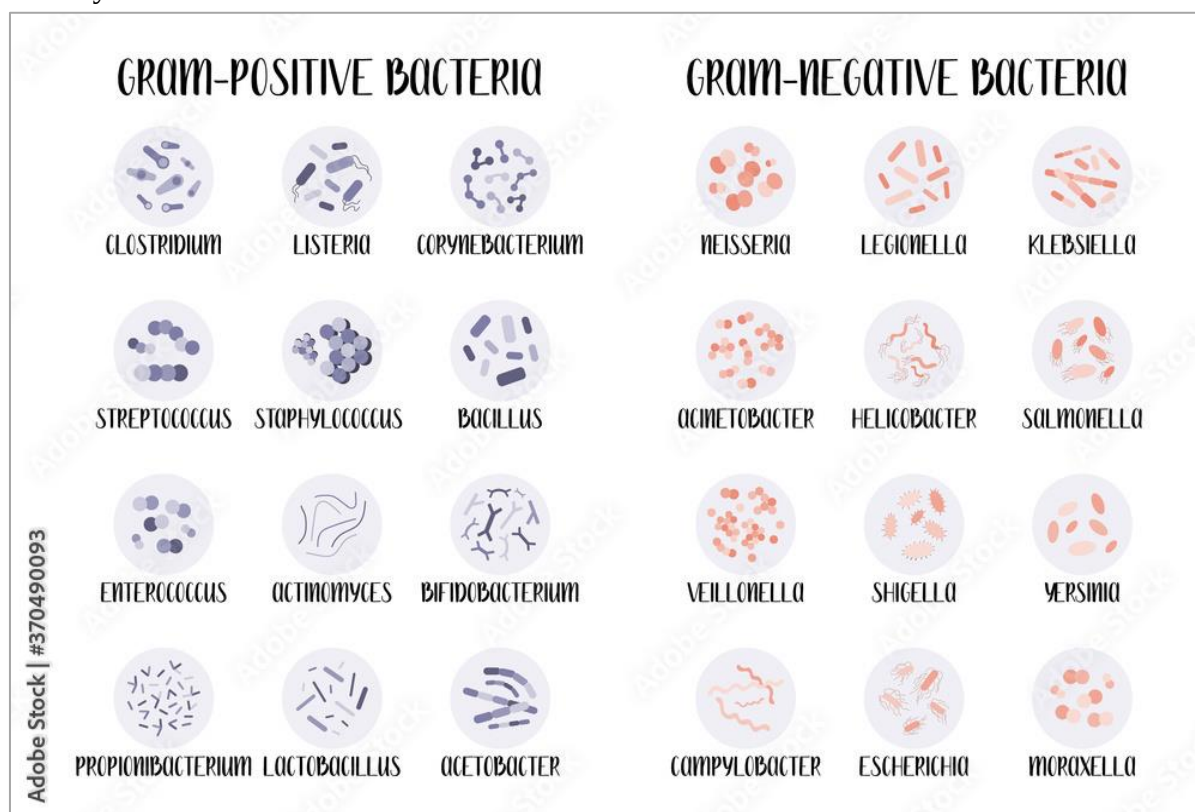


Chapter 3 : Major microbial groups

Certain bacteria present a significant risk to food safety and public health due to their capacity to cause spoilage and disease. This is driven by their **infectious potential**, which includes contamination, multiplication, harmful enzymatic activity, and toxin release (often endotoxins). Their impact is further defined by their **pathogenicity**, **virulence**, and **toxicity**, leading to foodborne poisoning, intoxication, or toxi-infection. Conversely, other bacteria are beneficial and are harnessed for their positive roles in food production and fermentation within the industry.



1. Gram- bacteria: *Enterobacteria*

Coliforms are Gram-negative, non-spore-forming, AAF bacilli (Aggregative Adherence Fimbriae) defined by their ability to ferment lactose with acid and gas production at 37/30°C within 48 hours. In food microbiology, they serve as crucial **indicator flora** for fecal contamination and general hygienic quality.

