UNIT 14 DETERIORATIVE FACTORS AND THEIR CONTROL

Structure

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14.0 OBJECTIVES

The objectives of this unit are to enable you to understand the importance of various deteriorative factors associated with food and their control. This unit gives brief account of shelf life of food, biochemical reactions and chemical reactions, nutritional changes in foods and various treatments needed for preservation of food grains etc. After going through this unit, you should be able to:

- know the various types of factors affecting the shelf life of food items
- learn the bio-chemical and chemical reactions which take place in food during storage
- explain about the insects, pests and rodents which cause spoilage and wastage of food
- distinguish the various anti microbial agents used in food
- learn about the various hygiene and sanitation practices to be followed for food processing

14.1 INTRODUCTION

Foods for human consumption can be divided into mainly plant and animal foods. Cereals, sugar products, fruits and vegetables are the main foods from plants. In addition to these are spices and food fungi. Foods from animals are meat, poultry, eggs, fish, seafood, milk and milk products. Sodium chloride is a mineral food, a

flavouring material, an essential nutrient and a chemical preservative. Some foods may be fortified with minerals e.g. iron and calcium compounds are added to flour. Some of the coloring and flavoring materials used in foods are synthetic. Vitamins usually are present in foods but may be added or consumed separately after chemical synthesis or production by microorganisms.

Food quality refers to the composite characteristics which differentiate individual units and enable determination of the degree of acceptability by the consumer and its defined as the measure of the degree of excellence. Consumption of unsafe, contaminated food leads to food borne diseases which cause morbidity and mortality. Microbiological quality of food comprises of these aspects: food safety, shelf-life and consistency. A food should not contain pathogens or its toxin and the total microbial load associated, should be within limits, so that it is not organoleptically spoilt in a short period of time. A food must be of consistent quality both with respect to safety and shelf-life. The principal knowledge of food microbiology helps in assuring a supply of wholesome and safe food to the consumer. The applications help in knowing the effect of processing and producing economically and consistently foods, which have good keeping qualities and are safe to eat. However, the control of processing and environment through Good Manufacturing Practices (GMP) and Hazard Analysis Critical Control Point (HACCP) is of greater importance than even the examination of the end-product. Other than traditional methods to detect food-borne pathogens which use culture media to select and propagate viable cells in food, there are rapid methods which comprise of biochemical tests, specialized media/substrate and DNA and anti-body based detection assays. However as they are used mainly for preliminary screening, they should be combined with the traditional routine analysis.

14.2 SHELF LIFE

Foods can be placed in three groups according to the shelf life.

i) **Stable or nonperishable foods:** These foods do not spoil unless handled carelessly and include products such as sugar, flour and dry beans.

ii) **Semi-perishable foods:** If these foods are properly handled and stored, they remain unspoilt for a fairly long period *e.g.* potatoes, garlic some varieties of apples etc.

iii) **Perishable foods:** This group includes most important daily foods that spoil readily unless special preservatives methods are used. Meats, fish, bread, bun, cakes, poultry, most of the fruits and vegetables, eggs and milk belong to this category.

The shelf life of any food depends on the physical state, structure of the food, chemical properties of the food and the environmental conditions of its production, handling, processing if any and storage.

14.3 CAUSES OF FOOD DETERIORATION

Food can be deteriorated by the following factors:

i) growth of microorganisms

ii) insects

iii) pests

iv) rodents

v) birds

vi) action of enzymes of the plant or animal food

vii) purely chemical reactions *i.e.* those not catalyzed by enzymes of the tissues or of microorganisms

viii) by physical changes such as those caused by freezing, burning, drying, pressure etc.

14.4 CHEMICAL REACTION

The chemical composition of a food determines how satisfactory it will be as a culture medium for microorganisms. Each organism has its own characteristic ability to utilize certain substances as a source of energy, a carbon source and a source of nitrogen.

