of *E. coli* O157:H7 survived longer in apple cider, fermented sausage, and hydrochloric acid than non-acid adapted cells.[12,13] However, *E. coli* O157:H7 is not unusually heat resistant[14] or salt tolerant[15] unless cells are preexposed to acid to become acid adapted. Acid-adapted *E. coli* O157:H7 cells have been determined to have increased heat tolerance.

In humans, three important manifestations of illness have been reported in *E. coli* O157:H7 infection. These include hemorrhagic colitis, hemolytic uremic syndrome, and thrombocytopenic purpura.[16] Two important factors attributed to the pathogenesis of *E. coli* O157:H7 include the ability of the pathogen to adhere to the intestinal mucosa of the host, and production of Shiga toxin I and/or Shiga toxin II.[16] Retrospective analysis of foods implicated in outbreaks of *E. coli* O157:H7 infection suggest a low infectious dose of the pathogen, probably less than a hundred cells.[17]

Salmonella Species

Salmonella spp. are facultatively anaerobic, gram-negative, rod-shaped bacteria belonging to the family *Enterobacteriaceae*. Members of the genus *Salmonella* have an optimum growth temperature of 37°C and utilize glucose with the production of acid and gas.[18] *Salmonella* spp. are widely distributed in nature. They colonize the intestinal tracts of humans, animals, birds, and reptiles, and are excreted in feces, which contaminate the environment, water, and foods.[19] Many food products, especially foods having contact with animal feces, including beef, pork, poultry, eggs, milk, fruits, and vegetables, have been associated

with outbreaks of salmonellosis.[20] *Salmonella* spp. can be divided into host-adapted serovars and those without any host preferences. Most of the foodborne serovars are in the latter group.

The ability of many strains of *Salmonella* to adapt to extreme environmental conditions emphasizes the potential risk of these microorganisms as foodborne pathogens. Although salmonellae optimally grow at 37°C, the genus *Salmonella* consists of strains which are capable of growth from 5° to 47°C.[21] *Salmonella* spp. can grow at pH values ranging from 4.5 to 7.0, with optimum growth observed near neutral pH.[19] Pre-exposure of *Salmonella* to mild acidic environments (pH 5.5 to 6.0) can induce in some strains an acid tolerance response, which enables the bacteria to survive for extended periods of time exposure to acidic and other adverse environmental conditions such as heat and low water activity.[22,23] However, most *Salmonella* spp. possess no unusual tolerance to salt and heat. A concentration of 3 to 4% NaCl can inhibit the growth of *Salmonella*.[24] Most salmonellae are sensitive to heat, hence ordinary pasteurization and cooking temperatures are capable of killing the pathogen.[25]

The most common species of *Salmonella* that cause foodborne salmonellosis in humans are *S. enterica* serovar Typhimurium and *S. enterica* serovar Enteritiditis.[26] A wide variety of foods, including beef, pork, milk, chicken, and turkey have been associated with outbreaks caused by *S.* Typhimurium. *S.* Enteritidis outbreaks, however, are most frequently associated with consumption of poultry products, especially eggs. During the period from 1985 to 1987, 77% of *S.* Enteritidis outbreaks in the U.S. was associated with Grade A shell eggs or foods containing eggs.[27] One of the major routes of *S.* Enteritidis contamination of intact eggs is through transovarian transmission of the pathogen to the yolk.[28]

S. enterica serovar Typhi is the causative agent of typhoid (enteric fever), a serious human disease. Typhoid fever has a long incubation period of 7 to 28 days, and is characterized by prolonged and spiking fever, abdominal pain, diarrhea, and headache.[18] The disease can be diagnosed by isolation of the pathogen from urine, blood, or stool specimens of affected individuals. *S. typhi* is an uncommon cause of foodborne illness in the U.S.

Campylobacter Species

The genus *Campylobacter* consists of 14 species, however, *C. jejuni* subsp. *jejuni* and *C. coli* are the dominant foodborne pathogens. *C. jejuni* is a slender, rod-shaped, microaerophilic bacterium that requires approximately 3 to 6% oxygen for growth. It can be differentiated from *C. coli* by its ability to hydrolyze hippurate.[29] *C. jejuni* is the most common bacterial agent causing diarrheal disease in humans in the U.S. and many other countries.[30] Many animals including poultry, swine, cattle, sheep, horses, and domestic pets harbor *C. jejuni* in their intestinal tracts, hence serving as reservoirs of human infection. Although a number of vehicles such as beef, pork, eggs, and untreated water have been implicated in outbreaks of campylobacter enteritis, chicken and unpasteurized milk are reported as the most commonly involved foods.[31] The organism does not survive well in the environment, being sensitive to drying, highly acidic conditions, and freezing. It is also readily killed in foods by adequate cooking.[32]

Usually campylobacter enteritis in humans is a self-limiting illness characterized by abdominal cramps, diarrhea, headache, and fever lasting up to four days. However, severe cases, involving bloody diarrhea and abdominal pain mimicking appendicitis, also occur.[29] Guillain-Barré syndrome (GBS) is an infrequent sequela to *Campylobacter* infection in humans. GBS is characterized by acute neuromuscular paralysis[32] and is estimated to occur in approximately one of every 1000 cases of campylobacter enteritis.[33] A few strains of *C. jejuni* reportedly produce a heat-labile enterotoxin similar to that produced by *Vibrio cholera*e and enterotoxigenic *E. coli*.[29] Some strains of *C. jejuni* and *C. coli* also can produce

a cytolethal distending toxin, which causes a rapid and specific cell cycle arrest in HeLa and Caco-2 cells.[30]

