FE 305 FOOD MICROBIOLOGY
Intoxications and Toxico-infections

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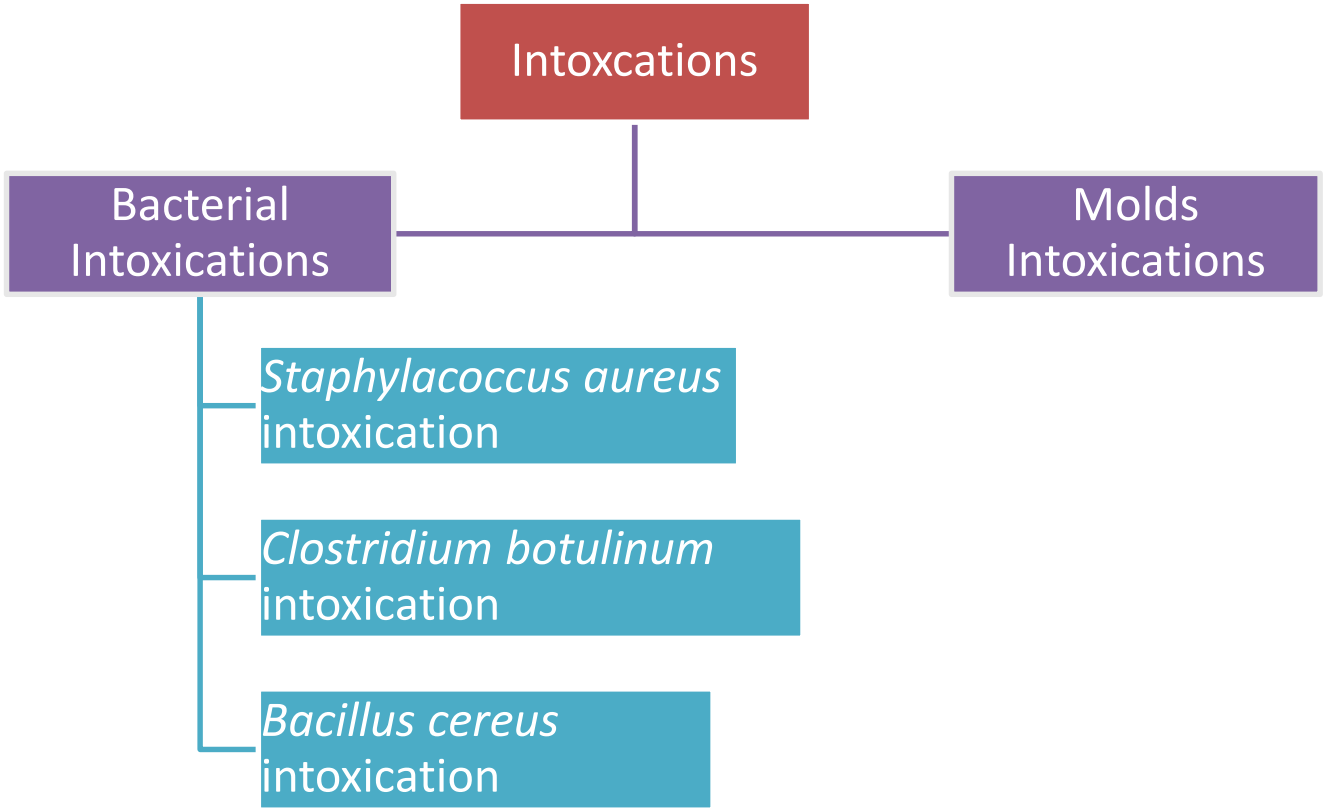
Foodborne Bacterial Intoxications

- **Food intoxication** is the poisoning caused by pathogenic microorganisms as a result of the consumption of toxin containing foods that is produced by microorganism in foods.
- As stated in the definition, for the formation of intoxication, it is necessary to consume not the pathogenic microorganisms, but the toxins they create in the food.
- Some general features seen in intoxications are:
  - The toxin is formed during the reproduction of the pathogen in food.
  - The toxin may or may not be heat resistant.
  - In general, the toxin must be taken with food for poisoning to occur.
  - Symptoms of the disease begin to appear shortly (30 minutes) after food consumption.
  - Symptoms of the disease vary according to the type of toxin.
  - No change in body temperature is observed.

Foodborne Bacterial Intoxications



Staphylococcus aureus intoxication

- *Staphylococci* are bacteria that look like bunches of grapes under the microscope. *S. aureus*, a local infectious agent, has the ability to reproduce in many foods. Most of the enterotoxin-producing strains show coagulase (+) properties. However, not all coagulase (+) *staphylococci* are enterotoxigens.
- It shows the fastest growth between 4-46 °C.
- It is resistant to low water activity (0.86) and is not affected by high salt concentration.
- Acid tolerant; It can grow at a pH of 4.8 under aerobic conditions.
- The toxins produced by *S. aureus* are serologically divided into six types (A, B, C1, C2, D, E). Their toxicities are also different; Enterotoxin-A is the most effective (minimum dose 1 ng/kg). The conditions required for this bacterium to grow and produce toxins differ according to the food.
- Enterotoxins are simple proteins. The most common toxin types in food poisoning cases are A and D. The most important feature of toxins is that they are resistant to heat treatment. In sterilization at 117°C, they are inactivated.

Staphylococcus aureus intoxication

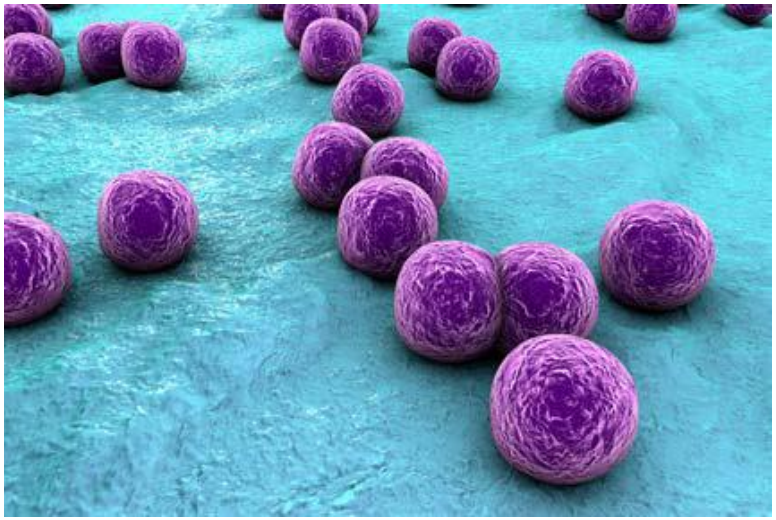
- *Staphylococci* are Gram (+), aerobic, sporeless and easily killed bacteria by pasteurization. When grown in solid culture media, they form colonies of various colors. They get their names according to these colors:
  - Golden yellow pigment producers: *Staphylococus aureus*
  - Those that make yellow pigment: *S. citrus*
  - Makers of white pigment: *S. albus*
- All *S. aureus* with enterotoxins beta hemolysis on blood agar; Coagulates (coagulates) plasma prepared from human or rabbit blood. They multiply in environments with 7.5% salt (NaCl) and ferment mannitol to produce acid and gas.
- The disease occurs 1-6 hours after people eat food contaminated with these bacteria.
- Symptoms include nausea, vomiting, severe abdominal pain and diarrhea.
- Symptoms can be seen in mild, moderate and severe degrees and disappear in a day or two.
- Recovery is normal and complete; it usually does not cause death and the majority of cases do not require treatment.