Chemical changes in stored food take place at rates governed by its temperature and moisture content. The effect of temperature is such that a 10^0C rise, within the temperature range likely to be encountered in temperate or tropical conditions, causes an approximately two fold increase in reaction rates. Cool storage retards changes such as fat oxidation, vitamin loss and container corrosion. Foods such as dried milk benefit from even a small reduction in storage temperature. Keeping such foods cool and dry will usually greatly reduce the rate of development of brown discolouration ('browning') and the associated off-flavours.

The chief factors which influence microbial activity in food are: hydrogen-ion concentration, moisture, oxidation-reduction potential, nutrients and the presence of inhibitory substances Every microorganism has a minimal, maximal and an optimal pH for growth. Yeasts and molds require low pH (below 4.5) for growth while bacteria are favoured by neutral pH. Microorganisms have an absolute demand for water without which no growth can occur. The water requirement is best expressed in terms of available water or water activity (aw). It is the vapour pressure of the solution (of solutes in water in most foods) divided by the vapour pressure of the solvent, usually water. For pure water aw is 1.0. The values permitting growth of spoilage organisms are given in Table 1.

Table 1:

Group of Microorganisms	Minimal aw value
Bacteria	0.91
Yeasts	0.88
Molds	0.80
Halophillic bacteria	0.75
Xerophillic (dry conditions) fungi	0.65
Osnophillic yeast	0.60

In general bacteria require more moisture to grow than yeasts and molds.

14.5 BIO-CHEMICAL REACTION

Putrefaction: Most of the nitrogen in foods is in the form of proteins which is hydrolyzed by enzyme protease increased by proteolytic microorganisms to a number of compounds, polypeptides, simpler peptides or amino acids before they

can serve as nitrogenous food for most organisms. Anaerobic decomposition of proteins, peptides or amino acids result in foul smelling, sulfur containing products such as hydrogen, ammonia, amines, indole etc.

Protein + Proteolytic microorganisms = Amino acids + amines + NH_3 + H_2S.

Carbohydrate fermentation: Carbohydrates are preferred by microorganisms to other energy-yielding foods. They are fermented by micro-organisms to form acids, alcohol and gases. Yeasts cause alcoholic fermentation under anaerobic conditions with ethanol and carbon dioxide as the principal products. Lactic acid bacteria bring about lactose (milk sugar) fermentation yielding lactic acid as the main product. Coliform bacteria carry out coliform type of fermentation with chief products as lactic acid, formic acid, ethanol, carbon dioxide, hydrogen acetoin, butanediol etc.

Carbohydrates + action by microorganisms = Acid + Alcohol + gases

Rancidity: Fats are hydrolyzed by microbial lipase produced by lipolytic microorganisms to produce glycerol and fatty acids. Sometimes autooxidation of fats is more common. This is called rancidity.

Fat + Lipase = Fatty acids + glycerol

14.6 MICRO ORGANISMS: GENERAL PRINCIPLES, CAUSES AND GROWTH

Microorganisms important in food are moulds, yeasts, bacteria, actinomycetes and viruses. They are microscopic in size but are easily visible to the naked eye when large numbers form colonies or surface films, except for viruses. Microorganisms in foods can be either beneficial as in fermented foods such as yoghurt, or harmful causing spoilage and disease. Deterioration of packaging and sealing materials due to mould growth often results in spillage; damp and mouldy cereal grain can become caked and difficult to handle; even the structure of stores can be weakened by certain micro-organisms. The significance of microorganisms in stored food commodities depends upon a number of conditions:

i) the types of microorganism

ii) the number present

iii) the type of food commodity and the treatments to which it has been exposed

iv) whether the foodstuff will be cooked

v) who will consume the food

General Biology

Recognition of all the microbial species that cause spoilage is not possible without a considerable amount of experience and laboratory equipment. However, adequate field recognition of the likely causes of spoilage can often be achieved by simple observation.

Bacteria are the smallest of the micro-organisms normally associated with food spoilage. They may be seen as slimy colonies on some spoilt commodities.

Actinomycetes are similar to bacteria. Actinomycetes will often impart a white, speckled apperance to the grain. *Thermoactinomyces* spp. grow on grain which heats up in store. Some species have been associated with the production of carcinogenic compounds in dried fish products.

