

BACHELOR OF VETERINARY MEDICINE

PRINCIPLES OF FOOD MICROBIOLOGY

LECTURE NOTES

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INTRODUCTION

Humans obtain food from animals and plants, and these must be healthy. Foods of animal origin are made up of water, carbohydrate, proteins and various minerals and vitamins. Foods of animal and plant origin are highly perishable. Food production occurs in specific areas and at certain periods of the year due to variation in weather conditions. Food has therefore to be collected and stored for use during periods of low or no food production or has to be stored /packed and transported over long distances to reach available markets

Generally, foods when fresh are free of microorganisms (bacteria, fungi, viruses etc), while food surfaces may have a low load of microorganisms. Food spoilage is complicated by the fact that food begins to deteriorate shortly after harvesting, gathering on slaughtering.

FOOD SPOILAGE

Food spoilage is defined as damage or injury to food rendering it unsuitable for human consumption. Food must be considered spoiled if it is contaminated with any pathogenic microorganisms or poisonous agents, e.g. Pesticides, heavy metals etc.

In most cases, there does not need to be an evident sign of spoilage. The food might look normal and only after eating it or by careful bacteriological and /or toxicological investigation, one is able to realize the defect. Food decay or decomposition is implied when the term 'spoiled' is used.

Table 1. Storage life of various foods

<u>Food product</u>	<u>Storage life (days) at 21°C</u>
Raw beef and mutton	1 – 2
Raw fish	1 – 2
Raw poultry	1 – 2
Dried salted or smoked beef meat, mutton or fish	360 or more
Fresh fruits	1 -7
Dried fruits	360 or more
Leafy vegetables	1 - 2
Root crops	1 - 20
Dried seeds	360 or more

CAUSES OF FOOD SPOILAGE

Food spoilage is ascribed to various causes;-

- (i) Growth and activity of microorganisms:- Bacteria, yeast and moulds are the key microorganisms that cause food spoilage. They produce various enzymes that digest the various constituents of food.
- (ii) Enzymes activity: This refers to the action of enzymes inherently found in animal or plant tissues. These enzymes digest the various food components after the death of plant or animal.

- (iii) Chemical reactions: - These are the chemical reactions which occur in plant or animal tissues, but not catalyzed by enzymes e.g. Oxidation of fats – causing rancidity.
- (iv) Vermin: vermin includes; weevils, ants, rats, cockroaches, mice, birds, larval stages of some insects. Vermin are important due to:
 - (a) Aesthetic aspect of their presence - e.g. weevils unsightly in food grains; maize and beans
 - (b) Possible transmission of pathogenic agents e.g. Salmonellosis by rats
 - (c) Consumption of food
- (v) Physical changes - These include those changes which are caused by freezing, burning, drying, pressure etc.

Microbial spoilage of foods

Bacteria, yeasts, and molds are the major causes of food spoilage. They produce various enzymes that digest various chemical constituents of food.

Molds are the major cause of spoilage of foods with reduced water activity e.g. cereals and cereal products, meats etc.

Bacteria are involved in spoilage of food with relatively high water activity eg milk and milk products, meat etc.

PRIMARY SOURCES OF MICROORGANISMS IN FOODS

There are eight primary environmental sources of microorganism in foods, namely;

- (i) **Soil and water:** - The two environments are placed together because many of bacteria and fungi that inhabit both have a lot in common. Soil organisms may enter the atmosphere by the action of the wind and later enter water bodies when it rains. They also enter water, when rain water flows over soil into bodies of water. Aquatic organisms can be deposited onto soil through cloud formation and subsequent rainfall. This common cycling results in soil and aquatic organisms being one and the same to a large degree.
- (ii) **Plants and plant products**
 It may be assumed that many or most microorganisms found in the soil and water will contaminate plants. Only a relatively small number of the microorganisms find the plant environment suitable for their over – all well – being. Those that persist on plant products do so by virtue of a capacity to adhere to plant surfaces so that they are not easily washed away and because they are able to obtain their nutritional requirements from these surfaces. Notable among these are the lactic acid bacteria and yeasts. Others are; *Corynebacteria*, *Pseudomonas* and *fungal* pathogens.

- (iii) **Food utensils**
When vegetables are harvested in containers and utensils, some or all of the surface organisms on the products contaminate contact surfaces. The cutting block in a meat market along with cutting knives and grinders are contaminated from initial sample, and this process leads to a buildup of organisms – this ensures a constant level of contamination by meat-borne organisms.
- (iv) **Gastro – intestinal tract – of man and animals**
This biota becomes a water source when polluted water is used to wash raw food products. The intestinal biota consists of e.g. Salmonellae. Any or all of the Enterobacteriaceae may be expected in faecal waste, along with intestinal pathogens, including some protozoa – pathogens in fecal waste, along with intestinal pathogens, including some protozoa e.g. E. histolytica, Giardia, lamblia, Toxoplasma gondii, Cryptosporidium pavum, Cyclospora cayetanensis.
- (v) **Food handlers:** The microorganism on the hands and outer garments of food handlers generally reflect the environment and habits of individuals. The organisms may be those from soil, water, dust and other environmental sources. Additional sources are the organism found in nasal cavities, the mouth, and on the skin and those from the gastro intestinal tract that may enter food through poor personal hygiene practices.
- (vi) **Animal hides and skins;** In the case of milk cows, the type of organism found in raw milk can be a reflection of the biota of the udder when proper milking procedures are not followed in milking and of the general environment of such animals. From both the udder and skin, organisms can contaminate the general environment, milk containers and the hands of handlers.
- (vii) **Animal feeds -** This is a source of Salmonellae to poultry and often from animals. In the case of silage, it is a known source of Listeria monocytogenes to dairy and meat animals. The organisms in dry animal feeds are spread throughout the animal environment and may be expected to occur on animal hides.
- (viii) **Air and dust –** The organisms found in air and dust, include most of gram positive bacteria, and among the fungi, a number of molds may be expected to occur in air and dust, along with some yeasts.

Table 2 – Relative Importance of Eight Sources of Bacteria and Protozoa to Foods

Organism	Soil and water	Plant/ product	Food utensils	Gastro intestinal tract	Food handlers	Animal feed	Animal hides	Acid and dust
<u>Bacteria</u>	Xx	X	-	-	-	-	-	-
<u>Aeromonus Bacillus</u>	Xx	X	X	-	X	X	X	X
<u>Compylobacter</u>	-	-	-	Xx	X	-	-	-
<u>Clostridium</u>	xx	X	X	X	X	x	x	xx
<u>Corynebacteria</u>	Xx	X	X	-	X	-	X	Xx
<u>Escherichia</u>	X	X	-	Xx	X	-	-	-
<u>Salmonella</u>	-	-	-	Xx	-	XX	-	-
<u>Staphylococcus</u>	-	-	-	X	Xx	-	X	-
<u>Vibrio</u>	Xx	-	-	X	-	-	-	-
<u>Protozoa</u>								
<u>E. histolitica</u>	Xx	-	-	X	X	-	-	-
<u>G.lambliia</u>	Xx	-	-	X	X	-	-	-
<u>T. gondii</u>	-	X	-	Xx	-	-	-	-

Xx – indicate a very important source

FACTORS AFFECTING MICROBIAL GROWTH IN FOOD

The factors affecting microbial growth fall into two categories; the Intrinsic factors and the Extrinsic factors.

(a) THE INTRINSIC FACTORS

These are the parameters (factors) of plant and animal tissues that are an inherent part of the tissues. These parameters are as follows:-

- (i) Hydrogen ion concentration (pH)
- (ii) Moisture content
- (iii) Oxidation-reduction Potential (Eh)
- (iv) Nutrient content of the food
- (v) Antimicrobial constituents (Substances)
- (vi) Biological structures.

(i) Hydrogen ion concentration (pH)

Most bacteria grow best at neutral or weakly alkaline pH, usually 6.6 – 7.5. Few bacteria may grow at pH below 4.0. Some bacteria can grow within a pH of 4.5 and 9.0 e.g. *Salmonella*. Bacteria tend to be more fastidious in their relationships to pH than molds and yeasts, with the pathogenic bacteria being most fastidious. Molds grow at pH of 1.5 – 11.0, while yeast grows at pH range of 1.5 – 8.5.

Fruits, soft drinks, vinegar and wines all have pH values all of which fall below the point at which bacteria normally grow. The excellent keeping quality of these products is due in great part to pH. It is a common observation that fruits generally undergo mold and yeast spoilage, and this is due to the capacity of these organisms to grow at pH values of

less than 3.5. Most of the meats and sea foods have a final ultimate pH of about 5.6 and above. This makes these products to be susceptible to bacteria as well as mold and yeast spoilage. Meat from fatigued animals spoils faster than that from rested animals, and this is a direct consequence of final pH attained upon completion of rigor mortis. Upon the death of a well rested animal, the usual 1% glycogen is converted to lactic acid, which directly causes a depression of pH values from about 7.4 to about 5.6 depending on the type of animal. The lowest pH value for beef is approx. 5.1 and the highest approx. 6.2 after rigor mortis.