Staphylococcus aureus intoxication

- Humans are the main reservoir;
  - on skin, in mouths, throats and nasal cavity.
  - about 30-50 % of healthy people carry *S. aureus*.
  - infected wounds, cuts, sinuses, pimples are abounding with *S. aureus*.
  - air, soil, water, sewage, plant surfaces, meats, poultry and dairy products.
- Foods involving in outbreaks: protein rich foods:
  - including meat, canned beef, salami, bacon, salads, sausages, cram-filled bakery foods, meat, meat-filled sandwiches, cheese, pudding, ice cream and cream-filled bakery products.

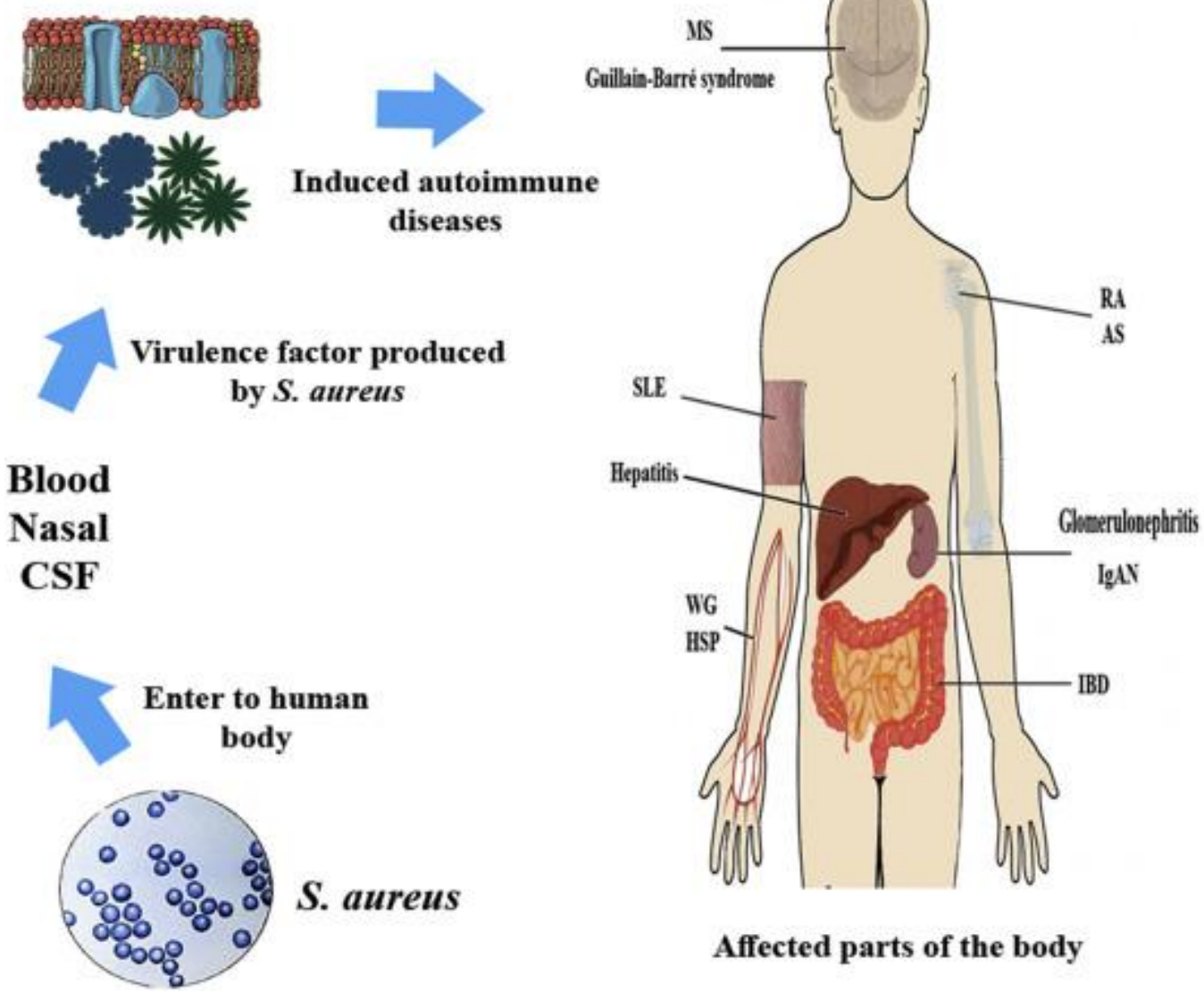
Staphylococcus aureus

a) Without Gram staining



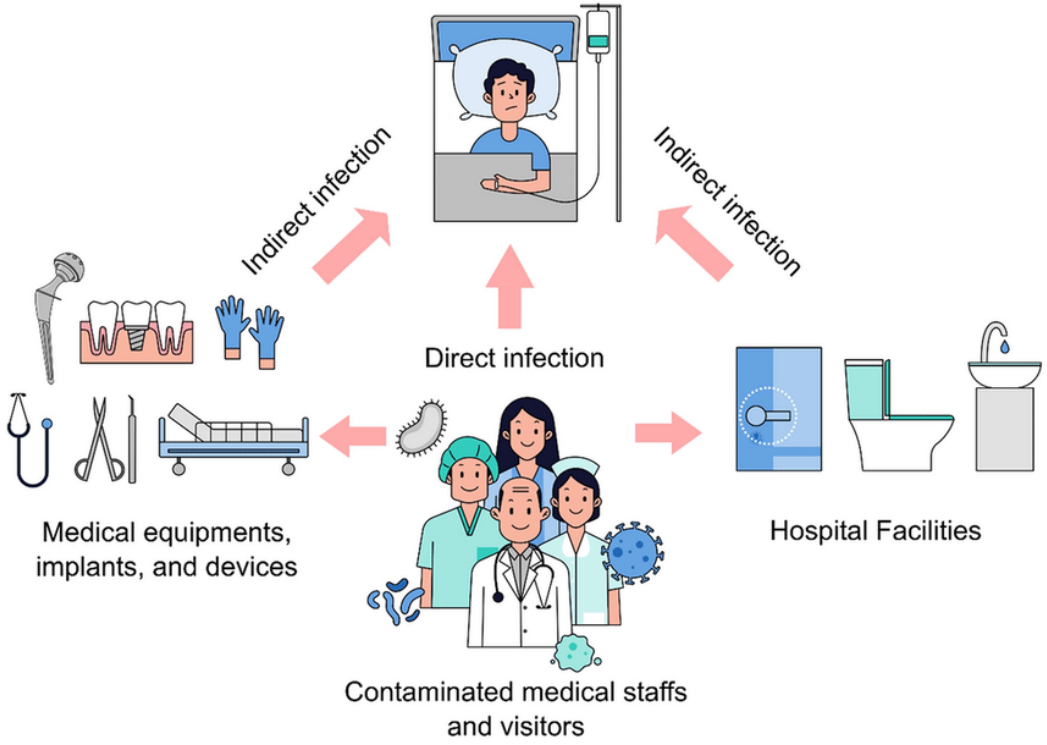
b) With Gram staining

S. aureus Intoxication Mechanism



Staphylococcus aureus intoxication

- The diagram below shows how this type of human poisoning occurs:



- Healthy people also carry these bacteria in their mouth, throat and nose. Infected wounds are particularly rich in staphylococcal bacteria. any part of the body that is inflamed; mouth, throat, sinuses, ears, nose, acne contain abundant staphylococcal bacteria.

Staphylococcus aureus intoxication

- Bacteria to food, carelessness of employees or coughing; Handling food without washing hands after touching places such as nose, mouth, ears, doorknobs, money or using tissues, and continuing food preparation work without washing hands thoroughly or not after going to the toilet, cause the transmission of this bacteria.
- Protein-rich foods and meals made with them are more suitable for staphylococcal poisoning. The way they are prepared also increases the growth of bacteria rapidly.
  - Meats, chicken, fish, milk, eggs, shellfish, salami, sausage, sausage, mincemeat dishes, soups and sauces made with broth, mixtures with eggs, sugar and milk, cream cakes, etc.
- Because these foods are less acidic, *Staphylococci* multiply easily in such foods. When high acid foods such as yogurt are mixed with eggs and used, their acidity decreases and can become a suitable environment for the reproduction of staphylococci.
- Foods that are handled and dealt with a lot during food preparation; Processes such as separating the meat from the bone, chopping, mincing in the machine, forging, shaping increase the contamination of food with bacteria.

