

Introduction to Food Microbiology

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Introduction

Food science is a discipline concerned with all aspects of food - beginning after harvesting, and ending with consumption by the consumer. It is considered one of the agricultural sciences, and it is a field which is entirely distinct from the field of nutrition. The field of food microbiology is a very broad one, encompassing the study of microorganisms which have both beneficial and deleterious effects on the quality and safety of raw and processed foods. It is important to understand the relationships among the various microorganisms making up the microflora of a food.

In fact food microbiologists are concerned with the practical implications of the microflora of the food and the food microorganisms that can cause spoilage of food and disease in humans. The primary tool of microbiologists is the ability to identify and quantitate food-borne microorganisms. However, the inherent inaccuracies in enumeration processes, and the natural variation found in all bacterial populations complicate the microbiologist's job. Moreover, they may be important from the aesthetic point of view. Of course, some useful bacteria may be important because they change the functional properties of food stuffs resulting in new tastes, odors or textures. Microorganisms in food include bacteria, molds, yeasts, algae, viruses, parasitic worms and protozoa. These organisms differ in size and shape and in their biochemical and cultural characteristics. The microorganisms described below are among the most important genera and species normally found in food products. Each microorganism has its own particular nutritional and environmental requirements.

Bacteria

Acinetobacter is a genus of Gram-negative bacteria belonging to the Gammaproteobacteria . Acinetobacter species are non-motile and oxidase-negative, and occur in pairs as observed under magnification. Young cultures show rod shaped morphology. They are strict aerobes that do not reduce nitrates. They are important soil and water organisms and are also found on many foods especially refrigerated fresh products. S A. baumannii is a frequent cause of nosocomial pneumonia, especially of late onset ventilator associated pneumonia. It can cause various other infections including skin and wound infections, bacteremia, and meningitis,

Bacillus cereus

Bacillus cereus is a thick long rod shaped Gram positive, catalase positive aerobic spore former and the organism is important in food borne illness. It is a normal inhabitant of soil and is isolated from a variety of foods. It is quite often a cause of diarrheal illness due to the consumption of desserts, meat, dishes, dairy products, rice, pasta etc that are cooked and kept at room temperature as it is thermotolerant. Some of the *B. cereus* strains are psychrotrophic as they grow at refrigeration temperature.

B. cereus is spread from soil and grass to cows udders and into the raw milk. It is also capable of establishing in cans. It is also capable of producing proteolytic and amylolytic enzymes and also phospholipase C (lecithinase). The production of these enzymes by these organisms can lead to the spoilage of foods. The diarrheal illness is caused by an enterotoxin produced during the vegetative growth of *B. cereus* in small intestine. The bacterium has a maximum growth temperature around 48°C to 50°C and pH range 4.9 to 9.3. Like other spores of mesophilic *Bacillus* species, spores of *B. cereus* are also resistant to heat and survive pasteurization temperature.

Bacillus subtilis

Bacillus subtilis, known also as the hay bacillus or grass bacillus, is a Gram-positive, catalase-positive bacterium commonly found in soil. A member of the genus *Bacillus*, *B. subtilis* is thin short rod-shaped, and has the ability to form a tough, protective endospore, allowing the organism to tolerate extreme environmental conditions. *B. subtilis* produces the proteolytic enzyme subtilisin. *B. subtilis* spores can survive the extreme heat during cooking. *B. subtilis* is responsible for causing ropiness a sticky, stringy consistency caused by bacterial production of long-chain polysaccharides in spoiled bread dough. A strain of *B. subtilis* formerly known as *Bacillus nattois* used in the commercial production of the Japanese food natto, as well as the similar Korean food cheonggukjang. It is used to produce amylase and also used to produce hyaluronic acid, which is useful in the joint-care sector in healthcare.