Shigella Species

The genus *Shigella* is divided into four major groups: *S. dysenteriae* (group A), *S. flexneri* (group B), *S. boydii* (group C), and *S. sonnei* (group D) based on the organism's somatic (O) antigen. Humans are the natural reservoir of *Shigella* spp. The fecal-oral route is the primary mode of transmission of shigellae, and proper personal hygiene and sanitary practices of cooks and food handlers can greatly reduce the occurrence of outbreaks of shigellosis. Most foodborne outbreaks of shigellosis are associated with ingestion of foods such as salads and water contaminated with human feces containing the pathogen. Shigellosis is characterized by diarrhea containing bloody mucus, which lasts one to two weeks. The infectious dose for *Shigella* infection is low. The ID_{50} of *S. flexneri* and *S. sonnei* in humans is approximately 5000 microorganisms, and that of *S. dysenteriae* is a few hundred cells; hence secondary transmission of *Shigella* by person-to-person contact frequently occurs in outbreaks of foodborne illness.

Yersinia enterocolitica

Swine have been identified as an important reservoir of *Yersinia enterocolitica*, in which the pathogen colonizes primarily the buccal cavity.[34] Although pork and pork products are considered to be the primary vehicles of *Y. enterocolitica*, a variety of other foods, including milk, beef, lamb, seafood, and vegetables, has been identified as vehicles of *Y. enterocolitica* infection.[35] One of the largest outbreaks of yersiniosis in the U.S. was associated with milk.[36] Water has also been a vehicle of several outbreaks of *Y. enterocolitica* infection.[36] Surveys have revealed that *Y. enterocolitica* is frequently present in foods, having been isolated from 11% of sandwiches, 15% of chilled foods, and 22% of raw milk in Europe.[37] However, most isolates from foods of non-pork origin are nonpathogenic for humans. Although contaminated foods are a major source of *Y. enterocolitica* infection, contaminated blood used in transfusions has resulted in many cases of *Y. enterocolitica* sepsis.[38] Several serovars of pathogenic *Y. enterocolitica* have been reported, which include O:3, O:5, O:8, and O:9,[39] with serovar 0:8 being common in the U.S.[40]

An unusual characteristic of *Y. enterocolitica* that influences food safety is its ability to grow at low temperatures, even as low as –1°C.[41] Several studies have revealed growth of *Y. enterocolitica* in foods stored at refrigeration temperature. *Y. enterocolitica* grew on pork, chicken, and beef at 0 to 1°C.[42,43] The ability of *Y. enterocolitica* to grow well at refrigeration temperature has been exploited for isolating the pathogen from foods, water, and stool specimens. Such samples are incubated at 4 to 8°C in an enrichment broth for several days to selectively culture *Y. enterocolitica* based on its psychrotrophic nature.

Vibrio Species

The genus *Vibrio* consists of 28 species, of which *V. parahaemolyticus*, *V. vulnificus*, and *V. cholerae* are the most important foodborne pathogens. Vibrios are associated with estuarine and marine waters, and their populations in surface waters and in seafoods are higher during the warm than cold months of the year.[44] *V. parahaemolyticus* is present in coastal waters of the U.S. and the world. A survey by the U.S. Food and Drug Administration revealed that 86% of 635 seafood samples contained *V. parahaemolyticus*, being isolated from clams, oysters, lobsters, scallops, shrimp, fish, and shellfish.[44] An important virulence

characteristic of pathogenic strains of *V. parahemolyticus* is their ability to produce a thermostable hemolysin (Kanagawa hemolysin).[45] Studies in humans on the infectious dose of pathogenic *V. parahemolyticus* strains revealed that ingestion of approximately 10^5 to 10^7 organisms can cause gastroenteritis.[44]

V. cholerae serovars O1 and O139, the causative agents of cholera in humans, are a part of the normal estuarine microflora, and foods such as raw fish, mussels, oysters, and clams have been associated with outbreaks of cholera.[46] Infected humans can serve as short-term carriers, shedding the pathogen in feces. Cholera is characterized by profuse diarrhea, potentially fatal in severe cases, and often described as "rice water" diarrhea due to the presence of prolific amounts of mucus in the stools. Gastroenteritis caused by non-O1 and non-O139 serovars of *V. cholerae* is usually mild in nature.

V. vulnificus is the most serious of the vibrios and is responsible for most of the seafood-associated deaths in the U.S., especially in Florida.[44] Although a number of seafoods has been associated with *V. vulnificus* infection, raw oysters are the most common vehicle associated with cases of illness.[47] This pathogen causes a fulminating septicemia with a 40 to 50% mortality rate.

Aeromonas hydrophila

Although *Aeromonas* species have been recognized as pathogens of cold-blooded animals, their potential to cause human infections, especially foodborne illness, received attention only recently. *A. hydrophila* has been isolated from drinking water, fresh and saline waters, and sewage.[48] It also has been isolated from a variety of foods such as fish, oyster, shellfish, raw milk, ground beef, chicken, and pork.[48] Although *A. hydrophila* is sensitive to highly acidic conditions and does not possess any unusual thermal resistance, some strains are psychrotrophic and grow at refrigeration temperature.[49] *A. hydrophila* can grow on a variety of refrigerated foods, including pork, asparagus, cauliflower, and broccoli.[50,51] However, considering the widespread occurrence of *A. hydrophila* in water and food and its relatively infrequent association with human illness, it is likely that most strains of this bacterium are not pathogenic for humans. *A. hydrophila* infection in humans is characterized by watery diarrhea and mild fever. Virulent strains of *A. hydrophila* produce a 52-kDa polypeptide, which possesses enterotoxic, cytotoxic, and hemolytic activities.[52]