Staphylococcus aureus intoxication

- *S. aureus* grows between 4-46°C and dies at 47°C and higher.
- Foods should store hot, that is, cooked food to be served immediately, hot (60°C and above), and cold foods cold (5 °C and below). Food that will not be served immediately or leftover cooked food should be cooled to a safe temperature quickly. This can be done by immersing the container in cold water and stirring occasionally if it will not spoil the appearance and consistency of the food.
- Foods such as cooked foods and easily perishable meat should not be kept at room temperature for more than two hours. This prevents bacteria from multiplying and making toxins.
- Since the toxins produced by *S. aureus* are resistant to high temperatures, they cannot be destroyed easily. Although it loses its effect gradually, boiling for 20-60 minutes cannot completely neutralize the toxin. Although the cooking process kills the staphylococci, it cannot destroy the toxin, so poisoning can still occur.
- In addition to all these, people working in food preparation must be healthy and must follow the rules of health and cleanliness during work.

Clostridium botulinum intoxication (Botulism)

- Botulism is an intoxication caused by *Clostridum botulinum* neurotoxin and can occur in three different ways.
  - The most common of these is food intoxication, which is caused by the consumption of toxin-formed food.
  - The second type, called "wound botulism", is very rare and occurs as a result of *C. botulinum* creating a toxin in an infected tissue.
  - In the type called "infant botulism", the toxin produced by *C. botulinum*, which colonizes and reproduces in the intestines, causes the disease. In wound and infant botulism, toxin is produced in-vivo and toxication occurs after infection by bacteria (toxico-infection).
- Botulism poisoning and its agent were first discovered by Van Emmergen in 1896. This poisoning occurs due to the handling of the exotoxins of *Clostridium botilunum* with food.
- Although this type of poisoning is quite common in Europe and especially in America, it is rarely seen in our country. This is based on the fact that Turkish Cuisine has well-cooked dishes.
  - Botulism is caused by the consumption of products such as canned food, sausage and pastrami, which are prepared improperly, especially at home.

Clostridium botulinum intoxication (Botulism)

- Although botulism is less common than other foodborne diseases, it is important because of its high mortality rate. From 766 botulism cases detected in the USA between 1899 and 1977, 1966 people were affected and 999 people (50.8%) died.
- According to the results of another study conducted in this country, 448 people were poisoned in 186 cases between 1960 and 1977 and 82 (18.3%) died; Between 1978 and 1983, 201 people were poisoned in 91 cases and 26 (12.9%) died.
- Low-acid canned vegetables, fish and meat products, smoked meats and fish are the most common foods in cases of botulism, although cases of botulism caused by different foods such as baked potatoes wrapped in aluminum foil, garlic stored in olive oil have also been reported.
- In 1978, 59 people got type A botulism from a baked potato wrapped in aluminum foil and kept at room temperature, and in 1985 36 people got type B botulism from garlic kept in olive oil.

Clostridium botulinum intoxication (Botulism)

- *C. botulinum* is an obligate anaerobic, gram-positive (+), non-encapsulated, peritric flagellated, motile, 0.5-0.8 x 3.0-8.0 mm, rod-shaped, spore-forming bacterium with oval or cylindrical spores. located terminally or subterminally in the cell.
- *C. botulinum* is divided into seven types according to the types of neurotoxins of different antigenic structures it produces. These neurotoxins; They are A, B, C, D, E, F and G.
- Types A and B, which are commonly found in the soil and on the earth, cause poisoning in humans, and very rarely, type E. Although the E type is isolated from the soil, it is mostly isolated from sea waters, fish guts, sea and lake bottom mud. The general characteristics of *C. botulinum* types included in this classification are given below.
  - Type A: Causes botulism in humans in the eastern United States, more toxic than Type B.
  - Type B: Common worldwide, is a human pathogen.
  - Type C: Poultry, cattle, etc., especially poultry and wild ducks. It is pathogenic in mammals.
  - Type D: Pathogenic in cattle and some other animals, especially in the Republic of South Africa. It does not cause disease in humans.
  - Type E: It lives at the bottom of streams, lakes and seas. It is pathogenic for humans and is more important in aquaculture.
  - Type F: It is similar to types A and B in terms of toxin properties and is a poisoning factor in humans. Isolated in Denmark.
  - Type G: isolated from soil in Argentina, not proven to cause foodborne botulism or typical infant botulism in humans. However, it has been reported to cause sudden death in children in Switzerland.

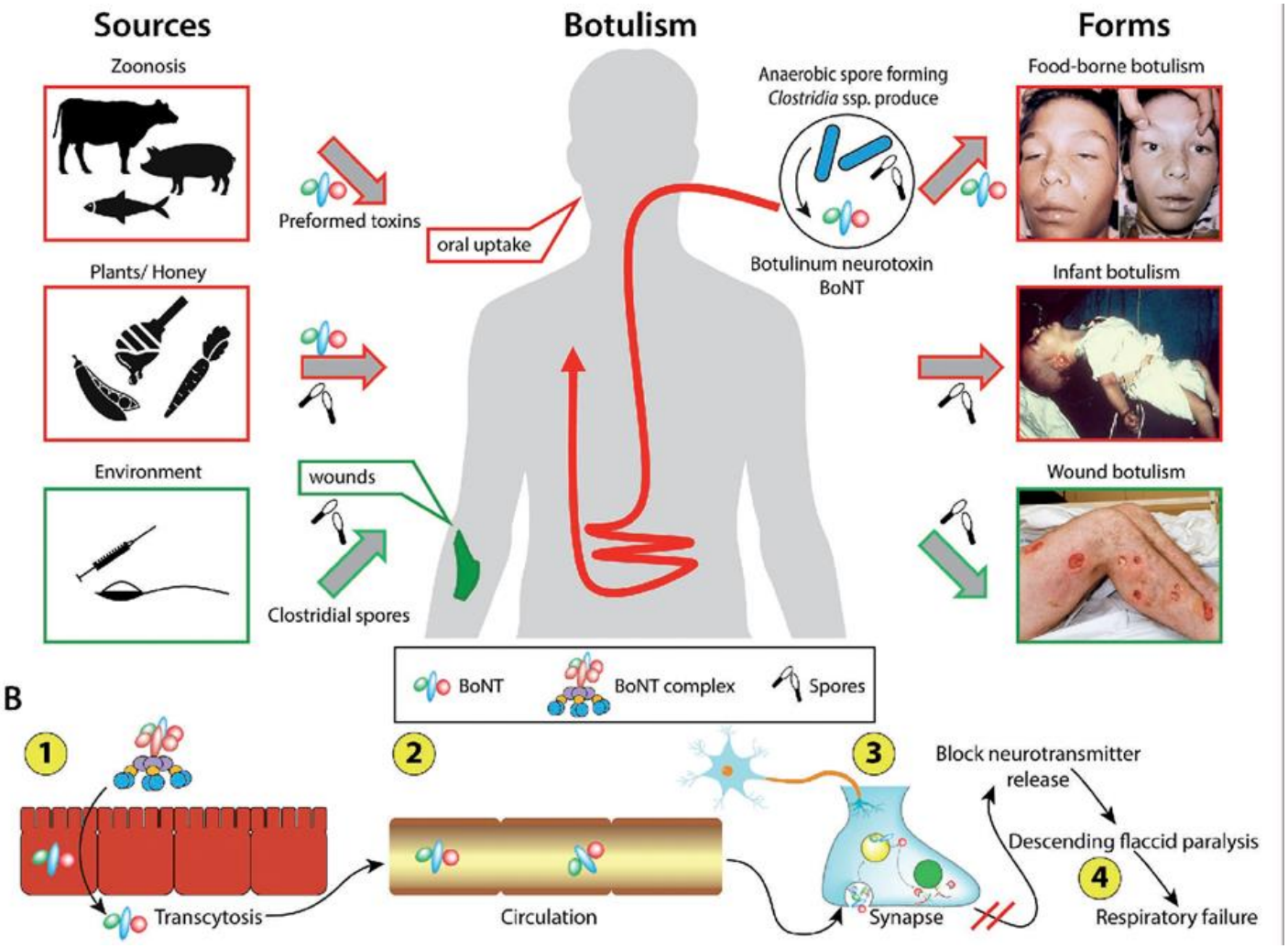
Clostridium botulinum intoxication (Botulism)