Carnobacterium

Carnobacterium is a genus of Gram-positive bacteria within the family Leuconostocaceae. *C. divergens* and *C. maltaromaticum* are found in the wild and in food products and can grow anaerobically. These species are not known to be pathogenic in humans but may cause disease in fish. The genus *Carnobacterium* contains nine species, but only *C. divergens* and *C. maltaromaticum* are frequently isolated from natural environments and foods. They are tolerant to freezing/thawing and high pressure and able to grow at low temperatures, anaerobically. They metabolize arginine and various carbohydrates, including chitin, and this may improve their survival in the environment. *Carnobacterium divergens* and *C. maltaromaticum* have been extensively studied as protective cultures in order to inhibit growth of *Listeria monocytogenes* in fish and meat products. Several carnobacterial bacteriocins have been identified and described. Carnobacteria can spoil chilled foods, but spoilage activity shows intraspecies and interspecies variation. Their production of tyramine in foods is critical for susceptible individuals, but carnobacteria are not otherwise human pathogens.

Corynebacterium

Corynebacterium is a genus of Gram-positive rod-shaped bacteria. They are widely distributed in nature and are mostly innocuous. Some are useful in industrial settings such as *C. glutamicum*. Others can cause human disease. *C. diphtheriae*, for example, is the pathogen responsible for diphtheria. Some species are known for their pathogenic effects in humans and other animals. Perhaps the most notable one is *C. diphtheriae*, which acquires the capacity to produce diphtheria toxin only after interacting with a bacteriophage. Diphtheria toxin is a single, 60,000 molecular weight protein composed of two peptide chains, fragment A and fragment B, held together by a disulfide bond.

1.2.6 Clostridium perfringens

C. perfringens is a Gram-positive encapsulated anaerobic non-motile bacterium commonly found on meat and meat products. It has the ability to cause food borne disease. It is a toxin producing organism-produces *C. perfringens* enterotoxin and β -toxin that are active on the human GI tract. It multiplies very rapidly in food (doubling time < 10 min). Spores are resistant to radiation, desiccation and heat and thus survive in incompletely or inadequately cooked foods. However, it tolerates moderate exposure to air. Vegetative cells of *C. perfringens* are also somewhat heat tolerant as they have relatively high growth temperature (43°C -45 °C) and can often grow at 50°C. They are not tolerant to refrigeration and freezing. No growth occurs at 6 °C . *C. perfringens* is present in soil and the other natural environment.

1.2.7 Clostridium botulinum

C. botulinum produces the most potent toxin known. It is a Gram-positive anaerobic rod shaped bacterium. Oval endospores are formed in stationary phase cultures. There are seven types of *C. botulinum* (A to G) based on the serological specificity of the neurotoxin produced. Botulism is a rare but very serious disease. The ingestion of neurotoxin produced by the organism in foods can lead to death. However, the toxin (a protein) is easily inactivated by heat. The organism can grow at temperature ranging from 10-48°C with optimum growth temperature at 37°C. Spores are highly heat resistant. The outgrowth of spores is inhibited at pH < 4.6, NaCl > 10% or water activity < 0.94. Botulinum spores are probably the most radiation resistant spores of public health concern. Contamination of foods is through soil and sediments where they are commonly present. The organism grows under obligate anaerobic conditions and produces toxin in under processed (improper canning) low acid foods at ambient temperature.

Campylobacter

Campylobacter are Gram negative nonspore forming rods. *Campylobacter jejuni* is an important food borne pathogen. It is one of the many species within the genus *Campylobacter*. *Campylobacter* species *C. jejuni* and *C. coli* cause diarrhea in humans. The organism is heat sensitive (destroyed by milk pasteurization temperature). It is also sensitive to freezing. The organism belongs to the family *Campylobacteriaceae*. The organisms are curved, S-shaped, or spiral rods that may form spherical or

coccoids forms in old cultures or cultures exposed to air for prolonged periods. Most of the species are microaerophilic. It is oxidase and catalase positive and does not grow in the presence of 3.5% NaCl or at 25 °C or below. The incidences reported for gastro enteritis by this organism are as high as in case of Salmonella. The organism is commonly present in raw milk, poultry products, fresh meats, pork sausages and ground beef.