Moulds and yeasts are part of larger group of micro-organisms called fungi. Moulds frequently produce a cottony or velvety appearance on spoilt commodities and are also often associated with discolouration of cereal products.

Table 2: Types of food spoilage caused by micro-organisms

Alteration of colour or texture	
Discolouration	– Fungal/bacterial pigments, chemical changes, browning reactions due to microbial heating
Caking	– fungal growth
Physical deterioration	– growth of fungi, bacteria and actinomycetes
Unpalatability	
Fermentation	– acid and gas produced from carbohydrates
Putridness	– Objectionable flavours and odours produced from protein breakdown.
Rancidity	– Objectionable odours and flavours produced by fat hydrolysis and oxidation.
Production of toxic chemicals	
Carcinogens	– *e.g.*, aflatoxins, formed by fungi in the food
Toxic chemicals	– Produced by micro-organisms that may cause chronic or acute toxicity if eaten.
Transmission of disease	– Due to the presence of pathogenic bacteria

Molds

They are aerobic, *i.e.* they require oxygen for growth and most of them require acidic pH for growth. Most of the molds possess a variety of hydrolytic enzymes. Molds of industrial importance are:

i) *Mucor sp*. (produces amylase and used in making oriental foods)

ii) *Rhizopus* sp. (bread mould). It causes spoilage of many foods, berries, fruits, vegetables, bread etc.

iii) *Thamnidium*: It is found on meat in chilling storage

iv) *Aspergillus*: The aspergilli are very widespread. Many are involved in the spoilage of foods and some are useful in the preparation of certain foods. *Aspergillus niger* is used for commercial production of citric and gluconic acids and enzymes.

v) *Pencillium*: This genus is also widespread. *P.expansum* (blue-green spored mold) causes soft rot of fruits. P. camembertii and *P. roquefortii* are used in ripening of camembert and roquefort cheese.

vi) *Alternaria:* They cause spoilage of many foods. *Alternaria citri* causes rotting of citrus fruits.

vii) *Helminthosporium:* Many are plant pathogens but may grow saprophytically on vegetable materials.

Yeasts

Yeasts may be useful or harmful in foods. Yeast fermentations are involved in the manufacture of foods such as bread, beer, wines, vinegar and surface-ripened cheese. Yeasts are also grown for enzymes and for food. Yeasts are undesirable when they cause spoilage of sauerkraut, fruit juices, molasses, honey, meats, wine, beer and other foods. Yeast cells are spherical to ovoid in shape and reproduce asexually by budding or fission. Yeasts of industrial importance are:

i) *Saccharomyces: Saccharomyces cerevisiae* used in leavening of bread, as top yeast for ale, production of alcohol, glycerol and invertase. *S.cerevisiae* var. *ellipsodeus* is used for production of industrial alcohol, wines and distilled liquors.

ii) *Zygosaccharomyces:* They are osmophilic and are involved in the spoilage of honey, molasses etc.

iii) *Pichia & Hansenula* sp.: They form a pellicle on beers or wines.

iv) *Debaryomyces:* They form pellicles on meat brines and grows on cheese and sausages.

Fig.1: Fungal contamination of maize causing discoloration and caking of grains

Bacteria

Two genera, Bacillus (aerobic) and *Clostridium* (anaerobic) form endospores which may remain dormant for years and germinate under favourable conditions. Spores are formed due to nutrient depletion and are heat resistant structures (due to Dipicolinic acid) to tide over the unfavourable conditions. Some genera important in food are:

i) *Acetobacter*: They oxidize ethyl alcohol to acetic acid. They are found on fruits, vegetables, souring fruits and cause spoilage of alcoholic beverages.

ii) *Aeromonas*: It is a pathogen in humans, fish, frogs etc.

iii) *Alcaligenes*: It causes alkaline reaction and causes ropiness in milk.

iv) *Bacillus*: They are aerobic spore formers and may be mesophilic growing at moderate temperature or thermophilic (growing at above 45^0C temperature)