Table 3: pH value of some food products

Food type	Range of pH value
Beef	5.1- 6.2
Chicken	6.2 - 6.4
Milk	6.3 - 6.8
Cheese	4.9 - 5.9
Fish	6.6 - 6.8
Oysters	4.8 - 6.3
Fruits	<4.5 (most < 3.5)
Vegetables	3.0 - 6.1

Microorganisms that are able to grow in acid environment are called acidophilic microorganism. These microorganisms are able to grow at pH of around 2.0. Yeasts and molds grow under acid conditions. Other microorganisms such as *Vibrio cholerae* are sensitive to acids and prefer alkaline conditions. Most bacteria are killed in Strong acids or strong alkaline environment except the Mycobacteria.

(ii) Moisture content

One of the oldest methods of preserving foods is by drying or dessication. The preservation of food by drying is a direct consequence of removal or binding of moisture, without which microorganisms do not grow. It is generally accepted that the water requirement of microorganisms should be described in terms of water activity. Water activity (A_w) is a measurement of the amount of free water in a food medium. The amount of free water in food is important for growth of microorganisms. If there is lack of this free water, microorganism will not grow.

Water activity is defined as the vapour pressure of a food substance to that of water at the same temperature ($A_w = VP \text{ food} / VP \text{ water}$). This concept is related to relative humidity (RH) in the following way; $RH = 100 \times A_w$. Pure water has an A_w of 1.00, a 22% solution of NaCl solution (W/V) has an A_w of 0.86 and a saturated solution of NaCl, a A_w of 0.75. The A_w of most fresh foods is above 0.99. In general bacteria require high levels of A_w for growth than fungi, with gram negative bacteria having higher requirements than the gram –positive bacteria. Most spoilage bacteria do not grow below $A_w = 0.91$, whereas spoilage molds can grow at $A_w = 0.80$. With respect to food- poisoning bacteria, Staphylococcus aureus can grow at $A_w = 0.86$, whereas Clostridium botulinum does not grow at A_w below 0.94.

Table 4: Water activity of some food products

Food product	Water activity
Raw meat and Milk	0.99 -10.
Luncheon Meat	0.95
Boiled ham, sliced bacon	0.90
Dried grains	0.80

Table 5: Minimum water activity that supports growth of some microorganisms

Microorganism	Water activity
<i>Clostridium botulinum</i> , <i>Bacillus cereus</i> , <i>Pseudomonas aeruginosa</i> , <i>Salmonella spp</i> ,	0.95
<i>Staphylococcus aureus</i> , <i>Candida</i>	0.90
Most spoilage yeasts	0.88
Most spoilage molds	0.80

(iii) Oxidation – Reduction Potential

The O/R potential of a substrate may be defined generally as the ease with which the substrate loses or gains electrons. When an element or compound loses electrons, the substrate is oxidized, whereas a substrate that gains electrons becomes reduced. Therefore, a substrate that readily gives up electrons is a food reducing agent, and one that readily takes up electrons is a food oxidizing agent.

The O/R potential of a system is expressed by the symbol Eh. Aerobic microorganisms require positive Eh value (oxidized) for growth, while anaerobes require negative Eh values (reduced).

(iv) Nutrient content of the food

In order to grow and function normally microorganism of importance in the foods require the following;

- Water
- Proteins
- Carbohydrate
- Lipids
- Nitrogen
- Sulphur
- Phosphorous
- Vitamins
- Minerals e.g. Ca^{2+} , Fe^{2+}

Foods such as milk, meat, and eggs are rich in nutrients that are required by microorganism. These foods are therefore susceptible to microbial spoilage.

(V) Antimicrobial constituents (substances)

The resistance of some foods against attacks by microorganisms is due to presence of naturally occurring substances that possess and express antimicrobial activity. Some plant species are known to contain essential oils that possess antimicrobial activity e.g. Eugenol

in cloves, Allicin in garlic. Cow's milk contains several antimicrobial substances, including lactoferrin, conglutinin, and the lactoperoxidase system. Milk casein is known to be antimicrobial under certain conditions. Lysozyme in Eggs is known to exhibit some antimicrobial activity.

(vi) Biological structures

The natural covering of some foods provides excellent protection against the entry and subsequent damage by spoilage organisms. For example, meat has fascia, skin and other membranes that prevent entry. Eggs have shell and inner membranes that prevent yolk and egg white from infection.

(b). EXTRINSIC FACTORS

The extrinsic factors of foods are not substrate dependent. They are those properties of the storage environment that affect both the foods and their microorganisms. They are factors external to the food that affect microbial growth. They include:

- (i) Temperature of storage
- (ii) Presence and concentration of gases in the environment
- (ii) Relative humidity of food storage environment.

(1) Temperature of storage

Microorganisms, individually and as a group, grow over a very wide range of temperatures. The lowest temperature at which a microorganism has been reported to grow is -34°C while the highest was in excess of 100°C . Bacteria can therefore be grouped into three categories based on their temperature requirements for growth;-

(i) Psychrophilic microorganisms

These grow best at around 20°C , but also down to -10°C in unfrozen media. The psychrophilic bacteria can cause food spoilage at low temperatures. Several of the bacteria found in soil and water belong to this group.

(ii) Mesophilic microorganisms

These microorganisms grow best between $20^{\circ}\text{C} - 45^{\circ}\text{C}$. With optimal between $30^{\circ}\text{C} - 40^{\circ}\text{C}$. The optimum growth temperature is around 37°C . Some of the mesophiles such as *Pseudomonas aeruginosa* may grow at lower temperatures of between $5^{\circ}\text{C} - 45^{\circ}\text{C}$. None of the mesophiles are able to grow at temperatures of below 5°C or above 45°C . Most of the pathogenic bacteria belong to this category of microorganisms.

(iii) Thermophilic microorganisms

These grow at temperatures above 45°C , and often their optimum growth temperature is between $50^{\circ}\text{C} - 70^{\circ}\text{C}$. Growth of some bacteria may occur at 80°C . Bacteria in this group are mainly spore formers and are of importance in the food industry especially in processed foods.

2) **Concentration of gases in the environment**

Carbon dioxide (CO₂) is the single most important atmospheric gas that is used to control growth of microorganisms in food. CO₂ along with oxygen (O₂) are the two most important gases in modified atmosphere packaged (MAP) foods. Various microorganisms require for growth, either high oxygen (aerobic microorganisms), low oxygen tension (microaerobic microorganisms) or absence of oxygen (Anaerobic microorganisms). Some microorganisms (Facultative anaerobes) may grow either in high oxygen tension, or in the absence of oxygen.

Anaerobic or facultative aerobic spore formers are most likely to grow in canned foods. Microaerophilic bacteria are most likely to grow in vacuum packed foods, since they require low oxygen tension. Aerobic bacteria are likely to grow on the surface of raw meat. Aerobic molds are likely to grow in insufficiently dried or salted products.

3). **Relative humidity of food storage environment**

Relative humidity is defined as $100 \times a_w$. This is the amount of moisture in the atmosphere or food environment. Foods with low water activity placed in high humidity environment take up water. This increases their A_w , and hence spoilage of the food occurs easily. For example, dry grains stored in an environment with high humidity will take-up water and undergo spoilage by molds.

FOOD PRESERVATION

Food preservation is a process through which physical and/or chemical agents are used to prevent microbial spoilage of foods, thus allowing the food to be stored in a fit condition for future use. Food preservation aims at treating food in a manner to prolong its storage life. In food preservation, efforts are made to destroy organisms in the food or increase the period taken by the microorganisms to adapt to the food environment before they start to spoil the food.

For purpose of food preservation, foods are classified into three categories:-

(i) Perishable foods

These are the foods which deteriorate quickly after harvesting such as tomatoes, mangoes, papaya, peaches, plums and other juicy fruits. Also some juicy vegetables e.g. cucumber, snake gourd, bitter gourd etc are in this category. Meat, fish and poultry also fall in this category. These foods have a high level of moisture content and are highly susceptible to spoilage.

(ii) Semi – perishable foods

These foods have less moisture content e.g. beetroots, carrots, peas, green beans, pumpkins and apples. Eggs, pasteurized milk, highly smoked fish, and pickled vegetables belong to this group.

(iii) Non – perishable foods

These foods have very low moisture content e.g. mature food grain cereals, pulses and nuts. These foods are not easily susceptible to spoilage by microorganisms and enzymes.

FOOD PRESERVATION PRINCIPLES

Two general principles are employed in food preservation:

- (1) Inhibition principle
- (2) Killing principle

(1) Inhibition principle

In this principle, food preservation is achieved by inhibition of growth and multiplication of microorganisms. The inhibition principle may be achieved by any of the following:-

- (a) Reduction of water activity e.g. by drying or salting.
- (b) Reduction in pH e.g. by fermentation and addition of acids.
- (c) Use of preservatives e.g. sodium benzoate
- (d) Use of low temperature e.g. chilling or freezing.
- (e) Smoking – This has a drying and preservative effect.

Preservation of foods by inhibition methods does not necessarily imply destruction of organisms. On removal of the inhibiting influence, the food will undergo spoilage as the microorganisms present will grow and multiply to cause spoilage.