Erwinia

Erwinia is a genus of the family Enterobacteriaceae bacteria containing mostly plant pathogenic species. The organisms was named after the first phyto bacteriologist, Erwin Smith. It is a Gram negative bacterium related to E. coli, Shigella, Salmonella and Yersinia. It is primarily a rod-shaped bacterium. A well-known member of this genus is the species E. amylovora, which causes fire blight on apple, pear and other Rosaceous crops. Erwinia carotovora (also known as Pectobacterium carotovorum) is another species, which causes diseases in many plants. These species produce pectolytic enzymes that hydrolyze pectin between individual plant cells. . Decay caused by E. carotovora is often referred to as bacterial soft rot (BSR). Most plants or plant parts can resist invasion by the bacteria, unless some type of wound is present. High humidity and temperatures around 30°C favor development of decay.

Enterococcus

Enterococcus is a genus of lactic acid bacteria. Enterococci are Gram positive cocci that often occur in pairs (diplococci) or short chains and are difficult to distinguish from streptococci on physical characters mentioned above. The two species are commensal organisms in the intestine of humans. The Enterococci are facultative anaerobic organisms non spore forming that grows optimally at 35°C . However, they tolerate wide range of environmental conditions (10-45°C) pH (4.5 to 10.5) quite high NaCl concentration (.6.5%) and can survive heating at 60°C for 30 min. Catalase-negative, oxidase negative-bacteria of the genes Enterococcus are ubiquitous organisms that often occur in large numbers on vegetables, plant materials and foods especially those of animal origin such as meat and dairy products. Enterococci also constitute a large preparation of autochthonous bacteria associated with the mammalian gastro-intestinal tract. The resistance of enterococci to pasteurization temperatures and their adaptability to different substrates and growth conditions in food products manufactured from raw materials and in heat treated food products is of great significance. Enterococci may constitute an important part of the microflora of fermented cheese and meats.

Escherichia coli

E. coli strains are associated with food borne gastroenteritis. These are Gram-negative asprogenous rods that ferment lactose and produce dark colonies with a metallic sheen on Endo agar. The organism grows well on a large number of media and in many foods. They grow over a wide range of temperature (4 to 46 °C) and pH (4.4 to 9.0). However, they grow very slowly in foods held at refrigerator temp. (5 °C). They belong to the family Enterobacteriaceae. The organism is also an indicator of fecal pollution. The organism

is also capable of producing acid and gas and off-flavours in foods. *E. coli* strains involved in foodborne-illness can be placed into five groups: enteropathogenic (EPEC), enterotoxigenic (ETEC), enteroinvasive (EIEC), enterohemorrhagic (EHEC) and facultatively enteropathogenic (FEEC). The organism also grows in the presence of bile salts. The primary habitat of *E. coli* is the intestinal tract of most warm blooded animals. *E. coli* 0157: H7 strains are unusually tolerant of acidic environments.

Lactococcus

Lactococcus lactis subsp. *cremoris* *L. lactis* subsp. *lactis* biovar *diaeetylactis* *Lactococcus* is a genus of lactic acid bacteria that were formerly included in the genus *Streptococcus* Group N (Group N *Streptococci*). They are known as homofermentors meaning that they produce a single product of glucose fermentation. They are Gram-positive, catalase negative, non-motile coccus that are found singly, in pairs or in chains. Some of the strains of lactococci are known to grow at or below 7 °C. Lactococci are intimately associated with dairy products. These organisms are commonly used in the dairy industry in the manufacture of fermented dairy products like cheeses. They can be used in single strain starter cultures or in mixed strain cultures with other lactic acid bacteria such as *Lactobacillus* and *Streptococcus*. Their main purpose in dairy production is the rapid acidification of milk. This causes drop in the pH of fermented product which prevents the growth of spoilage and pathogenic bacteria. These bacteria also play a role in the flavor of the final product. Dairy lactococci have also been exploited for several industrial fermentations in the biotechnology industry. They are easily grown at industrial scale up on cheap whey based media. *Lactococcus lactis* subsp. *lactis* includes species formerly designated as *S. lactis* subsp. *lactis*. *L. lactis* subsp. *cremoris* is distinguished from *L. Lactis* subsp. *lactis* by the inability to (i) grow at 40 °C (ii) grow in 4% NaCl (iii) hydrolyse arginine and (iv) ferment ribose.