- **Typical coliforms:** This group includes *Escherichia coli*, *Citrobacter*, *Enterobacter*, and *Klebsiella*.
- **Exclusion:** Pathogens like *Salmonella* and *Shigella* do not fit the coliform definition.
- **Fecal specificity:** The subgroup of **thermotolerant coliforms** (tolerant to 44-45°C) provides definitive evidence of fecal contamination, predominantly represented by *E. coli*.

- ***Escherichia***

Escherichia coli is the principal species within the *Escherichia* genus. It is characterized by key biochemical markers: motility, a positive indole test, lactose fermentation, and gas production from glucose, while being negative for citrate utilization, urease, Voges-Proskauer, hydrogen sulfide, and gelatin hydrolysis.



As a **normal and abundant commensal** in the intestinal tracts of humans and animals, it is routinely shed in feces. However, certain **pathogenic serotypes** can cause specific intestinal diseases.

A prime example is the **enterohemorrhagic strain *E. coli* O157:H7**, a regulated pathogenic agent (AFNOR standard) responsible for severe **hemorrhagic colitis**. Its pathogenicity is largely due to the production of potent **enterotoxins** (like Shiga toxin).

Enterotoxins are protein-based bacterial toxins that act within the intestine. They bind to the epithelium, disrupting ion absorption (Na⁺, Cl⁻) and causing a massive outflow of water, leading to **diarrhea**, with or without tissue damage.

Human infection typically occurs through the **consumption of contaminated food**, most notably undercooked ground beef, but also via raw milk, water, unwashed fruits, and vegetables.

- ***Citrobacter***

AAF, bacilli/coccobacilli, motile, glucose +, nitrate reduction to nitrite +, indole -, ONPG +, urease -, VP -, LDC-, ODC+, ADH+, lactose +, citrate+ and H₂S +/-.

These bacteria can be found in soil, water, sewage and the human intestine. They cause urinary tract infections, bacteraemia, brain abscesses, pneumonia and other neonatal infections (meningitis).

- ***Enterobacter***

Bacilli, motile, glucose +, nitrate reduction to nitrite +, indole -, ONPG +, urease -, VP+, LDC+, ODC+, ADH-, lactose +, citrate+ and H₂S-.

Habitat is the human and animal intestine, also found in faeces, sewage, soil, and dairy products. Strains of the *Enterobacter* genus are often responsible for nosocomial infections of the urinary tract or wounds and meningitis.

- ***Klebsiella***

Asporeous, immobile, glucose +, lactose +, sucrose +, mannitol + with gas production, urease +, VP+/RM -, indole -, H₂S- and gelatinase -. Saprophytes of the upper respiratory and digestive tracts of humans and animals. Responsible for bronchitis, tonsillitis, otitis, and urinary and digestive tract infections.

- ***Salmonella***

Motile, glucose + without gas production, lactose -, citrate +, ONPG-, VP-, indole -, H₂S+, RM+, LDC+, ODC+, ADH-, urease-.

Some salmonella bacteria are enteroinvasive and penetrate the cells of the intestinal mucosa. Typhoid and paratyphoid fevers are caused by *Salmonella Typhi*, Para A, Para B and Para C. -*Salmonella* bacteria cause gastroenteritis.

- ***Shigella***

Shigella are enterobacteria with low metabolic activity. They are genotypically very similar to *Escherichia*. Spore-, immobile, citrate-, glucose+ without gas production, lactose-, gelatinase-, ONPG+/-, H₂S-, RM+, VP-, urease-, catalase-, indole-, LDC-, ODC-, ADH-.

Strictly human, *Shigella dysenteriae*: toxin (Shiga toxin cfr EHEC). *Shigella* are therefore enteric invasive bacteria. *Shigella* are the causative agent of **bacillary dysentery**. This is an intestinal infection mainly localised in **the large intestine**, where the germ multiplies, causing **inflammation of the mucous membrane**. Dysenteric syndrome includes: severe abdominal pain, sometimes bloody diarrhoea, and fever.

- ***Yersinia***

Yersinia, motile (22°C), citrate-, glucose+, lactose-, ONPG+, H₂S-, VP+, urea +, indole +/-.