Food preservation by lowering pH

Many food products can be preserved by lowering pH so that the growth of spoilage and pathogenic bacteria is prevented. The lowering of pH can be achieved by addition of acids or fermentation. Fermentation is the breakdown of carbohydrates under anaerobic conditions into alcohol or lactic acid and carbon dioxide.

Food preservation by lowering water activity

Lowering of water activity may be achieved by:

- Addition of high content of salt eg sodium chloride, and sometimes nitrates and nitrites.
- Addition of high content of sugar.
- Drying:- sun/air drying, electrical drying or freeze drying.

The salting procedure

The salting procedure may be performed in four ways:-

- Dry cure in which the meat or fish is rubbed with salt.
- Pickling - The products are immersed in pickle or brine, usually containing about 15% salt.
- The injection cure – concentrated salt solution is injected in muscle tissues.
- Direct salt addition method.

Food Preservation by addition of high Content of Sugar

The high osmotic pressure of sugar creates conditions that are unfavourable for the growth of bacteria and reproduction of most species of bacteria, yeasts and molds. Approximately 70% of sucrose in solution will usually stop growth of all microorganisms in foods. Monosaccharides eg glucose (dextrose) and fructose are more effective in reducing the water activity, than disaccharides eg sucrose.

Thermophiles are more susceptible to the action of sugar than other bacteria. Osmophilic yeasts are able to tolerate very high concentrations of sugar and cause food spoilage.

Food preservation by use of low temperatures

Two methods are used in application of low temperatures to arrest microbial growth and multiplication; chilling and freezing. Chilling is keeping of food at temperatures between 0 – 15°C. The common chilling temperatures range between 4 - 5°C.

Freezing is keeping food at temperatures between -1°C - -35°C.

Effect of low temperatures

Low temperatures are used to retard chemical reactions and actions of food enzyme and to slow down or stop the growth and activity of microorganisms in the food. A low enough temperature will prevent growth of any microorganisms. Spores are not usually at all injured by freezing. However, most parasites are killed by freezing.

Food preservation by use of preservatives

A preservative is defined as any substance which is capable of inhibiting, retarding or arresting;-

- The growth of microorganisms,
- The deterioration of food due to activity of microorganisms
- The masking of the evidence of deterioration of any food

Chemical preservatives interfere with the cell membrane of microorganisms, their enzymes or their genetic mechanisms. Chemical preservatives are generally added after the foods are processed. Preservatives are divided into two types;

(i) Class one (1) preservatives – these includes :

- Common salt
- Sugar
- Dextrose
- Glucose (syrup)
- Wood smoke
- Spices
- Vinegar or acetic acid
- Honey

(ii) Class two (2) preservatives – There include

- Benzoic acid and Sodium benzoate
- Methyl and propyl p – hydroxy benzoates (parabens, sorbated, propionate)
- Sulphites (sulphur dioxide)
- Nitrites
- Hydrogen Peroxide
- Chlorine
- Carbon dioxide

Preservation of food by drying

This is one of the oldest methods of preserving foods. Large quantities of fruits are dried in different parts of the world. Dehydration extends the keeping qualities of the fruits well beyond their normal storage life, saves storage space and eases handling.

All the cereal grains are preserved by drying. Sun-drying remains the greatest food preservation action.

Milk is also preserved through drying in form of milk powder.

Meats are also preserved through drying.

(2) KILLING PRINCIPLE

In this principle, spoilage microorganisms in the food are destroyed (killed), and the food protected against subsequent contamination by being enclosed in an airtight container.

Methods employed to achieve the killing principle

- (1). Heat treatment through pasteurization or sterilization.
- (2). Irradiation with either ionizing or electromagnetic radiation e.g. Gamma rays, cobalt 60 radioactive particles. Radiation kills microorganisms by destruction of DNA and by creating toxic reactive compounds in a medium and in the microbial cells.
- (3) Use of gases - by use of ethylene oxide or ozone. The gases destroy both vegetative cells and spores.

PASTEURIZATION

This is the process of heat treatment, mainly of milk at specific temperatures and times. Pasteurization is aimed at destroying all pathogenic microorganisms without affecting the nutritive value of the food. Pasteurization is commonly used in milk processing.

There are three methods of pasteurization;-

- (a) Low temperature long time (LTLT) 63°C for 30mins.
- (b) High temperature short time (HTST) 72°C for 15 secs.
- (c) Flash Method 80°C for 1-2 secs.

STERILIZATION

This is the use of physical or chemical means to destroy all microorganisms including spores that are present in the food.

Sterilization may be achieved by;-

- (a) Heating at high temperature e.g. 100°C – 140°C. e.g. UHT milk – shelf life
Approx. 6 months
- (b) Irradiation - This kills bacteria, spores, and insects and also inactivates enzymes.

In practice, a combination of inhibition and killing principles and the various methods are used in food preservation depending on the food type.eg.

- Use of pasteurization and chilling of milk.
- Lowering of water activity and low temperature storage e.g. in fruits juices
- Use of preservatives and low temperature storage. e.g. in fruit juices.

Important Terminologies used in Heat Preservation

- D – Value
- Z – Value
- F – Value

(i) **Decimal reduction time (D – Value)** – This is time required at any temperature to destroy 90% of the spores or vegetative cells of a given organism. The higher the temperature, the faster is the rate of destruction and the shorter it takes to kill 90% of the cells. For example, D – Value of *Clostridium sporogenes* in a given food at 120°C is 1 minute, at 115°C is 4 minutes and at 110°C is 10 minutes.

The larger the initial number of vegetative cells or spores, the longer it will take to destroy 90% of the cells at a given temperature. D- Value is numerically equal to the number of minutes required for the survivor curve to traverse one log – cycle. If the initial number is one million cells/ml, one log cycle will reduce this number to 100 cells/ml.

(ii) **Z - Value**

This is the number of degrees the temperature has to be increased in order to reduce thermal death time ten – fold. The Z-Value is relatively constant and depends very little upon the environment. For spores of bacteria, the Z – Value used is 10°C. The spore killing effect of a heat treatment can be expressed as a function of temperature and the time the material has been exposed to that heat.

For example, when it takes 1 minute to kill 90% of the remaining spores, at 120°C, it will take 10 minutes to obtain the same effect (i.e. kill 90% of the spores) at 110°C.

(iii) **F – Value**

The value expresses the time taken to expose food to the same amount of heat required to destroy spores and vegetative cells of a particular organism using different temperatures.

- For example, food heated at 121.1°C for 2 minutes will give a value of F=2. To get the same F – Value at 111.1°C, one needs to heat the food for 20 minutes.

Heating such a food at 111.1°C for 2 minutes will give an F-Value of $2/10 = 0.2$. This means that one can obtain the same killing effect of spores and / or vegetative cells at a lower temperature, provided the time of exposure is longer.

Thus, F- Value shows the heat treatment given to a food product to destroy bacteria. As far as spore killing is concerned F – 1 is equal to 1 minute at 121°C (or 10 minutes at 111.1°C or 100 minutes at 101°C.)

References:

- Modern food microbiology. 7th Edition – James M.Jay et al.
- Technology of food preservation and processing
Written by EIRI Board of consultants & Engineers

FOOD – BORNE DISEASES (Food – borne infections and intoxications)

- Food – borne diseases are syndromes that are acquired as a result of eating foods that contain sufficient quantities of poisonous substance (Toxicants) or Pathogens.

Classifications of food-borne diseases

Food – borne diseases are often classified as:

1. Food-borne infections
2. Food – borne intoxications

1. FOOD – BORNE INFECTIONS

An infection is caused by the entrance of a pathogenic microorganism into the body, colonization, and the reaction of the tissues to the presence of the organism, their development or multiplication, or to the toxins they elaborate within the body.

Two types of food – borne infections are known:-

(i) One type results when the intestinal mucosa is penetrated and the infecting organism multiplies there – in (e.g. as occurs in diseases caused by *Salmonella*, *Shigella* and some strains of *Enteropathogenic Escherichia coli*) or passes to other tissues, where it multiplies or lodges (as occurs in diseases caused by Hepatitis A Virus, *Brucella*, *Trichinella spiralis*).

(ii) The 2nd type results when enterotoxins are released as the infecting organism multiplies, sporulates or lyses in the intestinal tract (as occurs in diseases caused by *Vibrio cholerae*, *Clostridium perfringens*, and some strains of *Enterotoxigenic E. coli*).

Food – Borne infections tend to have low incubation periods and are usually characterized by fever. Bacterial food infections include:- Cholera, Salmonellosis, typhoid fever, shigellosis, yersiniosis, *E.coli* infections, campylobacteriosis, *Vibrio*, parahaemolyticus, and listeriosis.

Mycotic food infections include:-

Candida spps, *Sporothrix* spp, *Wangiella* spp etc. Viral food – borne infections include:- Hepatitis A, Norwalk virus and Poliomyelitis virus.

SALMONELLOSIS

The *Salmonellae* bacteria constitute a group of bacteria with over 2000 different serotypes. These bacteria are capable of causing disease in animals and man when taken into the body in sufficient numbers. Many *Salmonella* species have a wide host range.

These are the organisms which commonly cause food poisoning.

However some salmonella species are restricted to a single host species eg; *Salmonella abortus ovis* causing abortion in ewes, and *Salmonella gallinarum*, the cause of fowl typhoid.