Lactobacillus

The organisms belonging to this important genus are rods usually long and slender and in some of the species form chains. They are aerotolerant/microaerophilic but some ferment sugars chiefly to lactic acids if they are homofermentative. The hetero fermentative species, besides lactic acid, also produce small amount of acetic acid, carbon dioxide and trace amounts of volatile compounds such acetaldehyde and alcohol. The homofermentative species of *Lactobacillus* include *L. bulgaricus*, *L. casei*, *L. helveticus*, *L. lactis*, *L. acidophilus* and grow optimally at 37 °C. *L. fermentum*, *L. brevis* are the typical example of hetero fermentative *Lactobacillus* and grow well at higher temperatures. Lactobacilli are of considerable importance in foods as they ferment sugar to lactic acid and other desirable flavouring compounds and are thus used in the production of fermented plant dairy and meat products. However, they are also implicated in the spoilage of wine and beer. The organism normally occurs on plant surfaces silage, manure and dairy products. They are quite fastidious in their nutritional requirements as they are unable to synthesize certain vitamins they require and, therefore, media need to be supplemented with these vitamins for their growth. Some of the strains are psychotropic in nature and are thus involved in the spoilage of refrigerated meats.

On the other hand thermotolerant properties (resistance to pasteurization temperature) of some of the thermophilic strains of lactobacilli are quite useful in the manufacture of certain varieties of cheeses e.g. Swiss cheese. Some strains of lactobacilli also show probiotic attributes and are finding application in functional probiotic foods and in pharmaceutical preparations.

Leuconostoc

Leuconostoc is a genus of Gram-positive bacteria, placed within the family of Leuconostocaceae. They are generally ovoid cocci often forming chains. Leuconostoc spp. are intrinsically resistant to vancomycin and are catalase-negative (which distinguishes them from staphylococci). All species within this genus are heterofermentative and are able to produce dextran from sucrose. They are generally slime-forming. Blamed for causing the 'stink' when creating a sourdough starter, some species are also capable of causing human infection. Leuconostoc spp. along with other lactic acid bacteria such as *Pediococcus* and *Lactobacillus* spp., is responsible for the fermentation of cabbage, to sauerkraut. In this process the sugars in fresh cabbage are transformed to lactic acid which give it a sour flavour and good keeping qualities.

Listeria monocytogenes

Listeria monocytogenes in foods has attracted worldwide attention due to the serious illness it causes in human beings. The Listeria are Gram positive non spore forming, nonacid-fast rods. The organism is catalase positive and produces lactic acid from glucose and other fermentable sugars. The organism grows well in brain heart infusion (BHI), trypticase soy, and tryptose broths. However, the medium should be fortified with B. vitamins and the amino acids. It is a mesophilic organism with optimal growth temperature 37°C but it can grow at refrigerator temperature also. Strains grows over the temperature range of 1°C to 45°C and pH range.

Listeria monocytogenes is widely distributed in nature and can be isolated from decaying vegetation, soil, animal feces, sewage, silage and water. The organism has been found in raw milk, pork, raw poultry, ground beef and vegetables. The HTST treatment of pasteurization is good enough to destroy the organism in milk. The most significant virulence factor associated with *L. monocytogenes* is listeriolysin O. The virulent strains produce β -hemolysis on blood agar and acid from rhamnose. *L. monocytogenes* grows well in moderate salt concentrations (6.5%). *L. monocytogenes* is unique among foodborne pathogens while other pathogens excrete toxins or multiply in the blood stream, *L. monocytogenes* enters the host's cells and grows inside the cell. In humans it crosses the intestinal barrier after entering by the oral route. Ready to Eat (RTE) foods that are preserved by refrigeration pose a special challenge with regard to *L. monocytogenes* infection.