Plesiomonas shigelloides

P. shigelloides has been implicated in several cases of sporadic and epidemic gastroenteritis.[53] The pathogen is present in fresh and estuarine waters, and has been isolated from various aquatic animals.[49] Seafoods such as fish, crabs, and oysters have been associated with cases of *P. shigelloides* infection. The most common symptoms of *P. shigelloides* infection include abdominal pain, nausea, chills, fever, and diarrhea. Potential virulence factors of *P. shigelloides* include cytotoxic enterotoxin, invasins, and β-hemolysin.[49]

Listeria monocytogenes

L. monocytogenes has emerged into a highly significant and fatal foodborne pathogen throughout the world, especially in the U.S. There is an estimated approximately 2500 cases of listeriosis annually in the U.S., with a mortality rate of ca. 25%.[1] A large outbreak of listeriosis involving more than 100 cases and associated with eating contaminated turkey frankfurters occurred during 1998-99.[54] During this period of time there were more

than 35 recalls of a number of different food products contaminated with listeriae.[54] *L. monocytogenes* is widespread in nature, occurring in soil, vegetation, and untreated water. Humans and a wide variety of farm animals, including cattle, sheep, goat, pig, and poultry, are known sources of *L. monocytogenes*.[55] *L. monocytogenes* also occurs frequently in food processing facilities, especially in moist areas such as floor drains, floors, and processing equipment.[56] *L. monocytogenes* can also grow in biofilms attached to a variety of processing plant surfaces such as stainless steel, glass, and rubber.[57]

A wide spectrum of foods, including milk, cheese, beef, pork, chicken, seafoods, fruits, and vegetables, has been identified as vehicles of *L. monocytogenes*.[55] However, ready-to-eat cooked foods such as low-acid soft cheese, pâtes, and cooked poultry meat which can support the growth of listeriae to large populations (>10^6 cells per gram) when held at refrigeration temperature for several weeks, have been regarded as high-risk foods.[58,59] *L. monocytogenes* possesses several characteristics which enable the pathogen to successfully contaminate, survive, and grow in foods, thereby resulting in outbreaks. These traits include an ability to grow at refrigeration temperature and in a medium with minimal nutrients, ability to survive in acidic conditions, e.g., pH 4.2, ability to tolerate up to 10% sodium chloride, ability to survive incomplete cooking or subminimal pasteurization treatments, and the ability to survive in biofilms on equipment in food processing plants and resist superficial cleaning and disinfection treatments.[54]

Human listeriosis is an uncommon illness with a high mortality rate. Clinical manifestations range from mild influenza-like symptoms to meningitis and meningoencephalitis. Pregnant females infected with the pathogen may not present symptoms of illness or may exhibit only mild influenza-like symptoms. However, spontaneous abortion, premature birth, or stillbirth are frequent sequela to listeriosis in pregnant females.[59] Although the infective dose of *L. monocytogenes* is not known, published reports indicate that it is likely more than 100 CFU per gram of food.[59] However, the infective dose depends on the age, condition of health, and immunological status of the host. Important virulence factors of *L. monocytogenes* include intracellular invasin and production of listeriolysin O.[60]

Staphylococcus aureus

Pre-formed, heat stable enterotoxin that can resist boiling for several minutes is the agent responsible for staphylococcal food poisoning. Humans are the principal reservoir of *S. aureus* strains involved in outbreaks of foodborne illness. Colonized humans can be long-term carriers of *S. aureus*, and thereby contaminate foods and other humans.[61] The organism commonly resides in the throat and nasal cavity, and on the skin, especially in boils and carbuncles.[61] Protein-rich foods such as ham, poultry, fish, dairy products, custards, cream-filled bakery products, and salads containing cooked meat, chicken, and potatoes are the vehicles most frequently associated with *S. aureus* food poisoning.[62] *S. aureus* is usually overgrown by competing bacterial flora in raw foods; hence raw foods are not typical vehicles of staphylococcal food poisoning. Cooking eliminates most of the normal bacterial flora of raw foods, thereby enabling the growth of *S. aureus*, which can be introduced by infected cooks and food handlers into foods after cooking. The incubation period of staphylococcal food poisoning is very short, with symptoms observed within two to six hours after eating toxin-contaminated food. Symptoms include nausea, vomiting, diarrhea, and abdominal pain.

S. aureus can grow within a wide range of pH values from 4 to 9.3, with optimum growth occurring at pH 6 to 7. *S. aureus* has an exceptional tolerance to sodium chloride, being able to grow in foods in the presence of 7 to 10% NaCl, with some strains tolerating up to 20% NaCl.[62] *S. aureus* has the unique ability to grow at a water activity as low as 0.83

to 0.86, which is unusual for a nonhalophilic bacterium.[63] *S. aureus* produces nine different enterotoxins which are quite heat resistant, losing their serological activity at 121°C but not at 100°C for several minutes.[63]

Clostridium botulinum

Foodborne botulism is an intoxication caused by ingestion of foods containing pre-formed botulinal toxin, which is produced by *C. botulinum* under anaerobic conditions. There are seven types of *C. botulinum* (A, B, C, D, E, F, and G) classified on the basis of the antigenic specificity of the neurotoxin they produce.[64] The organism is present in soil, vegetation, and sedimentation under water. Type A strains are proteolytic, whereas type E strains are nonproteolytic.[65] Another classification divides *C. botulinum* into four groups: group 1 (type A strains and proteolytic strains of types B and F), group II (type E strains and nonproteolytic strains of B and F), group III (type C and D strains), and group IV (type G strains). Types A, B, E, and F are associated with botulism in humans. Type A *C. botulinum* occurs frequently in soils of the western U.S., whereas type B strains are more often present in the eastern states and in Europe.[65] Type E strains are largely associated with aquatic environments and fish. Foods most often associated with cases of botulism include fish, meat, honey, and home-canned vegetables.[64] Type A cases of botulism in the U.S. are frequently associated with temperature-abused, home-prepared foods. Proteolytic type A, B, and F strains produce heat-resistant spores, which pose a safety concern in low-acid canned foods. In contrast, nonproteolytic type B, E, and F strains produce heat-labile spores, which are of concern in pasteurized or unheated foods.[65] The minimum pH for growth of group I and group II strains is 4.6 and 5, respectively.[64] Group I strains can grow at a minimum water activity of 0.94, whereas group II strains do not grow below a water activity of 0.97.[66] The proteolytic strains of *C. botulinum* are generally more resistant to heat than nonproteolytic strains.