Some salmonella serotypes are associated with human disease and are not known to affect animals e.g. *S. typhi* and *S. paratyphi*. Salmonella are ubiquitous in the gut of human and animal and these act as sources of contamination to food.

People who are carries of the Salmonellae contaminate the food. A heavy dose of up to 10,000 – 1 million organisms per gram of food is required to cause infection. Salmonellae grow well in food and can exist for a considerable period in faeces, and on pastures.

Common food poisoning serotypes

Some of the Salmonella species involved in food poisoning include: Salmonella typhimurium, Salmonella enteritidis, Salmonella dublin, Salmonella softenburg, Salmonella virchow, Salmonella Montevideo, Salmonella infantis and Salmonella newport.

These species of salmonella are also involved in causing diarrhea in animals – i.e. they are **ZOONOTIC**.

Heat resistance

The Salmonellae are killed by temperatures attained in commercial pasteurization. They can remain alive in moist earth for one year and in dry earth for 16 months. They are not destroyed in carcasses or offal maintained at chilling or freezing temperatures, or in the usual pickling solutions.

Salmonella food – poisoning outbreaks

Outbreaks of Salmonellosis occur in different forms:-

- (a) Sporadic cases involving only one or two persons in a house-hold.
- (b) Family out-breaks in which several members of the family are affected
- (c) Large out-breaks caused by a widely distributed food item.
- (d) Institutional out breaks which may be caused by a contaminated single food item

Factors associated with salmonella food poisoning outbreaks

These include:-

- Consumption of inadequately cooked or thawed meat or poultry.
- Cross contamination of food from infected food handlers.
- Presence of flies, cockroaches, rats that act as vectors of the disease in the food environment.
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Transmission

Salmonellae reach food in many different ways:-

- (a) Directly from slaughter animals to food.
- (b) From human excreta, and transfer to food through hands, utensils, equipment, flies etc.

Food poisoning is more likely to occur if the total number of bacteria present is high. A small number of bacteria may have no ill – effect.

Foods involved

Any food contaminated with Salmonellae may be involved. However animal derived foods are commonly involved and these include:-

- Meat and meat products
- Milk and milk products
- Eggs and egg products

Clinical symptoms

The ordinary symptoms include, abdominal pain, headache, diarrhea, fever, vomiting, prostration and malaise.

In severe cases, there is septicaemia, with leucopenia, endocarditis, pericarditis.

Severe cases are encountered in babies, young children, the sick and elderly persons. The mortality is usually low, but may be up to about 13%.

Control measures

- Efficient refrigeration and hygienic handling of food.
- Consumption of properly cooked meat.
- Complete thawing of frozen meats and adequate cooking.
- Heat processing of meat, milk, fish and poultry to destroy Salmonella organisms in food.

TYPHOID AND PARATYPHOID FEVERS (ENTERIC FEVERS)

Enteric fevers include; typhoid and paratyphoid fevers caused by Salmonella typhi and Salmonella paratyphi, A, B, and C respectively.

The serotypes are similar to other Salmonella bacteria, but unlike them, they are pathogens of man.

S. typhi possess capsular (Vi) antigen in addition to the usual O and H antigens found in other serotypes.

Disease symptoms

The incubation period is usually 2 weeks, but might vary between 3 and 28 days for typhoid fever and between 1 and 15 days for paratyphoid fever.

The enteric fevers are generally septicaemic infections with a frequent, if not constant bacteraemia during the first two weeks of the disease.

The abdominal symptoms are severe, while fever and illness may continue for 4 – 6 weeks.

Transmission

The typhoid and paratyphoid bacteria are essentially human pathogens and are acquired mostly from human sources, namely; patients and carriers.

The bacteria may be transmitted through contamination of water, milk or food by flies. Only a few organisms are needed to cause the disease.

Control measures

The following methods may be applied:-

- Hygienic control of food and water
- Detection and treatment of chronic carriers
- Vaccinate using TAB – Vaccine. The vaccine contains a mixed culture of *S. typhi* and *S. paratyphi*. The vaccine confers protection for 5 – 7 years.

CAMPYLOBACTERIOSIS

Campylobacter are a group of tiny, strictly micro-aerophilic curved or spiral gram negative rods.

Campylobacter jejuni and *Campylobacter Coli* cause food poisoning and are associated with acute enterocolitis in man.

Campylobacter jejuni occurs in large number in cattle faeces, and poultry as normal flora. *Campylobacter Coli* are commonly associated with human diarrhea and enteritis in pigs mostly in association with *Treponema hyodysenteriae*.

Disease in man

Campylobacter jejuni and *C. coli* cause illness characterized by diarrhea, abdominal pain, fever, nausea, vomiting, and abdominal clamps.

The jejunum, ileum and colon are primarily affected, resulting in acute inflammation and occasionally abscess formation.

The disease is self – limiting. Incubation period ranges between 2-11 days with an average of 3-5 days.

The disease is preceded by fever, followed by foul smelling and watery diarrhea, which runs for 3-4 days. The diarrhea may sometimes contain blood and mucus in faeces. Abdominal pain is associated with backache, and a high mortality. The condition is self-limiting but may last for up to 10 days.

Mode of infection

Campylobacter infection occurs through ingestion of campylobacter organisms in contaminated food stuff. Foods involved include; Meat from infected animals, unpasteurized milk and possibly cross-contamination from these sources to foods eaten uncooked or unrefrigerated. Among the meats, poultry constitutes the greatest potential source of infection to humans. Up to 1,000,000 bacterial organisms /g of feces is present in poultry gut and feces.

Carelessness in the kitchen e.g., cutting chickens with the same knife used to cut other foods without proper cleaning prior to use may enhance exposure. Pork is a major source of *Campylobacter coli*. Contamination of pork occurs during slaughter.

Preventive measures

The following preventive measures may be applied:-

- Thorough cooking of all foodstuffs derived from animal sources.
- Prevention of contamination after cooking.
- Proper refrigeration of foods
- Recognition, control and prevention of campylobacter infections in animals.
- Maintenance of high standards of hygiene.

ESCHERICHIA COLI FOOD – BORNE INFECTION

Escherichia coli are potential food poisoning pathogens which are widely distributed in low numbers in food environments.

E. coli Strains involved in food borne infection fall into the following groups:-

- (i) Entero pathogenic *E.coli* (EPEC)
- (ii) Enterotoxigenic *E.coli* (ETEC)
- (iii) Entero invasive *E.coli* (EIEC)
- (iv) Enterohemorrhagic *E.coli* (EHEC)

Each group is composed of unique O: H Serotypes. Each group posses virulence factors characteristic of that group. The serotypes are characterized by using O – somatic and H – flagella antigens

Entero – Pathogenic E. coli (EPEC)

These stains generally do not produce enterotoxins, although they can cause diarrhea. They possess adherence factor plasmids that enable adherence to the intestinal mucosa. After colonizing the intestinal mucosa, attachment effacement (A/E) lesions are produced, resulting in destruction of brush border microvilli. EPEC strains cause diarrhea in children under 1 year of age.

Enterotoxigenic E. coli (ETEC)

These strains attach to and colonize the small intestine by means of fimbrial colonization factor antigens (CFAs) unlike EPEC strains, which cause diarrhea primarily in the very young, ETEC strains cause diarrhea in both children and adults. ETEC strains are among the leading causes of travellers diarrhea. The ETEC disease syndromes are rarely accompanied by fever and the diarrhea is sudden.

Enteroinvasive E.coli (EIEC)

These strains generally do not produce enterotoxins as do ETECs, but they enter and multiply in epithelial cells and then spread to adjacent cells in a manner similar to the Shigellae.

EIEC strains cause a disease that is characterized by watery diarrhea in most patients. In addition, there is fever, nausea and abdominal clumps. Bloody diarrhea may occur on less than 10% of the patients. The presence of mucus and polynonphonuclear leucocytes in the stool is typical of these strains. Illness is usually self-limiting, lasting for 2-3 days.

A relatively high dose (10^8 cells) is necessary to produce the disease in volunteers. The median incubation period is 18 hrs (range 2-48 hour). Food borne spread is the most common mode of transmission. Person to person spread has also been reported. Dysentery is rarely reported, and the very young and very old are the most susceptible members of the population.

Entero-hemorrhagic E.coli (EHEC)

EHEC infection is caused by *Escherichia coli* serotype O157:H7. It causes hemorrhage colitis in humans that is characterized by diarrhea, abdominal pain, which may be severe and vomiting. Few patients develop fever. Illness lasts for 4-8 days, although it may extend to 13 days for severe cases. Patients with complications have bloody diarrhea,

acute ulceration or ischemic colitis and submucosal oedema with severe colonic inflammation.

The organism E. Coli O157:H7 is heat sensitive, but resistant to freezing. It grows poorly at 44°C-45°C, with no growth at 45°C, suggesting that its presence may not be detected by fecal coliform assay test.

Death occurs in patients who develop hemolytic uremic syndrome (HUS). HUS can be recognized by acute renal failure. Micro-angiopathic hemolytic anaemia and thrombocytopenia.