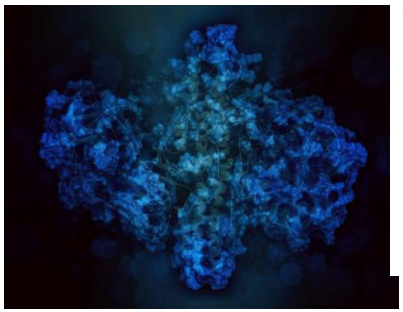
- When *C. botulinum* is classified according to its cultural and physiological characteristics, it is divided into 4 groups.
  - Group I: All strains of type A (proteolytic) and proteolytic strains of types B and F
  - Group II: All strains of type E (not proteolytic) and non-proteolytic strains of types B and F
  - Group III: Types C and D. Both types are non-proteolytic.
  - Group IV: Type G strains, proteolytic activity is weak.
- The growth and toxin production of *C. botulinum* in foods depends on several factors.
  - the type and characteristics of the food (nutrient content, water ratio and aw, pH value, O-R potential, salt content, etc.)
  - storage temperature and time.
- *C. botulinum* grows best at neutral pH. In general, pH 4.7 and below cannot produce toxins. The maximum pH value for growth and toxin formation is 8.89.
- *C. botulinum* generally does not grow below pH 4.5, so acidic foods are considered safe for botulism.
- Botulism cases are known to be caused by some foods such as tomato juice and fruit preserves with a pH below 4.5.

Clostridium botulinum intoxication (Botulism)

- The conditions that allow the development of *C. botulinum* in such foods are explained as follows:
  - The development of microorganisms (*Penicillum, Aspergillus, Mycoderma, Trichosporan* some *Bacillus* species) that will increase the pH in a normally acidic food and the increase of the food pH to a level suitable for the development of *C. botulinum*.
  - In foods whose pH is lowered by adding acid, acidification is not performed homogeneously and regions with higher food pH remain (GDL)
- Meat, fish and low and medium acid canned foods create favorable environments for toxin production. The minimum aw value for growth is 0.94 for A and B types and 0.97 for E type.
- The growth temperature ranges are 10-48°C in A and B types, and 3.3-45°C in E types. Optimum temperatures are between 26-35°C.
- The toxin has a high molecular protein structure and its molecular weight is between 200,000 and 900,000.
- *C. botulinum* neurotoxin is the most potent biological toxin known and 1 gram has the power to kill 10 million people. It has been reported that the LD_{50} value is 1 ng/kg body weight.

Mechanism of foodborne botulism





C. botulinum toxins

Clostridium botulinum intoxication (Botulism)

- The sensitivity to high temperature is an important feature of *C. botulinum* toxin. The maximum formation temperature of the toxin is 35°C.
  - Type A toxin is inactivated within 6-10 minutes at 80°C, and by heat treatment
  - Type B toxin at 90°C for 15 minutes.
  - Spores can be killed in 15 minutes at 115°C.
  - Spores of Type E are inactivated in 15 minutes at 80°C.
  - The toxin is adversely affected by direct sunlight and air. It does not lose its effect even at –79°C in an airless environment and in the dark. It is also resistant to acids and is not affected by stomach acid.
- Poisoning is usually caused by canned meat and vegetables (low acidity), smoked sausage, raw bacon, and pickled fish with a salt concentration of less than 5%.
- The spores of the bacteria germinate where there is a lack of oxygen. Inactive protoxin is released in bacterial colonies. Protoxins are converted into toxins by the action of enzymes that break down protein in the intestines of animals that eat them.
- Toxins cause paralysis at the junction between nerve and muscle, preventing the release of the chemical carrier acetylcholine, which transmits electrical nerve impulses that cause muscle contraction. Boiling food prevents intoxication as the poison breaks down in boiling water within 10 minutes.

Clostridium botulinum intoxication (Botulism)

- Low-acid home canned foods are the leading cause of *C. botulinum* intoxication. A study conducted in the USA reported that 72% of botulism cases were caused by homemade preserves. While canned meat and meat products are the most common group in *C. botulinum* poisonings in European countries, vegetables, especially low-acid canned vegetables (corn, beans, spinach, asparagus, beets) are common in the USA. In Canada, Japan, post-Soviet countries and Iran, fish and fish products usually cause botulism.
- The incubation period in food intoxication is generally 12-36 hours, but it can be between 2 hours and 8 days.
- The shorter the incubation period, the higher the mortality rate. At the beginning of the disease, gastrointestinal symptoms such as nausea, vomiting and diarrhea and weakness, dizziness, headache can be seen. Constipation occurs when the typical symptoms of botulism begin.
- **Botulism**; It starts with weakness and weakness in the muscles, these symptoms are drooping of the eyelid, mydriasis (development of the pupil), blurred and double vision (diplopia), dryness in the mouth, difficulty in speaking and swallowing, feeling of warmth, relaxation of the facial muscles, weakness of the tongue and extremity muscles, paralysis. followed by symptoms such as an increase in Respiration is not regular. Increasing paralysis occurs in involuntary muscles, and in severe cases, death occurs within 3-5 days from respiratory failure as a result of respiratory paralysis. Since death is attributed to respiratory failure, providing artificial respiration at an early stage may save the patient's life.

Clostridium botulinum intoxication (Botulism)

- Death can also occur in periods varying between 24 hours and 2-3 weeks.
- Recovery is possible if the patient passes the 10-day cycle.
- Mortality rate can vary between 30-80% depending on the type of toxin and geographical regions. Full recovery takes months (6 months or longer) if the patient survives. Partial paralysis can last 6-8 months.
- The only known treatment is neutralization of the toxin with antitoxin. Special serums prepared against the toxin of the bacteria are applied. First, polyvalent serum (including types A and B) is used. If it is possible to determine the toxin type, it is treated with a monovalent serum of that type.
- However, treatment often fails after typical symptoms begin. Removal of the existing toxin from the body (emesis, gastric lavage, etc.) and artificial respiration therapy help. Since death is usually seen due to paralysis of the respiratory muscles, artificial respiration may be required by performing a trecotomy.

Clostridium botulinum intoxication (Botulism)

- Points to be considered by the food industry:
  - One of these, and the most important, is the work of the food industry, and the exact application of the prescribed temperature degrees and times for canned foods and foods.
  - All foods that have partially or completely anaerobic parts that will allow the growth of *C. botulinum* must be protected with pH, salt, chemical preservatives (such as sodium nitrite or nitrate in meat products) and other similar antimicrobial applications to inhibit the growth of this bacterium.
- Considerations for the consumer:
  - The most important thing is not to use canned foods with swollen, rusty, damaged, leaking, foamy appearance when opened, discolored and odorless canned foods. Such preserves should not even be tasted for control purposes. This type of suspicious food should be destroyed.
  - Food that has been cooked and kept at room temperature but not reheated should not be eaten. Raw or cooked foods should not be kept at room temperature after they have been frozen and thawed, and those that have been kept or forgotten should not be eaten. Also, canned foods should be stored in a cool, shaded and dry place..