Clostridium perfringens

C. perfringens strains are grouped into five types: A, B, C, D, and E, based on the type(s) of toxin(s) produced. *C. perfringens* foodborne illness is almost exclusively associated with type A isolates of *C. perfringens. C. perfringens* is commonly present in soil, dust, water, and in the intestinal tracts of humans and animals.[67] It is frequently present in foods; about 50% of raw or frozen meat and poultry contain *C. perfringens*.[68] Spores produced by *C. perfringens* are quite heat resistant, and can survive boiling for up to one hour.[68] *C. perfringens* spores can survive in cooked foods, and if not properly cooled before refrigerated storage, the spores will germinate and vegetative cells can grow to large populations during holding at growth temperatures. Large populations of *C. perfringens* cells ($>10^6$/g) ingested with contaminated food will enter the small intestine, multiply, and sporulate. During sporulation in the small intestine *C. perfringens* enterotoxin is produced, which induces a diarrheal response. Although vegetative cells of *C. perfringens* are sensitive to cold temperature and freezing, spores tolerate cold temperature well and can survive in refrigerated foods.

Bacillus cereus

B. cereus is a spore-forming pathogen present in soil and on vegetation. It is frequently isolated from foods such as meat, spices, vegetables, dairy products, and cereal grains,

especially fried rice.[69] There are two types of foodborne illness caused by *B. cereus*, i.e., a diarrheagenic illness and an emetic syndrome.[70] The diarrheal syndrome is usually mild and is characterized by abdominal cramps, nausea, and watery stools. Types of foods implicated in outbreaks of diarrheal syndrome include cereal food products containing corn and corn starch, mashed potatoes, vegetables, milk, and cooked meat products. The emetic syndrome is more severe and acute in nature, characterized by severe vomiting. Refried or rewarmed boiled rice dishes are frequently implicated in outbreaks of emetic syndrome.[71] The dose of *B. cereus* required to produce diarrheal illness is estimated at more than 10^5 cells/g.[72]

Brucella Species

Brucella spp. are pathogens in many animals, causing sterility and abortion. In humans, *Brucella* is the etiologic agent of undulant fever. The genus *Brucella* consists of six species, of which those of principal concern are *B. abortus*, *B. suis*, and *B. melitensis*.[73] *B. abortus* causes disease in cattle, *B. suis* in swine, and *B. melitensis* is the primary pathogen of sheep. *B. melitensis* is the most pathogenic species for humans. Human brucellosis is primarily an occupational disease of veterinarians and meat industry workers. Brucellosis can be transmitted by aerosols and dust. Foodborne brucellosis can be transmitted to humans by consumption of meat and milk products from infected farm animals. The most common food vehicle of brucellosis for humans is unpasteurized milk.[73] Meat is a less common source of foodborne brucellosis because the organisms are destroyed by cooking.

Helicobacter pylori

H. pylori is a human pathogen causing chronic gastritis, gastric ulcer, and gastric carcinoma.[74,75] Although, humans are the primary host of *H. pylori*, the bacterium has been isolated from cats.[58] *H. pylori* does not survive well outside its host, but it has been detected in water and vegetables.[76,77] A study on the effect of environmental and substrate factors on the growth of *H. pylori* indicated that the pathogen likely lacks the ability to grow in most foods.[78] However, *H. pylori* may survive for long periods in low-acid environments under refrigerated conditions. Presently, the mode of transmission of *H. pylori* in human infection has not been elucidated, but contaminated water and food are considered potential vehicles.

Viral Foodborne Pathogens (Table 55.2)

Recent estimates by the Centers for Disease Control and Prevention of the incidence of foodborne illness in the U.S. indicate that viruses are responsible for approximately 67% of the total foodborne illnesses of known etiology annually.[1] Viruses are obligate intracellular microorganisms, and most foodborne viruses contain RNA rather than DNA. Since viruses require a host for multiplication, they cannot grow in foods. Foodborne viruses are generally enteric in nature, causing illness through ingestion of foods and water contaminated with human feces. Viruses disseminated through foods also can be spread by person-to-person contact. Hepatitis A virus, Norwalk-like viruses, and possibly rotavirus are among the most significant of the foodborne viruses.