Control Measures

The following control measures may be applied in the control of E. coli infections:-

- Proper cooking of hamburgers and other meats.
- Avoidance of cross-contamination of foods in the kitchen.
- Practicing of food personal hygiene eg. Proper washing of hands before handling foods.

SHIGELLOSIS (Bacillary dysentery)

Shigellosis is caused by members of the genus *Shigella* which belongs to the family *Enterobacteriaceae*, as do the *Salmonella* and *Escherichiae*. Only four species are recognized namely: *Sh. Dysenteriae*, *Sh. Flexneri*, *Shigella Boydii* and *Sh. Sonei*. *Sh. Dysenteriae* is a primary pathogenic that causes classic bacillary dysentery. The *shigella* have no known non human animal reservoirs. The *Shigella* are phylogenetically closer to the *Escherichiae* than to the *Salmonellae*.

All strains of *Shigella* possess potent exotoxins which are carbohydrate –lipid protein complexes.

The infective dose is smaller than that of *Salmonellae*, except *S.typhi*. (ingestion of mean of 344 cells per meal and 10.5- 12 cells per glass of water will cause infection.)

Clinical Symptoms

The illness begins 1-4 days after ingestion of bacteria and lasts 4-7 days. Symptoms of shigellosis include: water or bloody diarrhea, fever, stomach cramps, nausea or vomiting, dehydration and prostration in severe cases and convulsions in young children. The diarrhea which starts as a thin watery discharge quickly loses its fecal character to be composed of nothing but pus, mucus threads and blood. At this stage, there are agonizing pains and tenesmus .

Death from Bacillary dysentery is uncommon when efficient treatment is provided. However *Sh. Dysenteriae* infections have been reported to have a case fatality rate of 20% and the mortality rate is higher in children than in adults.

Transmission

Human cases and human carriers are the only important sources of infection. Spread of the diseases is by fecal-oral route, and person-person transmission is common. The bacteria leave the body in stool of an infected person or carrier and infect another person through contaminated hands, food, water or carrier and infects another person through contaminated hands, food, water or objects (toys, pens etc). Any type of food can

transmit *Shigella* pathogens to cause disease in man. Flies can spread *Shigella* germs when they get into contact with infected stool and then contaminate drinking water or food. *Shigella* organisms may remain viable in tap water for as long as 6 months, and in sea water for 2-5 months.

Preventive measures.

The following preventive measures may be applied:-

- Practice food hygiene and sanitation.
- Wash hands well with water and soap after visiting the ablution.
- The home and surrounding should be kept clean to prevent contamination of food and water supply.
- Practice proper disposal of human water or sewage.
- Keep kitchen work surfaces clean.
- Eat properly cooked food.
- Drink properly pasteurized milk and other liquid foods such as juices.
- Reconstitute juices with potable water.

CHOLERA

Cholera is caused by *Vibrio cholerae*. The bacteria are ingested in drink or food. In natural infection, the dosage is usually very small.

Infection with *V. cholerae* occurs by the ORAL route through the ingestion of water or food contaminated with the bacteria. The dosage is usually very small – ie only exposure to small numbers of *Vibrio bacteria* cause the disease.

The bacteria multiply in the small intestines and produce an enterotoxin that either stimulate the mucosal cells to secrete large quantities of isotonic fluid, or increase the permeability of the vascular endothelium. The *Vibrio cholerae* are very sensitive to acid and in volunteer feeding tests, it is usually possible to produce clinical cholera with prior neutralization of gastric acid.

Transmission

Cholera is not easily spread by direct contact and although some epidemics have been attributed to direct contact, there is no conclusive proof.

The most important mode of spread is through the environment, particularly by contaminated water from rivers, tanks or canals.

- Man is the only natural host of the *Vibrio cholerae*. Spread of infection is from person –person through contaminated water or foods. Shrimps and vegetables are frequent sources of infection.

Cholera is an infection of crowded poor communities and it tends to persist in such areas. Cholera outbreaks occur either as explosive epidemics usually in non-endemic areas or as protracted epidemic waves in endemic areas.

Clinical symptoms.

The incubation period of cholera is variable ranging from 1-5 days. In outbreaks with a common source of infection, the usual incubation period is about 48hrs (2 days).

Typical cholera has a sudden onset of effortless vomiting and painless profuse watery diarrhea, which quickly assumes the characteristic “rice - water” appearance. The frequent watery stool may be accompanied by small parts of the mucosa being liberated from the intestines.

Patients may produce upto 20-30 stools per day thus losing many litres of water and electrolytes.

Patients therefore exhibit extreme dehydration and weakness. Urination is suppressed, the skin becomes wrinkled, the eyeballs are sunken and the voice becomes weak and husky. The blood pressure falls, the heart sounds become barely audible and the pulse becomes rapid and weak just before death.

Death may occur in 12-24 hours from the onset of the disease due to rapid dehydration.

Diagnosis

A vibrio immobilization test with dark field microscopy may be applied. In the acute stage, the bacteria are abundant in watery stool (upto $10^7 - 10^9$ organisms per ml of stool). Bacteriological examination can be done on stool, suspect water and food. Specific identification of biotype and serotype is carried out serologically (eg CFT) and by phage sensitivity tests.

Control Measures

The following measures may be applied:-

- Provision of portable water
- Proper sewage disposal
- Proper cooking and hygienic handling of food.
- Observation of high standards of personal hygiene.
- Vaccination- the heat killed, phenol preserved vaccine confers protection which lasts for 3-6 months.

Vibrio parahaemolyticus food-borne infection

V. haemolyticus is a pathogenic bacterium, whose natural habitat is the sea. Human infections occur solely from sea foods eg. Oysters, shrimps, crabs, lobsters, clams and relaxed shellfish. Cross-contamination may lead to other foods becoming vehicles.

Symptoms of the disease.

V. parahaemolyticus causes gastro-enteritis and extra intestinal infections in man. The mean incubation period is 16.7 hrs (range 3-76 hrs).

Symptoms of the disease include:- Diarrhoea (95%) Cramps (92%), weakness (90%), Chills (55%), Headache (48%) and vomiting (12%).

Symptoms last from 1-8 days with a mean of 4.6 days.

Vibrio vulnificus food-borne infection

V. vulnificus causes a severe food-borne infection. The case fatality rate for the septicemic form exceeds 50%.

In immuno-compromised individuals, the Vibrio infection causes fever, nausea, myalgia and abdominal cramps, 24-48hrs after eating contaminated food.

The organism can cross the intestinal mucosa rapidly leading to sepsis within 36 hours of the initial onset of symptoms.

Cases are most commonly reported in warm-weather months and are often associated with eating raw oysters.

LISTERIA MONOCYTOGENES INFECTION

Listeria monocytogenes is a small coccoid, gram positive, none encapsulated, motile rod. It is pathogenic to both animals and human beings.

The organism is widespread in nature and is a transient constituent of the intestinal flora excreted by 1-10% of healthy humans. It is an extremely hardy organism and may survive for years in the cold in naturally infected sources.

Transmission

L. monocytogenes infection occurs after consumption of raw vegetables, salads, raw milk, soft cheese, meat and meat products, ice-cream, poultry, sea foods. Ready- to-eat foods are important in causing *Listeria* food poisoning.

Clinical signs

In man the most common manifestation of infection is meningitis in infants. Other manifestations include:- septicemia, abortion in pregnant women, localized external or internal abscesses, endocarditis , conjunctivitis, flu-like illness and pharyngitis.

Pregnant women, infants and elderly people are particularly at risk of infection with *L. monocytogenes*.

The infection is fatal in susceptible individuals with a mortality of 25-30%. Deaths have been reported in fetuses, neonates and other individuals with compromised health status.

YERSINIA ENTEROCOLITICA INFECTIONS

This organism has been isolated from beef, lamb, pork, sea foods, vegetables, milk and cakes, vacuum –packed meat. Of all the sources, swine appears to be a major source of strains pathogenic to man. Virulence appears to be as a result of tissue invasiveness capacity of this organism.

Symptoms

Symptoms develop several days following ingestion of contaminated food.

The symptom includes; abdominal pain, diarrhea, fever, vomiting, headache and pharyngitis. Children appear to be more susceptible than adults. The organisms may be shed in stool from upto 40 days following illness.

A variety of systemic involvement may occur as a consequence of the gastroenteritis syndrome. These involvements include: pseudo appendicitis, mesenteric lymphadenitis, peritonitis, terminal ileitis, reactive arthritis, colon and neck abscesses, cholecystitis, intesusception etc. The organism can be recovered from urine, blood, celebrosprinal fluid and eye discharge of infected individuals.

VIRAL FOODBORNE INFECTIONS

Viruses are common pathogens transmitted through food. Hepatitis A and Norwalk-like viruses (Novoviruses) are the most important viral foodborne pathogens. These viruses are highly infectious and may lead to widespread outbreaks.

Characteristics of viral foodborne infections include:-

- Only a few viral particles are required for the disease to develop.
- High numbers of viral particles are transmitted via faeces of infected persons (upto 10^{11} particles per gram of faeces).
- Specific living cells are necessary for virus replication. Accordingly viruses cannot multiply in foods or water.
- Foodborne viruses are relatively stable and acid resistant outside the host cells.