Bacillus sp. Is proteolytic and lipolytic and causes spoilage of many foods. Source is soil.

v) *Clostridium*: They are anaerobic spore formers and many species actively ferment carbohydrates with the production of acids and gases (CO_2+H_2). Source is soil.

vi) *Escherichia*: It is found in faeces and is pathogenic to humans.

vii) *Halobacterium*: They are strict halophiles (salt loving microorganisms) and cause discolouration on salted fish etc.

viii) *Klebsiella*: *Klebsiella pneumoniae* causes pneumonia in humans.

ix) *Lactobacillus*: They are microaerophilic (grow in very less O_2 concentration) and are rod shaped with ability to ferment sugars to lactic acid. They are used in production of fermented dairy products. They may also cause spoilage of wine, beer, refrigerated meats etc.

x) *Listeria*: *Listeria monocytogenes* causes listeriosis.

xi) *Mycobacterium: Mycobacterium tuberculosis* causes tuberculosis and spreads mainly from raw milk from infected cows.

xii) *Salmonella*: They are enteric pathogens and cause food infections.

xiii) *Serratia*: *Serratia marcescens* produces red discolouration in milk and on surface of foods.

xiv) *Shigella*:They are transported by foods and cause bacillary dysentery.

xv) *Staphylococcus*: They cause food poisoning.

xvi) *Streptococus*: *Streptococus pyogenes* causes human septic sore throat and scarlet fever.

xvii) *Vibrio*: *Vibrio cholerae* causes cholera.

xviii) *Yersinia*: *Yersinia peslis* causes human plague.

Viruses

Viruses are the smallest and simplest form of organisms. Unlike bacteria, viruses are not complete cells. Viral pathogens are often transmitted by infected food handlers through contact with sewage. Viruses do not require potentially hazardous foods to survive. Also viruses do not increase in number while they are in food, *e.g.* hepatitis A, Norwalk & Rota virus. Viruses contaminate food through poor hygiene by food handlers, through a contaminated water supply or from sewage contaminated water.

14.7 INSECTS, PESTS AND RODENTS

Insect and mite infestations cause weight loss and deterioration in quality by their presence and feeding activities. When insects are there in food, especially the grains, they produce heat, moisture, waste products and secretions. The heat, moisture and contamination have an adverse effect on the quality of grains and makes them susceptible to further deterioration. Increase in moisture content encourages the growth of molds. Infestation of the beetle *Tribolium* spp., produce large quantities of the chemical benzoquinone, which is toxic and imparts an unpleasant taste and smell. The larvae of mothslips *Ephetia* sp. *Plodia* and *Corcyra* sp., bind flour and grain together into a solid mass. Thus insects and mites can lead to direct loss of food, decrease in quality and results in economic losses. Great care should be taken to prevent high value processed foods e.g. biscuits and meals from being infested. It is impractical to remove insects from such foods so high standard of

hygiene should be ensured in their preparation and storage.

Insects that attack grain are divided into two groups: primary pests and secondary pests. Primary pests are those that are able to attack and breed in previously undamaged cereal and pulse grains. Damage takes the form of holes and cavities within grains. Examples are *Sitophilus* sp., *Rhyzopertha dominica*, *Sitotroga cerealella* etc. Many of these species attack the commodity in the field prior to harvest. Secondary pests are not capable of successfully attacking undamaged grains and solid commodities. They are able to attack materials already infested by primary pests or damaged either by poor threshing, drying and handling. They are also able to attack processed commodities. Examples are *Tribolium* spp; *Oryzaephilus* spp, *Corcyra* spp; *Plodia* spp; etc. There are few species like *Trogoderma Granarium* which do not fit in either category. Dried fish can be attacked by all pest species because its' surface has crevices. Primary pests are usually more destructive than secondary pests in short term storage of cereal grains. However on some commodities for example flour, milled rice, processed and blended foods, secondary pests, may form the majority of insects present.