TABLE 55.2

Viral Foodborne Pathogens

Microorganism	Significant Characteristics	Sources/ Reservoirs	Vehicles	Estimated No. of Foodborne Cases Annually in USA[121]	Incubation Period, Symptoms and Duration	Detection Methods	Control/Prevention
Hepatitis A virus	Single-stranded RNA virus, spherical in shape, remains viable for long periods of time in foods stored at refrigeration temperature, virus multiplies in the gut epitheliums before being carried by blood to the liver. Virus is shed in feces before symptoms of liver damage become apparent	Humans, sewage-polluted waters	Raw or undercooked shellfish and seafoods harvested from sewage-polluted water, ready-to-eat foods such as salads prepared by infected food handler	4170	15 to 45 days, usually ca. 25 days Loss of appetite, nausea, abdominal pain, fever, jaundice, dark urine, pale stools Duration is a few weeks to months	Cultural methods[164, 165] Enzyme immunoassay[166] PCR[167, 168]	Avoid consumption of raw seafoods, disinfection of drinking water, good personal hygiene and food handling practices, vaccination of professional food handlers, safe sewage disposal
Norwalk-like viruses (small round structured viruses; SRSV)	Single-stranded RNA virus, spherical in shape, does not multiply in any known laboratory host	Humans, sewage-polluted waters	Raw or undercooked shellfish and seafoods harvested from sewage polluted water, drinking water	9,200,000	1 to 2 days Loss of appetite, nausea, abdominal pain, diarrhea, vomiting, headache Duration is 2 days	Enzyme immunoassay[169] PCR[168]	Avoid consumption of raw seafoods, disinfection of drinking water, good personal hygiene and food handling practices, hygienic sewage disposal, treatment of wastewater used for irrigation
Rotavirus	Double-stranded RNA virus, icosahedral in shape	Humans	To be determined	39,000	1 to 3 days Vomiting, abdominal pain followed by watery diarrhea Duration is 6 to 8 days	Cultural methods[164, 165] ELISA[170] PCR[171]	Avoid consumption of raw seafoods Avoid drinking of untreated water, Good personal hygiene

Hepatitis A virus

Hepatitis A virus is a member of the family *Picornaviridae* and is transmitted by the fecal-oral route. Raw shellfish harvested from waters contaminated by human sewage is among the foods most frequently associated with outbreaks of hepatitis A virus.[79] Hepatitis A virus is more resistant to heat and drying than other picornaviruses.[79] The incubation period for onset of symptoms of hepatitis A infection ranges from 15 to 45 days, and symptoms include nausea, abdominal pain, jaundice, and fever. The virus is shed in feces by infected humans many days before the onset of symptoms, indicating the importance of good personal hygienic practices of cooks and food handlers who could otherwise contaminate food during the period of asymptomatic fecal shedding.

Norwalk-like viruses

Norwalk-like viruses belong to the family *Calciviridae*, and are often referred to as small, round structured viruses. Viruses of this type are believed to be the most common cause of foodborne viral diseases in the U.S. Raw or undercooked shellfish and other seafoods are common vehicles of Norwalk-like viruses. The incubation period of infection ranges from 24 to 48 h, and symptoms include nausea, vomiting, and diarrhea. Infected humans shed the virus in feces for up to a week after symptoms have subsided. Although little information is available on the stability of these viruses in foods, qualitative studies in human volunteers indicate that the viruses are infective for up to 3 h when exposed to a medium at pH 2.2 at room temperature or for 60 minutes at pH 7 at 60°C.[80]

Rotavirus

Rotavirus is the most common cause of diarrhea in children worldwide, especially in developing countries. In the U.S., there are an estimated 3.9 million cases of rotavirus diarrhea each year; however, only 39,000 cases are estimated to be acquired through contaminated foods.[1] Rotavirus infection has an incubation period of one to three days, and is characterized by fever, vomiting, and diarrhea. The virus is shed in the feces of infected humans and can survive on vegetables at 4° or 20°C for many days.[81] It also has been shown to survive the process of making soft cheese.[81]

Fungal Foodborne Pathogens (Table 55.3)

Molds are widely distributed in nature and are an integral part of the microflora of foods. Although molds are major spoilage agents of many foods, many molds also produce mycotoxins of which some are carcinogenic and mutagenic. Mycotoxins are secondary metabolites produced by molds usually at the end of their exponential phase of growth. Some of the principal species of molds which produce mycotoxins in foods are described here.

Aspergillus Species

A. flavus and *A. parasiticus* are the most important toxigenic foodborne aspergilli. A wide variety of foods such as nuts, corn, oil seeds, and sorghum are potential vehicles of these

TABLE 55.3

Fungal Foodborne Pathogens

Microorganism/ Toxin	Significant Characteristics	Sources/ Reservoirs of Fungi	Vehicles of Toxins	Toxic Effects	Detection Methods	Control/ Prevention
Aspergillus parasiticus and *Aspergillus flavus*/ Aflatoxin	Growth at 10°-43°C, optimal growth at 32°C, produces aflatoxins at 12°-40°C, growth at pH 3 to 11	Environment, soil, vegetation	Corn, peanuts, cottonseed	Effects of aflatoxin in animals: Acute: hemorrhage in the gastrointestinal tract, liver damage, death Chronic: cirrhosis of liver, liver tumors, immunosuppression	Cultural methods[172, 173, 174] ELISA[175] PCR[176]	Proper storage of cereal products, detoxification of mycotoxins in cereal products by treatment with hydrogen peroxide, ammonia
Penicillium expansum/ Patulin; *Penicillium citrinum*/Citrinin	*P. expansum* is psychrotrophic, capable of growth at -2° to -3°C, optimal growth at 25°C	Environment, soil, vegetation	*P. expansum*: Fruits, especially apples and pears *P. citrinum*: Cereals, especially rice, wheat, corn	Effects of patulin: Gastrointestinal, neurological, and immunological effects in animals Citrinin: fatty degeneration and necrosis of kidneys of pigs and dogs; significance in human health is unresolved	Cultural methods[173, 174, 177] Gas chromatography[178]	Avoid consumption of rotten apples and pears, proper storage of cereal products
Fusarium graminearum/ Deoxynivalernol, nivalenol, zearalenone	Growth at 5°C but not at 37°C, optimal growth at 25°C	Environment, soil, vegetation	Cereals, especially wheat, barley and corn	Effects of deoxynivolenol: nausea, vomiting, abdominal pain, diarrhea, headache, fever, chills, throat irritation	Cultural methods followed by morphology[179, 180] PCR[181]	Proper storage of cereal products

aspergilli. *A. flavus* and *A. parasiticus* produce aflatoxins, which are difuranocoumarin derivatives.[82] The common types of aflatoxins produced are B_1, B_2, G_1, and G_2.[83] Aflatoxicosis in animals can be acute or chronic. Acute cases are characterized by severe liver damage, whereas liver cirrhosis, liver cancer, and teratogenesis occur in chronic toxicity. Chronic intake of aflatoxins in animals can lead to poor feed conversion and low weight gain.