Bacillus cereus intoxication

- *Bacillus cereus* is a bacterium belonging to the *Bacillaceae* family. It is a spore forming, aerobic bacterium. It has peritric flagella. Cell sizes are between 1-2,2 x 3.0-5,0 mm. Optimum growing temperature varies between 28-35°C depending on the strains, but is generally 30°C.
- Gram-positive, rod shaped, motile, aerobic, endospore. Spores of *B. cereu*s are relatively heat-resistant. Widely distributed in the environment: soil, dust, water. Foods: meat, eggs, rice, pasta, vegetables and dairy products.
- *B. cereus* is very common in milk and dairy products and is a thermoduric dairy bacteria. Its spores remain viable after 30 minutes of pasteurization at 63°C. In the absence of contamination with spore and vegetative cells and post-pasteurization, *B. cereus* is the most important organism affecting milk quality under room conditions. This organism causes sweet coagulation in milk, as well as a deterioration called bitty cream.
- Although it is very common in milk, the most important reason for the low incidence of *B. cereus* poisoning in dairy products is that these products are not consumed as a result of the feeling of spoilage in milk and dairy products in the presence of high numbers of *B. cereus*. Apart from milk, it is often isolated from the surfaces of rice, other grains, starch, spices, dry foods, meat and chicken.
- *B. cereus* was first reported by Hauge (1950-1955) as an etiologic agent in four separate cases of food poisoning affecting 600 people in Norway. In all four cases, the food that caused the poisoning was vanilla sauce that was prepared and kept for one day. After that, *B. cereus* cases were detected in many European countries.

Bacillus cereus intoxication

- A very large food group is the source of *B. cereus* food poisoning. Especially starchy foods that are kept for a while before serving are susceptible to *B. cereus* poisoning.
- The high number of organisms isolated from foods indicates the presence of a pre-incubation period. *B. cereus* food poisoning is an intoxication and it synthesizes extracellular toxin after the number of bacteria reaches a certain level.
- Toxin formation by *B. cereus* depends on the presence of certain nutrients in the environment. The organism synthesizes and secretes its toxin during the logarithmic phase.
- *B. cereus* toxin is sensitive to trypsin and pepsin enzymes and becomes inactive with a 0.01% dose of these enzymes in 60 minutes at 37°C. The toxin does not lose its activity in 30 minutes at 45°C, but becomes inactive in 30 minutes at 56°C.
- The emetic (vomiting) toxin, on the other hand, is resistant to heat and pH, and resistant to trypsin and pepsin enzymes. It can be produced in a range of 15-50°C, but the optimum temperature is 35-40°C.

Bacillus cereus intoxication

- *B. cereus* causes two types of foodpoisoning:
  - Emetic type: emetic enterotoxin is preformed in foods, cyclic peptides. Incubation period 1-6 h generally 2-5 h, syptoms: nausea, vomiting, malaise, diarrhea.
    - Foods: fried and cooked rice, paste, pizza.
  - Diarrheal type (toxicoinfection): symptoms appear within 8 to 24 h ingested cells survive in stomach, multiply and produce spores; release enterotoxin.
    - Diarrheal enterotoxin(s): is a heat-label protein: hemolysin, nonhemolytic toxin and cytotoxin
    - Foods: grain foods, especially corn and corn starch, mashed potatoes, vegetables, minced meat, some meat products, pudding and soups .
    - Symptoms: abdominal pain, watery diarrhea, nausea, sometimes vomiting.
- In the prevention of *B. cereus* poisoning, it is important to prepare foods at risk for this bacteria in small portions just before consumption and to undergo adequate heat treatment. Among the measures that can be taken are the rapid cooling of the foods that will not be consumed immediately and the cold storage and, if reheating is to be applied, the heating process to exceed 74°C.

Foodborne Toxico-infections

- The disease that occurs as a result of the consumption of food containing live microorganisms and the production of toxins in the human body by the microorganism is called toxicoinfection.
- Features of foodborne toxicoinfection are:
  - If the bacteria are spores, a large number of cells must be consumed. Cells of spore-forming bacteria do not multiply in the digestive tract, but produce toxins.
  - Large numbers of cells are not required for gram negative bacteria. Because few cells multiply rapidly in the digestive tract and secrete toxins.
  - The ingested cells secrete toxins, even if they are dead cells.
    - Such as
      - *Enteropathogenic Escherichia coli (EPEC)*
      - *Enterotoxigenic Escherichia coli* (ETEC)
      - *Clostridium perfringens*
      - *Bacillus cereus*
      - *Vibrio cholerae*
      - *Aeromonas hydrophila*

Enteropathogenic *Escherichia coli* species

- *Escherichia coli* species are usually commensal in the intestines of humans and warm-blooded animals. However, some strains are pathogenic and cause diarrhea. These are examined under four main groups: enteropathogenic (EPEC), enteroinvasive (EIEC), enterotoxigenic (ETEC) and enterohemorrhagic *Escherichia coli* (EHEC).
- EPEC: A disease seen in children in developing countries. It causes diarrhea. Diarrhea looks like rice water and has mucus. Pork and meat pie cause infection.
- ETEC: It is known as the tourist disease. It is caused by water contaminated with fecal material and food washed with this water. The infective dose is 10^6. It produces a toxin similar to *Vibrio cholera*. It produces two enterotoxins that are resistant and sensitive to high temperature.
- EIEC: *Shigella dysenteriae* produces a toxin-like toxin. The infective dose is 10^6-10^8 cells.
- EHEC: The most common species is *E. coli O157:H7. E. coli O157:H7* has been reported to cause mainly three conditions: Hemorrhagic colitis, especially in children, hemolytic uremic syndrome (HUS) and thrombotic thrombocytopenic purpura (TTP).

Enteropathogenic *Escherichia coli* species

- In hemorrhagic colitis, sudden abdominal cramps begin, followed by watery diarrhea within 24 hours. Then comes the period when most of the stool consists of blood. During this time, vomiting may occur, fever is absent or very little. The duration of the disease is usually between 2-9 days.
- Hemolytic uremic syndrome (HUS) is the cause of acute kidney failure in children. This situation occurs when the coagulation mechanism in endothelial cells is activated by the effect of toxins, and small clots are formed, which block the vessels of the kidneys and other organs and cause waste materials to be stored in the blood. As a result, patients need blood transfusion and dialysis, heart failure, seizures and coma can be seen.
- Thrombotic thrombocytopenic purpura is similar to HUS except that it is related to the nervous system. It is a syndrome usually seen in adults and causes anemia, irregular neurological symptoms, fever and mild azotemia. It often results in death as blood clots usually form in the brains of patients. This microorganism can rarely cause bleeding bladder inflammation, balantitis, convulsions, sepsis and anemia with other organisms.

Enterotoxigenic ***Escherichia coli*** **(ETEC)**

- The difference of ETEC from pathogenic *E. coli* is that the cells multiply and produce toxins after ingestion. ETEC is the leading cause of traveler's diarrhea. It is popularly known as the "**tourist disease**". Symptoms of the disease are similar to cholera.

Properties

- ETEC is a gram-negative, non-spore-forming, motile, facultative anaerobe, short rod-shaped bacterium. The optimum growth temperature is 30-37 ℃. Its cells can be destroyed at pasteurization temperature.

Sources

- Water and food contaminated with ETEC cause this disease. Vegetables, salads, potatoes, cheese, fish, meat and meat products are risky foods. ETEC is transmitted to water and food via feaces due to poor hygiene. In addition, inadequate cooking and improper storage conditions are among the most important causes of the disease.