INFECTIOUS VIRAL HEPATITIS A INFECTION

The incubation period is long, being on average 30 days (range 15-50 days). It is a systemic infection characterized by gastrointestinal manifestations and liver injury, fever, malaise, anorexia, nausea, abdominal discomfort, bile in urine and jaundice. The duration of the disease could be from a few weeks to several months.

NORWALK-LIKE VIRUS (NOVOVIRUS) FOODBORNE INFECTION

Novovirus infection is relatively mild with an incubation period of 3 days. Clinical manifestation/ symptoms include vomiting and diarrhea, and rarely convulsions have been reported.

Asymptomatic infections are common and may contribute to the spread of the infections. Infections have resulted from eating of raw oysters.

RICKETTSIAL FOODBORNE INFECTIONS

Q-Fever

Q –fever is caused by *Coxiella burnetti* and has an incubation period of 2-4 weeks.

Transmission

Q- fever is mostly an occupational disease among people who handle livestock and raw animal products (eg farm and slaughterhouse workers etc). Infection occurs through consumption of raw milk, contaminated butter and cheese. Infection may also occur through contact with infected placentae, contaminated straw beddings and animal carcasses or slaughterhouse offals.

Clinical signs

Symptoms include a sudden onset of fever, dry cough and chest pain due to pneumonitis. Hepatic disorders which include slight jaundice occur in severe cases. Mortality is low with complete recovery.

Diagnosis

The following methods may be applied:-

- Serology including CFT, micro-agglutination test and FAT.
- Isolation of agent (in well equipped laboratories due to the high risk of infection).

Control

The following control methods are applicable

- Pasteurization of milk and milk products. Heating at 63°C/30 min or 72°C for 15 seconds.
- Safe disposal of offal.

FOODBORNE INTOXICATIONS

These are diseases caused by food containing the following:

1. Biotoxins which are found in tissues of certain plants and animals.
2. Metabolic products (Toxins) formed and excreted by microorganisms (such as bacteria, fungi and algae), while they multiply in food, or in gastro-intestinal tract of man.
3. Poisonous substances, which may be intentionally or unintentionally added to food during production, processing, transportation or storage.

Food borne intoxications have short incubation periods (minutes to hours) and are characterized by lack of fever.

Foodborne intoxications can be classified into:

- (a) bacterial intoxications
- (b) Fungal intoxications
- (c) Chemical intoxications
- (d) Plant toxicants
- (e) Poisonous animals

BACTERIAL FOODBORNE INTOXICATIONS

1. *Staphylococcus aureus* intoxication
2. *Bacillus cereus* food borne intoxication
3. *Clostridium perfringens* food borne intoxication
4. *Clostridium botulinum* food borne intoxication

1. Staphylococcus aureus food borne intoxication

This is a type of food borne intoxication caused by consumption of food contaminated with staphylococcal enterotoxins produced by certain strains of *Staphylococcus aureus* while growing in food.

Staphylococcus aureus produces five serologically different Enterotoxins that are involved in food borne intoxication. These are:-

1. Staphylococcal enterotoxin A (SEA)
2. Staphylococcal Enterotoxin B (SEB)
3. Staphylococcal Enterotoxin C (SEC)

4. Staphylococcal Enterotoxin D (SED)
5. Staphylococcal Enterotoxin E (SEE)

Individual strains of Staph. Aureus may produce one or more enterotoxin types while growing in food.

Growth conditions

Staph. aureus is a facultative anaerobe, non-spore forming gram positive cocci. It grows at a temperature range of 12-44°C (optimum 37°C) and pH range of 4.0 – 9.83 (Optimum 7.4-7.6).

Growth occurs in an environment containing upto 18% sodium chloride and water activity of 0.85-0.88 when growing aerobically and 0.9 under anaerobic conditions.

Toxin Production

Toxin production occurs at growth temperature of 12-44°C, pH 4.2 and salt concentration of $\leq 10\%$.

No toxin production occurs at temperature below 12°C, pH < 4.2 and > 10% salt concentration.

Nature of enterotoxin

All the Staph. aureus enterotoxins are heat stable (withstand heating at 100°C for one hour) and ordinary cooking procedures, pasteurization and drying do not inactivate these enterotoxins.

They are pH stable and resistant to most proteolytic enzymes (eg trypsin, chymotrypsin, renin and pepsin). The enterotoxins are also not affected by irradiation.

All the five enterotoxins have the same potency.

Competition with other organisms

Staph. aureus is a poor competitor and therefore grows poorly or not at all when growing with other microorganisms. Majority of Staph. aureus food poisonings are due to foods in which the microbial flora is substantially reduced, such as cooked, canned or pasteurized food.

Vehicle foods

The following foods are involved in Staph. aureus poisoning:-

Milk and milk products, including pasteurized milk, yoghurt, chocolate milk, fermented milk, cream filled pastries, poultry, fish, shellfish, meat and meat products, non-meat salads, egg and egg products, vegetables and cereal products.

Reservoirs

Staphylococci are found in varying numbers in air, dust, water, food, faeces and sewage.

The primary habitat of Staph. aureus is the mucous membranes of the nasopharynx and skin of man and animals. The organism is found in the nose, skin, saliva, intestinal contents and in faeces. Human carriers of Staph. aureus are numerous and undoubtedly the source of a number of outbreaks.

Contamination of foods may be traced to food handlers with minor septic had infections or severe nasal infections. The nasal mucous membrane is another particularly important source of Staphylococci of human origin.

Disease symptoms in man

The incubation period is 1-6 hours after consumption of food contaminated with at least 1.0 µg of enterotoxin. The clinical signs include; salivation, nausea, vomiting, abdominal cramps, sometimes diarrhea with prostration. The poisoning has an attack rate of 5-100%, but fatalities which occur in children, the old and debilitated victims are rare. Duration of the illness is 24-72 hours. A dose of 10ug or more is needed in order to cause the disease.

Diagnosis

The following may be applied:-

- Use of clinical symptoms – incubation 1-6 hours
- Enumeration of organisms in food and stool (presence of ≥ 10.6 cfu/g) of *Staph.aureus* in food is indicative of the bacterial involvement in causation of the disease.
- Detection of the enterotoxin in suspect food, stool and in vomiting of victims using various methods which include:-
 - (a) Serological tests (eg ELISA, Reverse passive agglutination test)
 - (b) Biological tests (eg. Monkey feeding tests)
- Use of molecular biology techniques – Gene probing and polymerase chain reaction.

Preventive measures

The following measures may be applied:

- Practice good personal hygiene including good personal conduct in food establishments and when handling food.
- Use of spoons when serving foods to prevent contamination of cooked foods
- Fast cooling of cooked foods and keeping such foods at low temperatures.
- Discourage use of left-overs.

BACCILLUS CEREBUS FOOD BORNE INTOXICATION/Bacillus Cereus gastro-enteritis

B. cereus is an aerobic, spore forming rod normally present in soil, dust, vegetation and water. It is also capable of growing under anaerobic conditions. The bacteria are also found in meat and milk. Vegetative cells are generally 1.0-1.2 µm by 3.0 -5.0 µm, and the spores are ellipsoidal, central on paracentral. The bacteria grow in the temperature range of 10°C - 48°C with optimum growth occurring between 10°C – 35°C. Recent evidence indicates that spores of *B. cereus* serotype 1 are more resistant to heat than the spores of other serotypes.

In view of the wide distribution of *B. cereus* in nature and the environment and in the various foods, the organisms are inevitably ingested from time to time in small numbers and contribute to the transitory intestinal flora. The organism can be found in 14% of single fecal specimens of healthy individuals in a population.

The gastroenteritis is caused by consumption of Enterotoxins produced by certain strains of B. cereus.

B. cereus produces a wide variety of extracellular toxins and enzymes. Some of the Enterotoxins produced include:

(a) Hemolysin BL (HBL). This is a tripartite complex composed of B₁L₁ and L₂. Together, this complex exhibits hemolysis, cytolysis, dermonecrosis, vascular permeability and enterotoxic activity. Although no single enterotoxin has been demonstrated, it appears hemolysin BL (HBL) causes the diarrheagenic syndrome (diarrheal syndrome)

(b). Cereolysin (mouse lethal toxin, hemolysin 1). This enterotoxin has a molecular weight of 55KDa – This appears to have no role in the causation of gastroenteritis.

(c). The emetic (vomiting) type enterotoxin is an ionophoric, water insoluble peptide. It has a molecular weight of about 1.2 KDa. This toxin causes the Emetic Syndrome in man.

(d). The emetic toxin strains of B. cereus grow in the temperature range of 15-50°C., with an optimum of 35-40°C.

The enzymes produced by B. cereus include; lecithinase, proteases, B-lactamase, sphingomyelinase, etc.

Foods involved in B. cereus food intoxication;

Bacteria are frequently isolated from meat, eggs and dairy products; minced meat. Cereal dishes for example mashed potatoes, rice, vegetables, cream and milk pudding are also involved.

The disease in man

B. cereus is commonly considered to be a harmless saprophyte. However there are numerous reports implicating the bacteria in various human infections. These include: Bronchopneumonia, bacteremia and septicemia, meningitis and osteomyelitis.

Two main syndromes have been associated with the B. cereus infection in man.