Micrococcus

Micrococcus occurs in a wide range of environments, including water, dust, and soil. Micrococci are Grampositive spherical cells ranging from about 0.5 to 3 micrometers in diameter and typically appear in tetrads. Micrococcus has a substantial cell wall, which may comprise as much as 50% of the cell mass. Some species of Micrococcus, such as *M. luteus* and *M. roseus* (red) produce yellow or pink colonies when grown on mannitol salt agar. Micrococcus is generally thought to be a saprophytic or commensal organism, though it can be an opportunistic pathogen, particularly in hosts with compromised immune systems, such as HIV patients.

1.2.17 *Proteus* Since it belongs to the family of Enterobacteriaceae, general characters are applied on this genus: It is oxidase negative, but catalase and nitrate reductase positive. Three species *P. vulgaris*, *P. mirabilis*, and *P. penneri* are opportunistic human pathogens. *Proteus* includes pathogens responsible for many human urinary tract infections. *P. mirabilis* causes wound and urinary tract infections. Most strains of *P. mirabilis* are sensitive to ampicillin and cephalosporins. *P. vulgaris* is not sensitive to these antibiotics. However, this organism is isolated less often in the laboratory and usually only targets immune suppressed individuals. *P. vulgaris* occurs naturally in the intestines of humans and a wide variety of animals; also manure, soil and polluted waters. *P. mirabilis*, once attached to urinary tract, infects the kidney more commonly than *E. coli*. *P. mirabilis* are often found as free-living organisms in soil and water.

Propionibacterium spp.

(*P. freudenreichii*) Historically, *Propionibacterium* spp. are of interest because of their use as dairy starters (especially in the production of Swiss-type cheese) and their ability to produce propionic acid during growth. The genus *Propionibacterium* is generally split into —cutaneous and —dairy groups. The dairy *Propionibacterium* spp. can also be isolated primarily from dairy foods and silage. The species in dairy products include *P. jensenii*, *P. acidipropionici*, *P. theonii* and *P. freudenreichii*. *Propionibacteria* have a role in the production of flavour compounds in cheese by proteolysis and propionic acid production. Dairy strains of *propionibacteria* are autolytic under environmental conditions found in cheese and degrade peptides and amino acids that are present in the cheese. And The dairy species offer an interesting opportunity as novel probiotic organisms with the most obvious advantage being that they are considered safe for ingestion.

Pediococcus spp. (*Pediococcus pentosaceus*, *P. acidilactici*) *Pediococci* comprise a group of bacteria that are of economic importance in the brewing and food industries. Several species and strains of *pediococci* have been used as starter cultures in the fermentation of vegetables, meats, sausage products, fermented milks and associated with the development of flavor in Cheddar and other related cheese varieties. Some strains form capsular material that causes beer to become ropy and viscous. They are catalase negative and exhibit a homolactic type of fermentation and produce optically inactive lactic acid i.e. a mixture of the L(+) and D(-) type. They generally appear in tetrads.

1.2.20 *Pseudomonas fluorescens* *Pseudomonas fluorescens* is a common Gram-negative, rod-shaped, motile bacterium. The organism is psychrotrophic in

nature and grows at refrigeration temperature (7°C). It has an extremely versatile metabolism, and can be found in the soil and in water. It is an obligate aerobe, but certain strains are capable of using nitrate instead of oxygen as a final electron acceptor during cellular respiration. Optimal temperature for growth of *Pseudomonas fluorescens* is 25-30 °C. It tests positive for the oxidase. *Pseudomonas fluorescens* also a nonsaccharolytic organism. Heat-stable lipases and proteases are produced by *Pseudomonas fluorescens* and other similar pseudomonads.