Enterotoxigenic *Escherichia coli* (ETEC)

Symptoms

- Symptoms begin 1-3 days after consumption of the relevant food.
- Excessive watery diarrhea and vomiting are the most prominent symptoms of the disease.
- Complaints may take 3-4 days. In severe cases, dehydration and shock may occur with diarrhea.

Prevention

- To prevent ETEC infection, the following rules should be observed:
  - The most important rule is to act in accordance with the rules of hygiene and sanitation.
  - Food must be cooked very well. Cooked foods should be stored at appropriate temperatures.

Clostridium perfringens

The disease caused by *C. perfringens* after consumption through food is called **perfringens food poisoning**. The symptoms of Perfringens food poisoning are mild and the disease passes quickly. Since the disease is not usually registered by doctors, the number of cases in official records seems less than it is.

Properties

C. perfringens is a gram-positive, anaerobic, rod-shaped, non-motile and spore-forming bacterium. The optimum growth temperature is 45 ℃. 37℃ is the optimum temperature for sporulation and toxin production. It cannot grow at refrigerator temperature. Bacterial cells are heat sensitive. It can be destroyed at pasteurization temperature. However, its spores are heat resistant. It can survive for hours at 100 ℃.

Sources

C. perfringens occurs naturally in the intestines of humans and animals. It contaminates food through sewage, human and animal feces. This poisoning is caused by protein foods. Meat and meat products, soups containing broth, mince pies and salads are among the food products that cause poisoning. Since *C. perfiringens* is an anaerobic bacterium, it can multiply due to reduced oxygen during cooking.

Poisoning usually occurs after meals that have been cooked 1-2 days before, kept in the refrigerator, and consumed by reheating. In addition, foods that are produced in large quantities and left to cool slowly and foods that are kept waiting until service and insufficiently heated are risky.

Clostridium perfringens

Symptoms

- Symptoms of the disease begin 8-15 hours after the consumption of food. Abdominal pain, diarrhea and fatigue are seen. Within 24 hours, the disease ends on its own. It is a mild disease. For this reason, a doctor is rarely consulted.

Prevention

- To prevent Perfringens poisoning, the following should be considered:
  - Foods should be cooked very well and consumed without delay.
  - If the food is not to be consumed immediately after cooking, it should be kept in the refrigerator.
  - Food in the refrigerator should be consumed after it has been heated very well. Hygiene rules should be observed.

Bacillus cereus

Since most countries do not report food poisoning associated with *B. cereus*, data on the disease are very limited. *B. cereus* poisoning is also called rice syndrome because it is usually caused by rice dishes that have been left at room temperature.

Properties

- *B. cereus* is a gram-positive, spore-forming, motile and aerobic bacterium. The optimum growth temperature is 35-40 ℃. Cells can be destroyed at pasteurization temperature. However, its spores remain viable at high temperatures.

Sources

- *B. cereus* is a soil-borne bacterium. Therefore, it is commonly found in vegetables and fruits. Salad, meat, milk and dairy products, sauces, cereals, legumes, pasta and rice are risky foods.

Symptoms

- Symptoms of *B. cereus* poisoning appear 8-24 hours after consumption of the relevant food. A rapid recovery is seen. The most important symptoms are diarrhea, abdominal pain and nausea. Its symptoms are very similar to those of perfringens disease.

Prevention

- To prevent *B. cereus* poisoning, the following should be considered:
  - Cereals and dried legumes should never be stored in a humid and hot environment.
  - Rice dishes should be cooked in the amounts to be consumed daily. Reheating should not be done more than once.
  - Care should be taken to cook the food very well.
  - If the dishes are to be kept after cooking, they should be kept either at refrigerator temperature or above 60 ℃.
  - *B. cereus* cells can also proliferate at refrigerator temperature. Therefore, food should not be kept in the refrigerator for a long time.
  - Attention should be paid to personal hygiene and kitchen hygiene.

Bacillus cereus

Foodborne outbreaks

- *B. cereus* causes two types of food poisoning:
  - the emetic type and the diarrheal type.
- The **preformed emetic toxin** during vegetative growth of cells in food results in vomiting.
- **Ingestion of living** *B. cereus* cells that may produce enterotoxins in the small intestine, cause dierrheal illness.
  - Large numbers of cells or spores (10^5 to 10^8) need to be ingested together with food to produce gastroenteritis.
  - Diarrheal symptoms appear within 8 to 24 h (mostly within 12 h).
  - Symptoms include abdominal pain, watery diarrhea and may be nausea, sometimes vomiting.
  - Three different enterotoxins can cause food poisonings: hemolysin (lyse erythrocytes), nonhemolytic toxin ans cytotoxin.

Vibrio cholerae

V. cholera causes cholera disease and can cause epidemics with high mortality rates. From 1814 to 1992, there were seven major cholera epidemics in the world. Today, cases of cholera continue to be seen.

Properties

- *V. cholerae* is a gram-negative, non-sporeless, motile, facultative anaerobic or aerobic, S-shaped bacterium.
- It is heat sensitive and cold resistant. The optimum growth temperature is 30-37 ℃. While it cannot develop in acidic foods, alkaline foods are very suitable for its development. It can be destroyed at pasteurization temperatures. It can maintain its vitality at 5-10 ℃ in cooked foods.

Sources

- Like all *Vibrio* species, *V. cholerae* is associated with sea water and seafood. However, the main source of cholera disease is mostly through food and water contaminated with faeces of sick people. Undercooked fish, oysters, crab and shrimp are risky foods.

Vibrio cholerae

Sources

- *V. cholerae* is widely distributed in aquatic environments.
- The most common associated foods with cholera are crabs, shrimp, raw fish, oysters, clambs, rice, cooked rice, frozen coconut milk, raw vegetables and fruits.

Symptoms

- Symptoms appear between 12 hours and 5 days after eating water or food contaminated with *V. cholerae*. The most important symptom is watery diarrhea. Along with this, vomiting, weakness, muscle cramps and confusion are seen.
- Many people with cholera are carriers. He is not affected by the disease and does not show any symptoms. However, it infects the environment through feces and infects other people.

Prevention

- In order to prevent cholera disease, it is necessary to pay attention to the following points:
  - Cholera can be prevented by following the hygiene rules.
  - Since most of the epidemics are caused by water, care should be taken to clean the waters.
  - It is necessary to pay attention to the fact that the food is well cooked.
  - Seafood should not be consumed raw.
  - Food should be stored in the refrigerator. In addition, the freezing process reduces the number of *V. cholerae* in the food**.**

Aeromonas hydrophila

- *A. hydrophilia* are common inhabitants:
  - in salt and freshwater and environments during warmer summer.
- *A. hydrophila* presents in the *Vibrionaceae* family, is a Gram-negative bacilli, facultatively anaerobic, catalase- and oxidase-positive, glucose fermenting, motile (*A. media* and *A. salmonicida* non-motile) by polar flagella (monotrichous), mesophilic or psychrophilic.
- It does not grow at high salt concentration (<5 % NaCl).
- It grows optimally at 28°C.
- It can produce mildly heat-resistant extracellular lipase or protease enzymes.
- It is common in seawater, well water, sink water, sewage and a wide range of seafood, including fish (both wild and farmed), shrimps (raw and cooked), oysters, crabs, scallops, vegetables.

Aeromonas hydrophila

- The symptoms appear within 24 h following ingestion of foods containing viable *A. hydrophila* cells.
- Infective dose of *A. hydrophilia* ranges from 10^4 to 10^{10} cells.
- The most common symptom is watery diarrhea.
  - Others: fever and vomiting.
- Generally the disease is self-limiting and lasting within 2 weeks.