1. Diarrheal syndrome

This syndrome is rather mild with symptoms developing within 8-16 hours after consumption of food, more commonly within 12-13 hours, and lasting for 6-12 hours.

Symptoms consist of nausea, with vomiting being rare, cramp-like abdominal pains, tenesmus and watery stool. Fever is generally absent.

(2) Emetic Syndrome

This form of B. cereus food poisoning is more severe and acute than the diarrheal syndrome. The incubation period ranges from 1-6 hours, with 2-5 hours being most common. It is often associated with fried or boiled rice dishes.

The symptoms include; nausea, vomiting, abdominal cramps and sometimes diarrhea may occur.

Diagnosis

The following may be applied:-

- Suspect infection from clinical signs and history.
- Use of selective media – Enumeration of bacteria of same *B. cereus*; strain and serotype in significant numbers ≥ 15 cfu/g in incriminated foods, in faeces or vomitus of affected persons.
- .Detection of *B. cereus* Enterotoxins in foods through immunological and biological tests.

Prevention

As it is impossible to reduce the prevalence of *B. cereus* in nature, it must be accepted that sometimes this organism will be present in food. The organism is not dangerous when ingested in small numbers. Control must aim at destruction of the bacteria and prevention of germination of the spores and subsequent multiplication of vegetative cells in cooked foods. Freshly cooked food eaten hot immediately after cooking is safe.

Prevention of poisoning due to *B. cereus* is best achieved by confining the organism to small numbers by preparing the food, such as rice and meat, a short time before service, or if it has to be stored, by rapid and adequate cooling to a temperature sufficiently low to prevent growth of the bacteria. Good hygiene should be observed by food handlers and in food establishments.

CLOSTRIDIUM PERFRINGENS FOOD POISONING (INTOXICATION)

Actiology: *Cl. perfringens* is a gram positive, anaerobic, spore forming rod, widely distributed in nature. Based on the ability of the bacteria to produce exotoxins, five types are recognized. A, B, C, D, and E.

The food poisoning strains of *Cl. perfringens* belong to the type A. some type C strains produce enterotoxin and may cause a food poisoning syndrome.

The food-poisoning strains of *Cl. perfringens* are widely distributed in soil, water, foods, dust, spices and the intestinal tract of humans and animals. Because *Cl. perfringens* is a spore former, it can withstand adverse environmental conditions of drying, heating and certain toxic compounds.

Cl. perfringens is mesophilic, with a growth temperature of 37°C -45°C. The lowest temperature for growth is around 20°C and the highest is around 50°C.

The causation factor of *Cl. perfringens* food poisoning is an enterotoxin. The enterotoxin has a molecular weight of 35,000 daltons and an isoelectric point of 4.3. It is heat sensitive (biological activity destroyed at 60°C for 10 minutes), and is pronase sensitive but resistant to trypsin.

The food poisoning strains of *Cl. perfringens* are heat resistant and survive heating at 100°C for 1 hour.

Vehicle foods

The foods involved in *Cl. perfringes* outbreaks are often meat dishes prepared one day earlier and eaten the next day. The heat preparation of such foods is presumably inadequate to destroy the heat resistant endospores. Upon cooling and warming, the endospores germinate and multiply.

Foods that have been involved include red meats, chicken, fish, pork, fruits, vegetables, spices etc.

Food poisoning occurs when the number of cells reaches 10^7 - 10^8 cells/g of food. Growth is enhanced by anaerobic conditions achieved after removal of oxygen during the cooking process.

Mode of transmission

Cl. perfringens is transmitted to man through:

- Directly from slaughter animal
- Contamination of meat from containers, handlers, dust and water.
- Cross-contamination in the kitchen environment.

Symptoms of the disease in man

The symptoms appear between 6-24 hours especially 8-12 hours after ingestion of contaminated food. The symptoms are characterized by acute abdominal pain and diarrheal nausea, fever and vomiting is rare. Except in the elderly or in debilitated persons, the illness is of short duration of a day or less. The fatality rate is low.

Diagnosis

The following may be applied:-

- Suspect disease from clinical signs and History. Typical symptoms of (abdominal pain and profuse diarrhea), 12-24 hours following consumption of food.
- confirmation of disease through :-
 - Enumeration of *Cl. Perfringens* in food and stool (count of $\geq 10^5$ cfu/g of).
 - Detection of Enterotoxin in food and patients stool using serological methods eg ELISA.

Prevention:

Because *Cl. perfringens* poisoning often occurs in institutional cafeteria, some special precautions should be taken. These include:-

- Proper cooking of food and eating freshly prepared foods.
- Thorough washing and sanitation of containers.
- Hygiene handling of cooked food.
- First cooling of cooked food. Storing of food in small quantities will enhance cooling.
- Proper re-heating of cold cooked food before consumption.
- Storage of left-overs of unused foods in freezers.

Clostridium botulinum food poisoning (BOTULISM)

Actiology

Symptoms of Botulism are caused by the ingestion of a highly toxic, soluble Exotoxin produced by *Cl. botulinum* while growing in foods. *Cl. botulinum* is a gram positive, anaerobic spore-forming rod with oval to cylindrical, terminal to sub-terminal spores. On the basis of serological specificity of their toxins, seven types of *Cl. botulinum* are recognized: A, B, C, D, E, F and G. Type A, B, E, F and G cause disease in humans while type C causes botulism in fowls, cattle, ninks and other animals, and type D is associated with forage poisoning of cattle especially in South Africa. The type of enterotoxin (exotoxin) is also differentiated on the basis of their proteolytic activity. Type A and B are proteolytic as are some types of B and F strains. Type E is non- proteolytic, as are some B and F strains.

Distribution of *Cl. botulinum*

The organism is indigenous to soils and water. Type E is truely an aquatic organism and multiplies in dead aquatic animals and sediments and is disseminated by water currents and fish. The non-proteolytic *Cl. botulinum* are associated more with water than soil.

Growth characteristics

The proteolytic *Cl. botulinum* generally do not grow below 12.5°C, although a few reports exist in which growth was observed at 10°C. The upper temperature for these bacteria is 50°C. Therefore the proteolytics grow at a temperature range of 10°C to 50°C. The non-proteolytics grow at a temperature range of 3.5-45°C (with the optimum temperature being 35-37°C).

It is generally recognized that growth of *Cl. botulinum* does not occur at below pH 4.5. the minimum water activity (aw) which allows growth of *Cl. Botulinum* is 0.94. With respect to heat resistance, the proteolytic strains are much more resistant to heat than the non-proteolytics.

Proteolytic strains of *Cl. botulinum* produce an active botulinal toxin, while the non-proteolytics produce inactive pro-toxin which requires activation by trypsin.

Nature of Botulinal toxins

The toxins of *Cl. botulinum* are neurotoxins which are formed within the organism and are released upon autolysis. The botulinal neurotoxins (BONT) are the most toxic substances known, with the purified type A reported to have a minimum lethal dose in mice of 0.4-2.5 ng/kg by intravenous or intraperitoneal injection, and a 50% human lethal dose of about 1ng/kg of body weight.

Type A toxin has been reported to be more lethal than B or E.

Symptoms of botulism can be produced by either parenteral or anal administration of the toxin. The toxins may be absorbed into the blood stream through the respiratory mucous membranes as well as through the walls of the stomach and intestines.

The toxins are heat labile and are inactivated by heating at 80°C for 10mins. They are unstable at alkaline pH (but stable at pH below 7.0)

The toxins are not completely inactivated by the proteolytic enzymes of the stomach.

Vehicle foods

Types of food implicated in causation of Botulism include:-

- Home made (ie canned) vegetables.
- Canned fruits
- Canned meats, fish
- Poultry
- Home made fermented fresh foods have been incriminated, together with smoked, pickled and canned foods.
- Uncooked fresh foods are safe because they are eaten before the toxin has had time to develop; while, if the foods are cooked, the toxin is destroyed.

Nitrates/nitrites are used in canned meat as preservatives. These salts reduce the chances of growth of *Cl. botulinum* and inhibit toxin production.

Mode of transmission

The greatest hazards of botulism come from home-prepared and home-canned foods that are improperly handled or given inadequate heat treatment to destroy botulinal spores. The spores are present in animal tissues eg. meat and fish.

Symptoms of disease in man

The symptoms may develop 12-72 hours after the ingestion of foods containing the toxin. The symptoms consist of nausea, vomiting, fatigue, dizziness and headache, dryness of the skin, mouth and throat, constipation, lack of fever, paralysis of muscles, double vision, and finally respiratory failure and death. The mortality rate varies between 30% - 65% although upto 100% rate has been reported.

Preventive measures

The following methods may be used:

- Ensuring proper sterilization and preservation of canned meat, and proper heating of food before consumption to destroy heat labile neurotoxins. Food should be heated to a temperature of $\geq 80^{\circ}\text{C}$ and this temperature maintained for at least 10min.
- Pickled foods should be treated with brine containing not less than 10% common salt.
- Care should be taken in the manufacture of cans, their transport, handling, storage and their use during the packaging of the products.