Penicillium Species

The genus *Penicillium* consists of more than 150 species, of which nearly 100 produce known toxins. Three important foodborne toxigenic *Penicillium* species include *P. verrucosum*, *P. expansum*, and *P. citrinum*. *P. verrucosum* is present on grains grown in temperate zones, and is commonly associated with Scandanavian barley and wheat.[84] *P. verrucosum* produces Ochratoxin A, which has immunosuppressive and potential carcinogenic properties.[84] Ochratoxin A also has been associated with nephritis in pigs in Scandanavia.[85] *P. expansum*, which is frequently associated with fresh fruits, produces patulin, a toxin that produces immunological, neurological, and gastrointestinal toxic effects in animal models. *P. expansum* is commonly present in rotten apples and pears, and to a lesser extent in cereals. An unusual characteristic of *P. expansum* is its ability to grow at low temperature, i.e., –2° to –3°C.[84] *P. citrinin* is a widely occurring mold commonly present on rice, wheat, and corn. *P. citrinin* produces the metabolite citrinin. Although the toxicological effect of citrinin in humans is not known, it has been reported to cause renal toxicity in pigs and cats.[86]

Fusarium graminearum

F. graminearum is a toxigenic mold commonly present in soil and on cereals such as wheat and corn. It produces a number of mycotoxins, including deoxynevalenol and zearalenone.[87] Ingestion of foods containing deoxynevalenol produces illness termed Scabby grain intoxication, which is characterized by anorexia, nausea, vomiting, diarrhea, dizziness, and convulsions. Foods most frequently implicated as vehicles of deoxynevalenol include cereal grains, wheat, barley, and noodles.

Parasitic Foodborne Pathogens (Table 55.4)

Foods can be vehicles of several types of parasites, including protozoa, roundworms, and flatworms. Although foodborne transmission of parasites such as *Trichinella spiralis* and *Taenia solium* has been known for many years, the foodborne disease potential of many protozoan parasites such as *Cryptosporidium* and *Cyclospora* has only recently been recognized. Unlike bacteria, parasites do not multiply in foods. Moreover, parasites need at least one specific host to complete their life cycle. Many of the well-recognized parasites that can be transmitted to humans through foods are listed below.

Giardia lamblia

G. lamblia is a flagellated protozoan parasite that colonizes the intestinal tract of humans and animals. It is commonly present in lakes, rivers, and stagnated waters. The life cycle of *G. lamblia* includes flagellated trophozoites, which become pear-shaped cysts.[88] The

TABLE 55.4

Parasitic Foodborne Pathogens

Parasite	Significant Characteristics	Sources/ Reservoirs	Vehicles	Estimated No. of Foodborne Cases Annually in USA[1]	Incubation Period, Symptoms and Duration	Detection Methods	Control/ Prevention
Giardia lamblia	Flagellate protozoa, produces oval-shaped cysts ranging from 8 to 20 µm in length and 5 to 12 µm in width, cysts contain four nuclei and are resistant to chlorination used to disinfect water	Humans, animals, especially beavers and muskrats, water	Drinking water, raw fruits and vegetables contaminated with cysts, ready-to-eat foods such as salads contaminated by infected food handlers	200,000	4 to 25 days, usually 7 to 10 days Abdominal cramps, nausea, abdominal distension, diarrhea which can be chronic and relapsing, fatigue, weight loss, anorexia Duration is weeks to years	Immuno-fluorescence[182] Immunochromato-graphy[183] PCR[184]	Adequate cooking of foods, filtration of drinking water, good personal hygiene and food handling practices
Entamoeba histolytica	Amoeboid protozoa, anaerobe survives in environment in crypted form, cysts remain viable in feces for several days and in soil for at least 8 days at 30°C and for more than 1 month at 10°C, relatively resistant to chlorine	Humans, dogs, rats	Foods and water contaminated with feces or irrigation water	Unknown	2 to 4 weeks Abdominal pain, fever, vomiting, diarrhea containing blood and mucus, weight loss Duration is weeks to months	Microscopic examination ELISA[185] PCR[185, 186]	Good personal hygiene and food handling practices, adequate cooking of foods, filtration of water, hygienic disposal of sewage water, treatment of irrigation water
Cryptosporidium parvum	Obligate intracellular coccidian parasite, oocysts are spherical to oval in shape with an average size of 4.5 to 5.0 µm, oocysts are resistant to chlorination used to disinfect water	Humans, wild and domestic animals, especially calves	Contaminated drinking and recreational water, raw milk from infected cattle, fresh vegetables and other foods contaminated with feces from infected humans and animals	30,000	2 to 14 days Profuse, watery diarrhea, abdominal pain, nausea, vomiting Duration is few days to 3 weeks	Immunofluorescence assay[187] PCR[188]	Thorough cooking of food, avoid contact with infected animals, filtration of drinking water, good personal hygiene and food handling practices
Cyclospora cayetanesis	Obligate intracellular coccidian parasite, oocysts are spherical in shape with an average size of 8 to 10 µm	Humans	Water, fruits and vegetables contaminated with oocysts	14,600	1 week Watery diarrhea, abdominal pain, nausea, vomiting, anorexia, myalgia, weight loss Duration is a few days to 1 month	Staining and microscopic examination[189] PCR[190]	Good personal hygiene, filtration of drinking water