These enzymes cause milk to spoil, by causing bitterness, casein breakdown, and ropiness due to the production of slime and coagulation of proteins.

1.2.21 *Pseudomonas aeruginosa* It is a Gram-negative, aerobic, rod-shaped bacterium with unipolar motility. An opportunistic human pathogen, *P. aeruginosa* is also an opportunistic pathogen of plants. *P. aeruginosa* is the type species of the genus *Pseudomonas* (Migula). Gram-stained *Pseudomonas aeruginosa* bacteria (pink-red rods) secrete a variety of pigments, including pyocyanin (blue-green), pyoverdine (yellow-green and fluorescent), pyorubin (red-brown). *P. aeruginosa* is often preliminarily identified by its fluorescence and grape-like or tortilla-like odor in vitro. Definitive clinical identification of *P. aeruginosa* often includes identifying the production of pyocyanin and fluorescein, as well as its ability to grow at 42°C. *P. aeruginosa* is capable of growth in diesel and jet fuel, where it is known as a hydrocarbon-using microorganism (or "HUM bug"), causing microbial corrosion. *P. aeruginosa* is considered by many as a facultative anaerobe.

1.2.22 *Salmonella* (*S. typhimurium*, *S. typhi*, *S. enteritidis*) *Salmonella* spp. have been reported to be a leading cause of foodborne illnesses in humans. Foodborne salmonellosis scores over all other foodborne bacterial illnesses in humans. Enteric fever is a serious human disease associated with typhoid and paratyphoid strains.

Salmonella Enterobacteriaceae. The optimum growth temperature is 37-45 °C. The organism can also grow at about 7°C in foods. It ferments carbohydrates with its production of acid and gas. *Salmonella* are oxidase negative, catalase positive and grow on citrate as a sole carbon source and produce H₂S. Some *Salmonella* strains can grow at higher temperatures (54 °C) while others exhibit psychrotrophic properties. The organism has the ability to grow at pH values ranging from 4.5 to 9.5, with an optimum pH growth at 6.5 to 7.5. spp. are facultatively anaerobic, small Gram-negative, non spore forming, rod-shaped (2-4 μm) bacteria belonging to the family Enterobacteriaceae. Milk, meat and poultry are principle vehicles of human foodborne salmonellosis. Ingestion of only a few salmonella cells can be infectious. Low levels of salmonellae in a finished food products may, therefore, be of serious public health consequence.

1.2.23 *Serratia* *Serratia* is a genus of Gram-negative, facultatively anaerobic, rod-shaped bacteria of the Enterobacteriaceae family. The most common species in the genus, *S. marcescens*, is normally the only pathogen and usually causes nosocomial infections. However, rare strains of *S. plymuthica*, *S. liquefaciens*, *S. rubidaea*, and *S. odoriferae* have caused diseases through infection. Members of this genus produce characteristic red pigment, prodigiosin.

Streptococcus thermophiles

The only streptococcus species that is associated with food technology is *S. thermophilus* which is used in the manufacture of yoghurt (in co culture with *L. bulgaricus* and Dahi). *S. thermophilus* is a Gram positive facultative anaerobe and belongs to the family Streptococcaceae. It is catalase negative organism that is non-motile, non-spore forming and homofermentative and occurs in pairs to long chains. The spherical to avoid cells are with a diameter in the range of 0.7 to 0.9 μm . The optimum temperature for the growth of this organism is between 39°C to 45°C, although most species in the genus are able to grow at temperature ranging from 45-60°C. They do not grow at temperature below 20°C, but they can survive at 65 for 30 min. They ferment sugars with L (+) lactic acid as the major end product and produce around 0.6 to 0.8% lactic acid. They are able to grow in broth with 2.5% NaCl but fail to grow in 6.5% NaCl at pH 9.6 or in milk with 0.1% methylene blue (Bergey's Manual 1994). It is also classified as lactic acid bacteria (LAB). It is a very versatile organism. *S. thermophilus* has properties that make it one of the commercially most important lactic acid organism. *S. thermophilus* is used along with *Lactobacillus* spp., as a starter culture to manufacture several important fermented dairy foods including yoghurt and mozzarella cheese.