Prevention and control

- Foods stored at low temperature (below 5°C),
  - sufficiently cooking seafoods, disinfections of drinking water.
- Organic acids and inorganic acids are effective on *A. hydrophilia*.
- The vegetative cells of *A. hydrophilia* are easily destroyed by heating at 56°C for 10 min.
- *Aeromonas* spp. produce heat-stable exotoxins and exoenzymes which may survive at this heat treatment.

MOLD INTOXICATIONS FROM FOOD

MYCOTOXICOSIS

- Mycotoxins are secondary metabolic products secreted by molds that do not show antigenic properties and cause disease in humans and animals. Intoxication caused by the consumption of food and feed contaminated with mycotoxins is called mycotoxicosis.
- At least 14 of mycotoxins have carcinogenic effects, and most of them cause various acute or chronic diseases.
- Mycotoxicosis such as toxic leukopenia caused by *Fusarium* species, ergotism caused by *Claviceps* species, yellow rice poisoning caused by *Penicillium* species have historical importance and they have caused diseases in epidemics in the past, especially in Russia, Japan and Europe.
- Today, more than 300 mycotoxins are known and the most important of the mycotoxin producing genera are *Aspergillus, Penicillium, Fusarium* and *Alternaria.*

MYCOTOXICOSIS

- With the discovery of aflatoxin, secondary metabolites with toxic effects have gained importance and a very large group of substances on which numerous researches have been carried out for 40 years has been formed.
- At the point reached today, in order to protect people from the effects of this toxic group, the highest (tolerable) amounts of mycotoxins that can be found in food and feed are determined by legal regulations.
- The number of molds capable of synthesizing mycotoxins is around 350. However, this number is quite low among all molds.
- The vast majority of the thousands of mold species tested did not produce mycotoxins.
- Derivatives in the aflatoxin group are around 18.
- Ochratoxins also include 7 compounds with structural similarity. But the most important is OA (Ochratoxin A).
- There are 40 derivatives of trichothecenes, and it is claimed that 150 compounds are included in this group.
- Alternaria toxins also exhibit over 30 different metabolites.

MYCOTOXICOSIS

MOULD	COLOR	TOXIN PRODUCER	COMMENTS
Penicillium	Green to green-blue	Yes - Ochratoxin, Citrinin, Patulin	Several potential toxins associated with certain species. Most common toxin producer in silage.
Aspergillus	Yellow-green	Yes - Aflatoxin, Ochratoxin	Found in drought, heat-stressed conditions or insect infested fields.
Fusarium	White to pinkish - white	Yes- Zearalenone, Vomitoxin (DON), T-2 Toxin, Fumonisin	Common in cold, wet seasons. Certain strains produce extremely potent toxins.
Mucor	White / Grey	None	Found especially in sealed corn. Grows at low temperatures. Also found in manures and soils.
Rhizopus	Black/white	None	Requires high moisture and an advanced decay mould. Common bread mould.
Cladosporium	White	None	Produces yeast-like symptoms. Grows at low temperatures.

MYCOTOXICOSIS

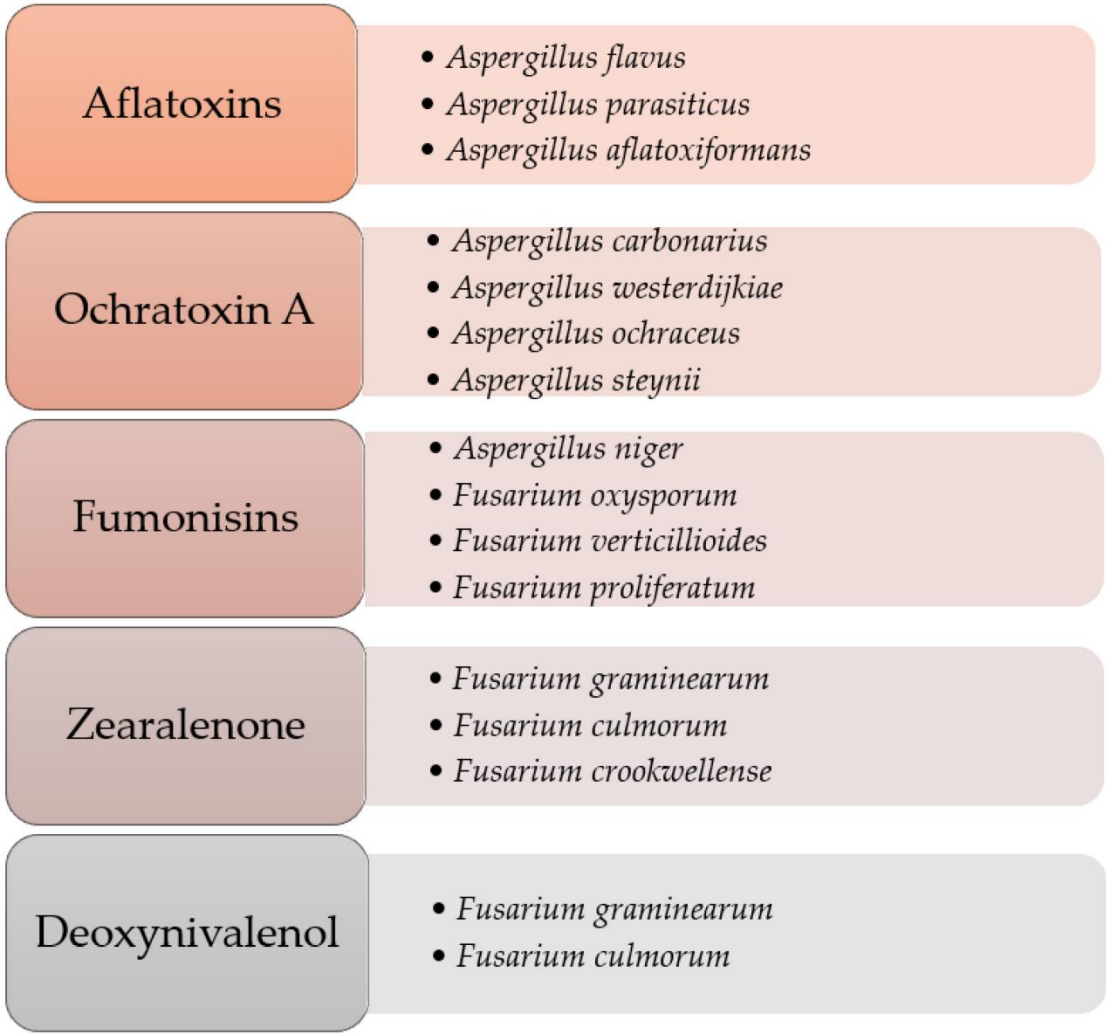
Mycotoxin	Molds	Foods	Toxicities
Aflatoxins	*A. flavus,* *A. parasiticus* *A. nomius*	Peanut, nuts, wheat, corn, rice, pepper, milk, spices, dried fruits, soybean.	Hemorrhages, carcicinogens, alterating digestive tract.
Citrinin	*P. citrinum* *P. expansum* *A. oryzae*	Wheat, barley, rye, oats, corn, rice.	Nephrotoxin (kidney damage), inhibit respiration.
Ochratoxin	*A. ochraceus* *P. cyclopium* *P. viridicatum*	Cereal grains, peanuts, wine, beer, coffee, dried fruits.	Hepatotoxin (toxicity on kidney), kidney tumor, liver damage, enteritis.
Patulin	*P. expansum* *P. patulum*	Feed, apple juice, fruit, vegetable.	Brain and lung hemmorrhage, convulsions, carcinogenesis.
Roquefortin	*P. roqueforti*	Blue cheese.	Neurotoxin, liver damage, hemorrhagic in digestive tract.
Zearalenone	*F. culmorum* *F. gramin.*	Corn, cereal crops.	Enlargements of mammary glands, estrogenic effects.