Rodents such as rats and mice can attack stored foods. Apart from the food eaten, spoilt or contaminated, there are additional 'invisible' losses such as the replacement or repair of packaging materials and the cost of re-bagging spilled food. Rats and mice also feed inedible materials including electrical wiring. Hence their presence in buildings can constitute a fire hazard. Three main rodents are *Rattus norvegicus* (Norway rat) *Rattus rattus* (Roof/ship rat) and *Mus musculus* (House mouse) and important diseases of man transmitted by them are Plague (*Yersinia pestis*), Rat bite fever (*Spirillum minus*), Food poisoning (*Salmonella* spp.), Leptospirosis (*Leptospira spp*.), Rickettsial Pox and Murine typhus (*Rickettsia* spp.) etc. Rodent control is essential for public health and can be done by the use of chemicals in food stores, trapping, proofing and improved hygiene.

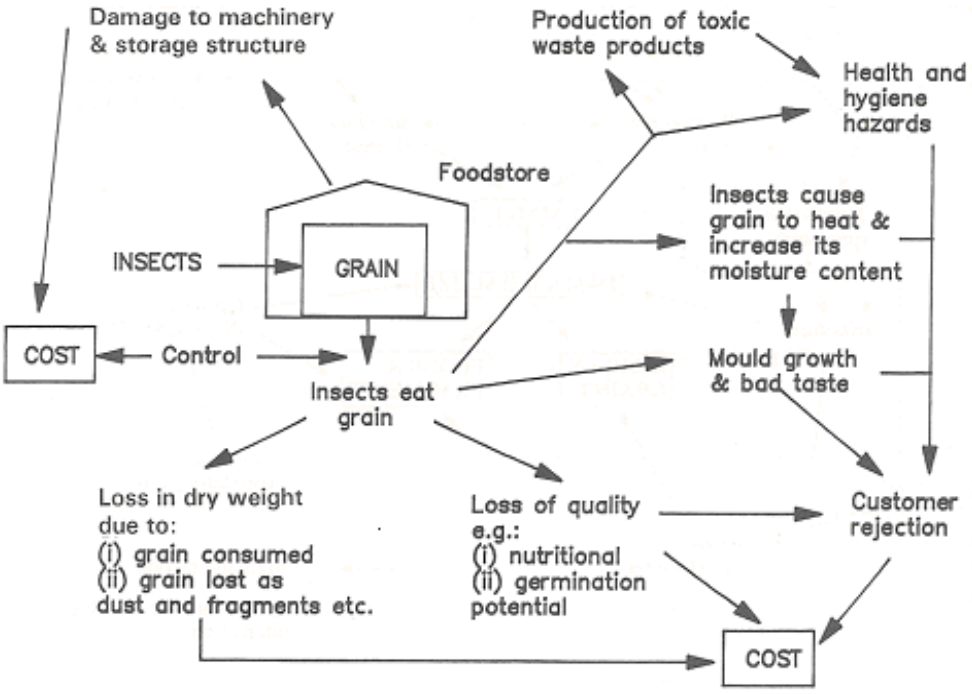

Fig. 2: Economic loss caused by insect and mite infestations

14.8 NUTRITIONAL CHANGES IN FOOD

Food has a variety of organic compounds in it. Numerous kinds of microorganisms can decompose them by hydrolysis caused by microbial enzymes, which lead to a decrease in the nutritional value of the food. There are three types of degradation processes:

i) **Protein biodegradation**: Most of the nitrogen in foods is in the form of proteins which is hydrolyzed by enzyme protease elaborated by proteolytic microorganisms to a number of compounds, polypeptides, simpler peptides or amino acids before they can serve as nitrogenous food for most organisms.

ii) **Carbohydrate biodegradation**: Carbohydrates are preferred by microorganisms to other energy-yielding foods. They are fermented by micro-organisms to form acids, alcohol and gases

iii) **Fat biodegradation:** Fats are hydrolyzed by microbial lipase produced by lipolytic microorganisms to produce glycerol and fatty acids. Sometimes autooxidation of fats is more common.