Though the natural habitat of *S. thermophilus* is yet to be established, most strains have been isolated from milk environments. 1.2.25 *Staphylococcus aureus* *Staphylococcus aureus* is commonly associated with humans. It is a Gram-positive catalase-positive coccus. *Staphylococcus aureus* is the common cause of foodborne gastroenteritis known as staphylococcal food poisoning. Staphylococcal gastroenteritis is caused by the ingestion of food that contains one or more enterotoxin which are produced by some strains of *S. aureus*. Although enterotoxin production is believed generally to be associated with coagulase and thermo nuclease producing *S. aureus* strains, many species of *Staphylococcus* that produce neither coagulase nor TNase are also known to produce enterotoxin. The main reservoir of *S. aureus* is the nasal cavity of human beings from where they find their way to the skin and wounds. Mastitis in animals due to *S. aureus* is quite common and from the infected udder the organism finds its way to the milk. The organism can grow well in NaCl concentrations of 7 to 10%. Though the optimum growth temperature of the organism is 37 °C , some strains can grow at a temperature as low as 6.7 °C. The organism can grow to water activity as low as 0.86.

Shigella Bacillary dysentery, or shigellosis, is caused by *Shigella* species. *Shigella* is a member of the family Enterobacteriaceae. The growth temperature varies from 10 to 48 °C. *Shigella*2S. *Shigella* does not usually survive well in low pH foods. *Shigella* is sensitive to ionizing radiations. species are non-motile, oxidase negative produce acid only from sugars; do not grow on citrate as sole carbon source, do not grow on KCN agar, and unlike *Salmonellae* do not produce H *Shigella* is an important disease in developed and developing countries. Disease is caused by ingestion of contaminated foods, and in some instances it subsequently leads to rapid dissemination through contaminated feces from infected individuals. The infective dose may be as low as 100 cells. Contamination of foods usually does not occur at the processing plant but rather through an infected food handler. Humans are the natural reservoir of *Shigella*. The organism is spread through the fecal-oral route.

Vibrio *Vibrio cholerae* and *V. parahaemolyticus* are the two important species of the genus *Vibrio*. *Vibrio cholerae* O1 causes cholera, one of the few food borne illnesses with epidemic and pandemic potential.

Vibrio cholerae are Gram-negative straight or curved rods and belong to the family Vibrionaceae. Important distinctions within the species are made on the basis of productions of cholera enterotoxin (CT) and serogroup. Vibrio cholerae is part of the normal free living bacterial flora in estuarine areas. Amongst the many different enrichment broths described for the isolation of vibrios alkaline peptone water is the most commonly used. Though V.

parahaemolyticus can grow in the presence of 1-8% NaCl, the best growth occurs in the salt concentration 2 to 4%. Yersinia Yersinia enterocolitica and Yersinia pestis are the two important human pathogens while Y. enterocolitica causes food borne gastroenteritis, Y. pestis is an agent of human plague. Y. enterocolitica also known as newly emerging human pathogen is a heterogeneous species that is divisible into a large number of subgroups. Y. enterocolitica is unusual because it can grow at temperatures below 4 °C. The generations time at the 28- 30 °C (Optimum growth temperature) is almost 34 min. It also survives in frozen foods. It grows better in processed foods such as pasteurized milk, vacuum packed meat, boiled eggs, boiled fish, and cottage cheese. Both the species can grow over a pH range of 4 to 10 (optimum pH is 7.6) and tolerate alkaline environment well. They can motile at a temperature < 30 °C. However, both these organisms are susceptible to pasteurization, ionizing and ultraviolet (UV) irradiation. The organism can also tolerate upto 5% NaCl. Infections with Yersinia species are due to transmittance of the organism from animals to humans. The organism is frequently present in pork, lamb, poultry and dairy products.

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