MYCOTOXICOSIS

Mycotoxins	Fungus	Health Effect	Source
Aflatoxins B1,B2,G1,G2, (M1)	Aspergillus and penicillium species	Hepatotoxin carcinogen immunosuppressant	Cereals, maize or corn, tree nuts, ground nuts
Ochratoxins: A, B, C, α, β, γ	Aspergillus and penicillium species	Nephrotoxin, immunosuppressant, renal, and hepatic carcinogens	Vegetables, cereals, vine fruits, wine, beer, coffee
Trichothecenes Toxins: NT-1, NT-2, T-2, HT-2 Diacetoxicirpeaol (DAS) Nivalelol (NIS) Deoxynivalenol (DON) Fusarenon-X	Fusarium species and Gilerellazeac species	Hepatotoxin carcinogen immunosuppressant, neurotoxin, necrosis, malabsorption, skin issues	Grains, rice, oats, cereals, maize or corn
Fumonisins: B1, B2, B3	Fusarium species	Esophagus cancer, liver cancer neurotoxic	Maize or corn, cereals, grains, beer, garlic, beans, asparagus
Zearalenone	Fusarium species	Reproductive issues, infertility, early development	Corn wheat, oats sorghum, sesame seed, hay, silage
Cyclopiazonic acid	Aspergillus and penicillium species	Convulsions	Cheese, maize or corn, ground nuts
Citrinin	Aspergillus and penicillium species	Neurotoxic, nephrotoxic, carcinogen	Rice, cheese, soy, maize or corn, cereals, grains
Patulin	Aspergillus and penicillium species	Neurotoxin, mutagenic, hepatotoxic, immunosuppressant	Fruits, juices, vegetables
Ergot Alkaloids	Claviceps species	Digestive disorders, nervous disorders, seizures, vomiting, headaches	Cereals, grains, maize or corn

MYCOTOXICOSIS

MOULD	MYCOTOXIN
Aspergillus	Aflatoxin
	Ochratoxin
Penicillium	Ochratoxin
Fusarium	DON (Deoxynivalenol)
	ZON (Zearalenone)
	T-2 Toxin
	Fumonisin



MYCOTOXICOSIS

Mycotoxins	Producing microorganisms	Foods mainly affected	Toxic impacts/effects
Aflatoxins	*Aspergillus flavus, Aspergillus parasiticus*	Nuts, groundnuts, pistachios, cereals, millet, also milk to a secondary degree	Hepatotoxic, mutagenic, carcinogenic, teratogenic
Citrinin	*Penicillium citrinum*	Barley, oats, maize, rice, walnuts	Nephrotoxic, mutagenic, carcinogenic (?)
Cyclopiazonic acid	*Aspergillus flavus, Penicillium cyclopium*	Groundnuts, maize, cheese	Neurotoxin
Deoxynivalenol (Vomitoxin)	*Fusarium graminearum, Fusarium culmorum*	Wheat, barley, maize, rye	Neurotoxin, immunotoxic, carcinogenic (?)
Ergot alkaloids	*Claviceps purpurea*	Rye, wheat, oats	Neurotoxin, circulatory disturbances, carcinogenic
Fumonisins	*Fusarium verticolloides*	Maize and products containing maize	Neurotoxin, hepatotoxic, harmful to the lungs, carcinogenic (?)
Moniliformin	*Fusarium verticolloides (moniliforme)*	Maize and products containing maize, wheat (cereals)	Harmful to the heart, hepatotoxic, nephrotoxic
Ochratoxin A	*Aspergillus ochraceus, Penicillium verrucosum*	Cereals, beans, green coffee beans	Carcinogenic, teratogenic, neurotoxic, nephrotoxic
Patulin	*Penicillium patulum, Aspergillus clavatus*	Apples, pears, beans, wheat	Carcinogenic, mutagenic, hepatotoxic
Penicillic acid	*Penicillium puberulum, Aspergillus ochraceus*	Maize, barley, beans	Neurotoxic
Sterigmatocystin	*Aspergillus versicolor, Aspergillus nidulans*	Cereals, green coffee beans, cheese	Teratogenic, skin irritant, carcinogenic (?)
T-2 Toxin/ HT-2-Toxin	*Fusarium sporotrichioides, Fusarium poae*	Maize, barley, oats, millet	Strong neurotoxin, skin irritant, carcinogenic
Zearalenone	*Fusarium graminearum*	Cereals	Mucous membrane irritant, estrogenic, causing infertility, carcinogenic (?)

TOXINS PRODUCED BY FUSARIUM SPECIES

- Many species of the *Fusarium* genus produce mycotoxins.
  - Of these toxins, there are over 80 derivatives of the group called "trichothenes", of which T-2 toxin, deoxynivalenol (DON= vomitoxin) and a few others have been naturally isolated from cereal products and feed.
  - Other mycotoxins produced by *Fusarium* species include toxins such as zeralenone, moniliformin, fusarin C, and fumonisin. One of the most important fusarium toxins is trichothecene, which causes toxic leukopenia.

Fumonisin

- Although fumonisin is predominantly seen in maize, it can also be seen in wheat with embryo darkening disease. Fumonisin was first isolated in 1988 as a metabolite of *Fusarium verticillioides* (*moniliforme*). 6 types of fumonisin have been distinguished. Fumonisin B1 (FB1) is thought to be the most toxic.

TOXINS PRODUCED BY PENICILLIUM SPECIES

Rubratoxin

- Rubratoxin, a mycotoxin produced by *Penicillium rubrum*, causes a hemorrhagic disease in cattle, pigs and chickens. This toxin, which can be found widely in nature and soil, has been isolated from peanuts, corn, legumes and sunflower.

Patulin

- This toxin was first isolated as an “antibiotic” and its antibacterial and antifungal effects were reported in various studies. It is considered as a potential carcinogen in humans after its carcinogenic and mutagenic effects were determined in mice.
- It produces many types of patulin, especially *Penicillium expansum*. However, the most toxin-producing species in fruits is *P. patulum*. Apple products such as apple juice, apple cider vinegar and fruits (banana, pear, peach and pineapple) are among the foods in which patulin is most isolated.
- Patulin is a heat-resistant toxin and remains stable after boiling at 100°C for 15 minutes. Therefore, the pasteurization process applied to fruit juices does not inactivate this toxin.

TOXINS PRODUCED BY ASPERGILLUS SPECIES

Ochratoxin

- Ochratoxins are toxic metabolites produced by different species and strains of *Aspergillus* group molds, especially species belonging to the *A. ochraceus* group, and the most toxic of them is Ochratoxin A (OA).
- Ochratoxins are isolated from many foods, especially cereals and legumes. Among the places where it is isolated; foods such as peanuts, red and black pepper, corn, wheat, coffee, nuts, bread, citrus fruits, some fermented fish products, moldy smoking tobacco and animal feeds, and animal tissues or fluids such as pig kidney, pig serum.

Aflatoxin

- Aflatoxins, a disease that killed more than 100,000 turkeys on a chicken farm in England in 1960, emerged. It was determined that the disease was caused by the molds in the peanut flours added to the animal feeds, and in the researches, it was determined that this mold was *Aspergillus flavus*, and the toxin made was named aflatoxin.
- Although it was stated in later studies that some other mold species, especially *A. flavus* and *A. parasiticus*, can produce aflatoxin, according to the latest findings, only *A. flavus* and *A. parasiticus* are molds that produce aflatoxin.
- Aflatoxin can be found in all kinds of foods where molds that produce aflatoxins grow.
- The foods in which aflatoxins are most commonly isolated are; peanuts, corn and grain. Apart from these, the foods in which aflatoxins are produced include oilseeds, cereals, dried fruits, red pepper and potatoes. Toxin is also isolated from the milk of animals fed with aflatoxin feed.