Natural reservoir: soil, water, animals and plants. Ingestion of contaminated food (milk, shellfish, meat, poultry). There are 57 O antigens and 16 H antigens, of which mainly types O:3, O:9 and O:8 are found in human infections. A heat-sensitive virulence plasmid is responsible for the enteric invasiveness of the strains.

2. Gram-: *Vibrio*

- ***Vibrio cholerae***

Curved comma-shaped bacilli, motile, NR+, catalase +, indole+, LDC+, AAF, oxidase +, spore-, capsules-, proteolytic and slightly glucidolytic. Alkaline media (pH=9.2) and saline media with 3% NaCl (alkaline saline peptone water).

Cholera is a serious disease that is most often transmitted through water. This bacterium is only tested for in foods suspected of causing the disease or in high-risk areas.

-Cholera enterotoxin.

- ***Vibrio parahaemolyticus***

This marine bacterium is increasingly recognised as the cause of food poisoning foodborne infection following the consumption of seafood.

3. Gram-: *Brucella*

Gram-negative coccobacilli, non-spore-forming, immobile, strictly aerobic, urease +, indole -, catalase +, nitrate reductase +.

Brucellosis is a serious infectious disease linked to the consumption of specific foods, with goat's cheese often being implicated.

4. Gram- : *Pseudomonas*

Gram-negative, motile, strictly aerobic bacilli, oxidase +, catalase +, presence of nitrate reductase. This flora, common due to its widespread presence, causes alterations in meat. The genus *Pseudomonas* has been isolated from meat and poultry.

5. Gram +: *Streptococci*

Faecal *streptococci* are part of the intestinal flora of humans and animals; their number varies from 10^5 to 10^7 per g of faecal matter. These germs are also widespread in nature (plants, etc.). Of all non-spore-forming bacteria, these germs are among the most resistant to unfavourable environmental conditions. These germs are more resistant than coliforms and *E. coli* to refrigeration, freezing, heating and salting. As a result, these bacteria can multiply in a large number of foods (acidic, salty, refrigerated products, etc.), which they can then spoil.

6. Gram+: *Staphylococcus*

S. aureus: Ferments mannitol, NaCl 75 g/l, Enterotoxins + Leukocidin + Haemolysins, Coagulase + Thermonuclease

7. Gram+: *Anaerobic sulphite-reducing bacteria*

Sulphite-reducing anaerobic bacteria are normal inhabitants of the human and animal intestine, but are frequently found in nature, particularly in soil and decaying organic matter.

- *Clostridium perfringens*
- *Clostridium botulinum*
- *Bacillus cereus* (another spore-forming bacterium, AAF).

8. Gram+: *Listeria* bacteria

- *Listeria monocytogenes*

A small Gram-positive bacillus with rounded ends, which can form a palisade, asporule, oxidase -, catalase +, esculin +, mesophile (cultivated between +3 and +45°C with an optimum at 37°C). The bacterium remains viable after several years of storage at 4°C.

9. Lactic acid bacteria

Lactic acid bacteria (LAB) are Gram-positive, non-sporulating bacteria with a coccal or rod-shaped morphology. A primary biochemical hallmark is their **catalase-negative** status, as they lack the enzyme catalase used to break down hydrogen peroxide. However, certain species may develop a non-enzymatic "pseudocatalase" activity.

LAB are **fermentative organisms** that generate ATP through **lactic acid fermentation** of carbohydrates. They do not respire using oxygen. Their glucose metabolism diverges into two main pathways:

- **Homofermentative:** Producing mainly lactic acid.
- **Heterofermentative:** Producing lactic acid along with other byproducts like ethanol and carbon dioxide. The pathway used depends on the bacterial species and growth conditions.

The group encompasses several genera, traditionally divided based on morphology and fermentation pathways:

❖ Cocci

- ***Lactococcus*:** Homofermentative. Historically grouped as "lactic streptococci." Crucial in dairy fermentations (e.g., *L. lactis* for cheese and buttermilk).
- ***Streptococcus*:** Homofermentative. Includes commensal, pathogenic, and industrially important species. *S. thermophilus* is vital for yogurt production.
- ***Enterococcus*:** Homofermentative. Often found in the intestines of humans and animals. Some are opportunistic pathogens (e.g., *E. faecalis*), while others are used as probiotics.
- ***Leuconostoc*:** Heterofermentative. Produces CO₂, lactate, and ethanol/acetic acid. Important in sauerkraut, kimchi, and some dairy products for flavor development.
- ***Pediococcus*:** Homofermentative. Tetrad-forming cocci. Used in fermented meats, vegetables, and some beers.