Toxoplasma gondii	Obligate intracellular coccidian protozoa	Cats, farm animals, transplacental transmission from infected mother to fetus	Raw or undercooked meat, raw goat milk, raw vegetables	112,500	5 to 23 days Fever, rash, headache, muscle pain, swelling of lymph nodes; transplacental infection may cause abortion Duration is variable	Cell culture and mouse inoculation[191] Immunoassay[192] PCR[191]	Prevent environmental contamination with cat feces, avoid consumption of raw meat and milk, safe disposal of cat feces, wash hands after contact with cats
Trichinella spiralis	Nematode with no free living stage in the life cycle, adult female worms are 3 to 4 mm in length, transmissible form is larval cyst which can occur in pork muscle	Wild and domestic animals, especially swine and horses	Raw or undercooked meat of animals containing encysted larvae such as swine or horses	50	Initial symptoms: 24 to 72 h, Systemic symptoms: 8 to 21 days Initial phase: abdominal pain, fever, nausea, vomiting, diarrhea Systemic phase: periorbital oedema, eosinophilia, myalgia, difficulty in breathing, thirst, profuse sweating, chills, weakness, prostration Duration is 2 weeks to 3 months	Microscopic examination ELISA[193] PCR[194]	Adequate cooking of meat, freezing of meat at -15°C for 30 days or at -35°C, preventing trichinosis in pigs by not feeding swine garbage containing infected meat
Anisakis spp.	Nematode, slender threadlike parasite measuring 1.5 to 1.6 cm in length and 0.1 cm in diameter	Sea mammals	Some undercooked salt water fish, sushi, herring, sashimi, ceviche	Unknown	4 to 12 h Epigastric pain, nausea, vomiting, sometimes hematemesis Duration is variable	ELISA[195] PCR[196]	Adequate cooking of saltwater fish, freezing fish at -23°C for 7 days
Taenia solium *Taenia saginata*	Tapeworm, dependent on the digestive system of the host for nutrition	Humans, cattle, swine	Raw or undercooked beef or pork	Unknown	Few days to >10 years Nausea, epigastric pain, nervousness, insomnia, anorexia, weight loss, digestive disturbances, weakness, dizziness Duration is weeks to months	Detection of eggs or proglottids in feces ELISA[197] PCR[198]	Adequate cooking of beef and pork, proper disposal of sewage and human wastes, freezing of meat at -10°C for 2 weeks
Diphyllobothrium latum	Largest human tapeworm	Saltwater fish, humans	Raw or undercooked saltwater fish	Unknown	Epigastric pain, nausea, abdominal pain, diarrhea, weakness, pernicious anemia Duration is months to years	Detection of eggs in feces	Adequate cooking of fish, proper disposal of sewage and human waste

cysts contaminate water or food through feces of infected animals or humans. Following ingestion of cyst-contaminated water or food, the trophozoites reach the small intestine, where they undergo excystation and multiply by binary fission. New trophozoites subsequently become cysts in the distal small intestine, and the encysted trophozoites are shed in the feces. The symptoms of giardiasis include abdominal pain, abdominal distension, nausea, vomiting, and diarrhea. Although water and foods contaminated with cysts are primary vehicles of giardiasis, little is known about the survival characteristics of the cysts in foods. In most cases of foodborne transmission, infected food handlers transfer the cysts to foods they prepare.

Entamoeba histolytica

E. histolytica is a protozoan parasite that causes amoebiasis or amoebic dysentery in humans. Although the parasite survives in the environment and water, humans are the principal source of amoebiasis. In humans, cysts containing the trophozites are released, which in turn multiply and are subsequently excreted in the feces as cysts.[89] Foods and water contaminated with the cysts transmit the disease. Since the fecal-oral route is the principal route of transmission of amoebiasis, personal hygiene of infected food handlers plays a critical role in preventing foodborne amoebiasis. Human amoebiasis can occur in two forms: intestinal amoebiasis and amoebic liver abscess, which is usually a sequela to the intestinal form. Intestinal amoebiasis is characterized by abdominal pain, vomiting, and watery diarrhea containing mucus and blood. Symptoms of the hepatic form of amoebiasis include wasting, painful and enlarged liver, weight loss, and anemia.

Cryptosporidium parvum

C. parvum is a protozoan parasite that infects a wide range of animals and humans. *C. parvum* is monoexenous in its life cycle, requiring only one host for its development.[88] Infected hosts shed in their feces oocysts of the parasite, subsequently contaminating the environment, food, and water. The life cycle of *C. parvum* can be summarized as follows.[88] Upon ingestion of contaminated water or food, or by inhalation of oocysts, sporozoites are released by excystation of oocysts into the gastrointestinal or respiratory tract. The sporozoites enter the epithelial cells and develop into trophozoites, which in turn differentiate into type I and type II meronts. The merozoites from type I meronts invade new tissues and develop into trophozoites to continue the life cycle. The merozoites from type II meronts invade infected cells and undergo sexual multiplication to give rise to male and female gametes. The zygotes resulting from fertilized gametes become infectious by sporulation, and the sporulated oocysts are excreted in feces.