FUNGAL INTOXICATIONS

These are caused by consumption of metabolites produced by fungi when they grow in foods. A large number of fungi (molds) produce toxic substances designated MYCOTOXINS. Some MYCOTOXINS are mutagenic and carcinogenic, some display specific organ-toxicity. At least 14 mycotoxins are known to be carcinogenic, with the aflatoxin being the most potent. Mycotoxins are produced as secondary metabolites.

Vehicle foods

Grains, oilseeds, fruits and vegetables are mostly involved if they are stored at high humidity (ie A_w of ≥ 0.75) or if they are not properly dried before storage. Poor storage of dry foods eg grains leads to growth of molds and subsequent production of mycotoxins. The most important mycotoxin in public health is the Aflatoxin.

AFLATOXINS

The consumption of aflatoxins causes aflatoxicosis. The aflatoxin is produced by *Aspergillus flavus*. Four main types of aflatoxins have been described:-

- (i) Aflatoxin B₁
- (ii) Aflatoxin B₂
- (iii) Aflatoxin G₁
- (iv) Aflatoxin G₂

Animals consuming feeds contaminated by aflatoxin B₁ secrete in their milk aflatoxin M₁ and M₂

Effects of Aflatoxins in man

While consumed in large doses, aflatoxins are lethal by causing acute hemorrhage syndrome. Sub-lethal doses cause histotoxic changes. Often consumption of small doses causes liver tumors as Aflatoxins are potent carcinogens.

Prevention of Aflatoxicosis

The following actions may be applied in prevention of aflatoxicosis

- Proper drying and storage of grains and other susceptible foods.
- Quality control of potentially hazardous foods to ensure that they do not contain above the allowable limits of 20ppb of aflatoxin before consumption. Appropriate analytical tests should be applied.
- Use of fungicides as seed dressing to protect stored cereals and other foods eg potatoes against fungal invasion.

CHEMICAL FOOD POISONINGS (Intoxications)

This is the type of food poisoning arising from ingestion of food containing poisonous chemicals.

These chemicals may be intentionally or unintentionally added to foods as a result of producing, processing, transporting or storage.

Chemical substances involved

Chemical food poisoning involves the following chemicals:

- Heavy metals eg Antimony, Mercury, Arsenic, Flouride, Lead, Cadminium, Cyanide etc.
- Pesticides and insecticides – eg DDT, BHC, Organochlorides and organophosphates.

- Herbicides
- Fungicides eg. Organomercurials
- Preservatives eg. Nitrites, nicotinate
- Antibiotics eg Penicillin, tetracyclines
- Radionuclides eg. Cesium, Strontium, Radium, Iodine isotopes etc

Chemicals may enter foods in the following manner:

- Accidental contamination eg by Heavy metals, pesticides, radionuclides.
- Intentional addition eg preservatives such as nitrites and nitrates.
- Leaching from containers eg Zinc-galvanized containers by acid foods, copper surfaces, lead pipes, asbestos roofs etc.
- Usage: presence of such chemicals in food as a result of their use in animals and crop husbandry.
- Malicious addition to cause harm. (this is rare).

Clinical signs in man

Chemical food intoxications exhibit a very short incubation period, usually a few minutes to a few hours.

The symptoms observed are usually dependant on the type of toxicant, but include: Nausea, headache, convulsions, gastrointestinal irritation, abdominal cramps, vomiting and diarrhea.

Prevention Measures

The following measures may be applied:-

- Do not use utensils or containers that are able to leach chemicals.
- Use coloured pesticides and apply proper storage of the pesticide.
- Prevent use or avoid use of dangerous additives eg Sodium nicotinate.
- Prevent contamination of foods by insecticides.
- Ensure observance of required withdrawal periods after use of pesticides and antibiotics in animal and crop husbandry.

BIOTOXICATIONS

These are disorders resulting from ingestion of a poisonous substance (a biotoxin) present in the body of a plant or animal. Biotoxins are derived from plants or animals as a result of metabolic activities. Only a small proportion of fish and shellfish consumed by man contain biotoxins.

Animal tissues may be rendered poisonous by bacterial and enzymatic decomposition, but some are naturally toxic.

Primary toxicity arises due to inherent toxicants that arise due to normal metabolic processes.

Secondary toxicity arises due to external toxicants contaminating animal tissues. Some of these toxicants are eg. Pesticides, heavy metals and drugs eg antibiotics.

There are various types of toxic fish eg Puffers, triggerfish and parrot fish. The fish toxin affects the peripheral nervous system. The fish may become poisonous by feeding on poisonous marine organisms. Some of the biotoxins associated with fish include: Ciguatera poisoning (ciguatera toxicity), tetraodon poisoning and scombroid toxicity.

Prevention of animal biotoxinations.

The following measures may be applied:-

- Avoid eating of unknown meats from vertebrates or invertebrates sources.
- Adhere to local eating customs and local quarantine regulations.
- Avoid sea foods and always heat foods to above 100°C to denature the inherent heat labile toxins that may be present in animal tissues.

FOOD BORNE ANIMAL PARASITES

The animal parasites that may be contracted by eating certain foods belong to three distinct groups:

- (i) Protozoa
- (ii) Flatworms
- (iii) Roundworms

The Protozoan diseases include:

- (i) **Giardiasis** – caused by *Giardia lamblia*, which is a flagellate Protozoan. These protozoa occur in water, the environment and foods. Most of the infections are asymptomatic with passage of cysts in the stool.
- (ii) **Amebiosis** (Amoebic dysentery). This is caused by *Entamoeba histolytica*. It is often transmitted by fecal oral route, although transmission is known to occur by water, food handlers, and foods.

Clinical Signs

Incubation period for amebiosis is 2-4 weeks. Its onset is insidious, with loose stooling and generally no fever. Mucus and blood are characteristic.

(iii) Toxoplasmosis

This is caused by *Toxoplasma gondii* which is a coccidian protozoan that is an obligate intracellular parasite.

(iv) Sarcocystosis

Of the more than 13 known species of the genus *Sarcocystis*, two are known to cause an extra intestinal disease in humans. One of these is obtained from cattle (*S. Hominis*) and the other from pigs (*S. suihominis*). Humans are the definitive hosts for both species.

(v) Cryptosporidiosis

Protozoan *Cryptosporidium parvum* is known to be a pathogen of at least 40 mammals and varying numbers of birds and reptiles.

It is estimated to cause diarrhea in 7-30% of HIV AIDS patients. In humans the disease is self-limiting in immunocompetent individuals, but it is a serious infection to the immune-compromised, such as AIDS patients. The fecal-oral route of transmission is the most important in water, milk and food.

C. parvum is an obligate intracellular coccidian parasite that carries out its life cycle in one host.

(vi) Cyclosporiasis

The protozoa that causes this disease, *Cyclospora cayentanensis* is a coccidian parasite. The protozoa is an intestinal pathogen that appears to paratize the epithelial cells (enterocytes) of the jejunum

(2) FLATWORMS:

All flatworms belong to the animal phylum *Platyhelminthes*

(i) Fasciolosis

This syndrome is also referred to as parasitic biliary cirrhosis and liver rot, and is caused by *Fasciola hepatica*. The disease among humans is cosmopolitan in distribution, and the organism exists where sheep and cattle are raised. They and the humans are the principal definitive hosts.

(ii) Paragonimiasis

This is caused by *Paragonimus spp*, especially *P.westermani*. The eggs of this parasite are expelled in sputum of definitive hosts (Humans and other animals). Paragonimiasis is accompanied by severe chronic coughing and sharp chest pains. Sputum is often reddish-brown or bloody.

(iii) Clonorchiasis

Caused by *Clonorchis Sinensis* which is the Chinese liver fluke, which causes oriental biliary cirrhosis. Symptoms of infection may not occur if the infection is mild, but in severe cases damage to the liver may occur. This may lead to liver cirrhosis and edema, and cancer.

(iii) Diphylobothriasis

This infection is contracted from eating of raw or undercooked fish. The causative organism is *Diphyllobothrium latum*. The definitive hosts are humans and other fish-eating mammals. The intermediate hosts are various freshwater fish and salmon, where larvae are formed.

(iv) Cysticercosis/Taeniasis

The syndrome in human is caused by two species of tapeworm:

- *Taenia sagnata* –beef tapeworm
- *Taenia solium* –Pork tapeworm

Humans are the definitive hosts, and the adult and sexually mature stages develop in humans. Most cases of teaniasis are asymptomatic.

3. ROUNDWORM:

The diseases caused by round worms include:

(i) Trichinosis

This is caused by *Trichinella Spiralis*. The adult forms of *Trichinella spiralis* live in the duodenal and jejunal mucosa of mammals such as swine, canines, bears, marine mammals and humans that have consumed *Trichnellae* infested meat.

(ii) Anisakiasis

This disease is caused by two closely related genera and species:- *Anisakis simplex* and *Pseudoterranova decipiens*. Both of these organisms have several intermediate hosts and generally more than one definitive host. Humans are not final hosts for either, and human disease occurs as a result of humans being accidental interlopers in the cycle of these worms. The definitive hosts are marine mammals eg whales for *Anisakis simplex*.

References:

- Modern food microbiology. 7th Edition – James M.Jay et al.
- Technology of food preservation and processing
Written by EIRI Board of consultants & Engineers.