❖ Rods (Bacilli)

- ***Lactobacillus* (historically a large genus, now reclassified):** Includes both homo- and heterofermentative species. Extremely diverse and prevalent in food fermentations (dairy, meat, vegetables, sourdough), as probiotics, and as commensals in the human body (e.g., vagina, gut). *Note: Recent taxonomy has split this genus into ~25 new genera (e.g., Lactiacaseibacillus, Limosilactobacillus, Lactiplantibacillus), though the term "lactobacilli" is still widely used.*
- ***Carnobacterium*:** Heterofermentative. Often isolated from chilled meat, fish, and certain dairy products.
- ***Weissella*:** Heterofermentative. Found in fermented foods and mucosal environments. Can produce dextran.

❖ Industrial and health significance

- **Food industry:** LAB are essential as starter cultures, preservatives (via acid production), and flavor/aroma agents in yogurt, cheese, sauerkraut, kimchi, sausages, and wine.
- **Probiotics:** Many strains, particularly from *Lactobacillus*, *Bifidobacterium* (which is technically an Actinobacterium but often grouped with LAB due to similar properties), and *Enterococcus*, are marketed for their purported health benefits.
- **Biotechnology:** Used in the production of bacteriocins (natural antimicrobials), exopolysaccharides, and various metabolites.

10. Acetic acid bacteria

Acetic acid bacteria (AAB), primarily from the genera *Acetobacter* and *Komagataeibacter*, are Gram-negative, aerobic organisms known for their unique and valuable metabolic trait: the oxidation of ethanol into **acetic acid**. This process, called acetous fermentation, is the essential biochemical foundation for the commercial production of **vinegar** from wine, cider, or other alcoholic substrates. While this makes them highly beneficial in the food industry, their oxidative metabolism can also be detrimental. Uncontrolled growth of AAB is a major cause of **wine spoilage**, where they convert the alcohol into acetic acid and ethyl acetate, imparting a sharp, sour vinegar note and ruining the product. Thus, their role is a classic example of controlled application versus unwanted contamination.

11. Yeasts and molds

• Yeast

In the microbial world, yeasts present a dual nature. **Beneficial yeasts**, such as *Saccharomyces cerevisiae*, are indispensable to humanity. They drive essential fermentation processes for producing bread, beer, wine, and bioethanol. Additionally, certain species like *Kluyveromyces lactis* are key in dairy fermentation, and some strains of *Saccharomyces boulardii* are used as probiotics for gut health. Conversely, **non-beneficial (or pathogenic) yeasts** can cause significant harm. The most notable is *Candida albicans*, an opportunistic pathogen responsible for infections ranging from superficial thrush to life-threatening systemic candidiasis. Other species, like *Cryptococcus neoformans*, can cause severe meningoencephalitis, particularly in immunocompromised individuals.

• Molds

Molds in food present a stark duality of value and risk. **Beneficial molds** are deliberately cultivated to produce iconic foods and enzymes. For instance, *Penicillium roqueforti* and *P. camemberti* are essential for creating the distinct flavors and textures of blue cheeses and soft-ripened cheeses like Camembert. *Aspergillus oryzae* (koji mold) is fundamental in fermenting soy sauce, miso, and sake. However, **non-beneficial molds** are major agents of food spoilage, causing visible rot, off-flavors, and economic loss. More critically, certain species, particularly

some *Aspergillus*, *Penicillium*, and *Fusarium* molds, can produce **mycotoxins**—potent, heat-stable chemical toxins that persist through processing. Ingestion of mycotoxin-contaminated food (e.g., aflatoxins in nuts, ochratoxin in grains) poses serious health threats, including carcinogenic, immunosuppressive, and teratogenic effects.