Cryptosporidiosis is a self-limiting disease with an incubation period of one to two weeks, and is characterized by profuse, watery diarrhea, abdominal pain, vomiting, and low-grade fever. Water is the most common source of *C. parvum* for human infections.[58] Oocysts of the pathogen have been detected in fresh vegetables, raw milk, sausage, and apple cider.[58] Infected food handlers can also transfer the oocysts to foods.[90, 91] C. *parvum* oocysts are sensitive to freezing and freeze-drying. The oocysts lose infectivity in distilled water stored at 4°C.[92] However, the oocysts are quite resistant to chlorine; no loss in infectivity was observed in water containing 1 to 3% chlorine for up to 18 hours.[93] However, the oocysts are sensitive to ozone, losing more than 90% infectivity in the presence of 1 ppm ozone for 5 minutes.[94]

Cyclospora cayetanensis

C. cayetanensis is an emerging foodborne, protozoan pathogen, especially in the U.S. The parasite was implicated in several foodborne outbreaks in the U.S. during 1996 and 1997.[95] *C. cayetanensis* is spread through infected feces and is transmitted to humans by the fecal-oral route. Water and foods, especially fruits and vegetables containing oocysts, are common vehicles of human infection. The symptoms of *C. cayetanensis* infection in humans include watery diarrhea, nausea, abdominal pain, vomiting, and weight loss. Presently, very little information is available on the effects of heat, freezing, and disinfection agents on *Cyclospora* oocysts. Preliminary studies revealed that exposure of oocysts to –20°C for 24 h or 60°C for 1 h prevented oocysts from sporulating. Exposing oocysts to 4° or 37°C for 14 days delayed sporulation.[96]

Toxoplasma gondii

T. gondii is an obligate intracellular protozoan parasite for which cats are the definitive hosts. In the intestines of cats, the parasite undergoes sexual reproduction to form oocysts, which are excreted in feces.[97] The oocysts undergo maturation and survive in the environment for months. Toxoplasmosis in humans results following ingestion of food or water contaminated with oocysts. Transmission also occurs from an infected pregnant mother to child by transplacental transmission.[89] Symptoms in healthy adults are usually mild, and include rash, headache, muscle pain, and swelling of lymph nodes. The oocysts are sensitive to both heat and cold;[98] hence the cysts are killed in properly cooked foods.[99]

Trichinella spiralis

T. spiralis is a roundworm that primarily infects wild and domestic animals, especially pigs. Humans contract trichinosis by consumption of raw or undercooked meat containing larvae of the parasite. Pigs are infected by consuming uncooked scraps of infected pork. The encysted larvae upon ingestion are liberated from the cyst in the intestine, where they sexually mature.[100] The mature male and female worms copulate in the lumen of the small intestine, giving rise to a new generation of larvae. The newly born larvae migrate to various tissues in the body. Those larvae that reach the striated muscles penetrate into the sarcolemma of the muscle fibers and develop to maturity as encapsulated cysts.[100] The larvae continue their life cycle when raw or undercooked meat, especially pork containing the larvae, is consumed by humans.

Anisakis Species

Anisakiasis in humans is caused by two foodborne roundworms. These include *A. simplex*, whose definitive host is whales, and *Pseudoterranova decipiens*, which primarily inhabits seals. The eggs of these roundworms are excreted in feces by their respective hosts. The eggs then undergo molting in suitable intermediate hosts and subsequently develop into larvae, which are ingested by fish.[101] Humans contract anisakiasis by consumption of raw or undercooked fish and seafoods containing the larvae. In noninvasive anisakiasis, the worms released from ingested foods migrate to the pharynx, resulting in "tingling throat syndrome."[101] The worms are ultimately expelled by coughing. In the invasive form of anisakiasis, the worms penetrate the intestinal mucosa, causing symptoms that include epigastric pain, nausea, vomiting, and diarrhea.

Taenia Species

The genus *Taenia* includes two meatborne pathogenic flat worms, *T. saginata* (beef tapeworm) and *T. solium* (pork tapeworm). The eggs of *T. saginata* survive in the environment, including on pastures, and are ingested by cattle in which they hatch into embryos.[100] The embryos migrate to skeletal muscles or the heart, and develop into larvae known as cysticercus bovis. Humans become infected by consuming raw or undercooked beef containing the larvae. Larvae that are released into the small intestine develop into mature, adult worms. The symptoms of *T. saginata* infection in humans include decreased appetite, headache, dizziness, diarrhea, and weight loss.

In the normal life cycle of *T. solium*, pigs serve as the intermediate host. Eggs ingested by pigs develop into embryos in the duodenum, penetrate the intestinal wall, migrate through the blood and the lymphatic system, and finally reach the skeletal muscles and myocardium, where they develop into larvae known as cysticercus cellulose. Humans consuming raw or undercooked pork are infected with the larvae, which develop into adult worms in the small intestine. The symptoms of *T. solium* infection in humans include discomfort, hunger pains, anorexia, and nervous disorders. Worms are passed in the feces. In the abnormal life cycle of *T. solium*, humans serve as intermediate hosts in which the larvae develop in striated muscles and in subcutaneous tissue.

Diphyllobothrium latum

D. latum is commonly referred to as the broad tapeworm because it is the largest human tapeworm.[101] Humans contract diphyllobothriasis by consuming raw or undercooked fish containing the larval forms called plerocercoids. Upon ingestion, the larvae develop into mature worms in the intestines. Eggs produced by mature worms are excreted in feces. If feces containing the eggs contaminate water, the eggs develop into free-swimming larvae called coricidia. Coricidia are ingested by crustaceans, where they develop into a juvenile stage known as procercoid. Following ingestion of infected crustaceans by fish, procercoids develop into plerocercoids to continue the life cycle. Diphyllobothriasis in humans is characterized by nausea, abdominal pain, diarrhea, weakness, and pernicious anemia.[101] Cases of diphyllobothriasis have been associated with eating raw salmon and sushi.

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