Thus microorganisms when proliferate on food sometimes elaborate hydrolytic enzymes which leads to hydrolysis of organic constituents, thus decreasing their content. Hence we can say that these hydrolytic microorganisms decrease the nutritional value of food.

14.9 FOOD BORNE DISEASES

Organisms important in food include bacteria, yeast, molds and viruses. Food borne diseases result from the following:

i) Bacteria

ii) Mycotoxins

iii) Viruses

iv) Rickettsias

v) Parasitic worms

vi) Protozoa

Food borne intoxication by bacteria:
This refers food borne illness caused by presence of a bacterial toxin formed in the food. But there has to be large number of organisms (opprox. 10^6/g of food) to produce toxin. They are of two types:

Clostridium botulinum: This food poisoning (botulism) occurs rare but is fatal. Symptoms due to the neurotoxin produced (7 types) is double vision, coated tongue, diarrhoea, paralysis and death within 3-6 days. The foods involved mainly are inadequately processed home-canned food or preserved meat and fish.

Staphylococcus: The toxin produced is an enterotoxin which causes gastro enteritis or inflammation of the lining of the intestinal tract. The foods involved are cream filled bakery goods, milk and milk products, meat and meat products, salads, puddings etc. ordinary cooking cannot destroy the toxin.

Food infections by bacteria

They are mild food poisoning and are of two types. First in those in which food is just a carrier of pathogens causing tuberculosis, diptheria, typhoid, cholera etc.

and second category in which food series as a culture medium for growth of pathogens like *Salmonella, Vibro, Escherichia coli, Clostridium perfringens,Yersinia Shigella listeria, Bacillus cereus* etc. The symptoms are generally same, nausea, chills, diarrhoea, cramps etc. and the number of organisms that can cause the disease are very small. The toxin causes gastro enteritus. Foods involved are fried rice, custards, cereal products, vegetable sprouts and mashed potatoes.

Mycotoxins

Mycotoxins are toxins produced by filamentous molds on foodstuffs. They are secondary metabolites that can cause death in humans and other animals. There are 300-400 compounds recognized as mycotoxins, out of which most common are aflatoxins and others are fumonisins, ochratoxin A, patulin, trichothecenes and zearalenone. Aflatoxins are produced by *Aspergillus flavus* and *Aspergillus parasiticus* and were first isolated from mold-contaminated peanuts in 1962, during which approximately 100,000 turkey birds died and the word mycotoxin was coined. Mycotoxins have been detected in cereals, oil-seeds fruits and vegetables, milk, bees, cocoa. raisins etc. both in field and in storage. Other moulds which produce mycotoxins are *Penicillium citrinum* which produces citrinin, *Fusarium* produces Funonisins and zearalenone and Ochratoxin A from *Aspergillus ochraceus.* Mycotoxins can be used as chemical warfare agents. They can cause carcinogenicity, dermal irritation and other metabolic disturbances.

Fig. 3: *Aspergillus ochraceus* causing discoloration and disintegration of wheat. Ochratoxin was isolated from the infected grains

Ways of controlling mycotoxins

- Prevention of mold growth by proper drying and storage of food.
- Removal of mold damaged food material before storage or processing.
- Proper sanitation with adequate moisture and humidity control of food. A water activity of 0.85 and a temperature range of 25-40 ^{0}C should be avoided as they are optimum conditions for production of mycotoxins. Short periods of high temperature also should be avoided because it causes depression of growth of fungus and accumulation of toxin.

– Use of recommended anti-fungal substances when required.
– Regular chemical and biological assays on food to detect mycotoxins.

Viruses

Viruses are ultramicroscopic parasites able to infect plants, animals and bacteria. There are two main viruses which are food borne:

Poliovirus type causing poliomyelites, infections hepatitis virus causing jaundice and foods usually involved are milk, beverages, sea-food etc. Prevention can be done by maintaining personal cleanliness, adequate heating of foods and disinfection of water. The most important are human calciviruses (Norwalk like viruses) and the hepatitis A virus. Almost all viruses enter food as a result of faecal contamination. They cannot multiply in food. They can be inactivated by high temperature but can persist in refrigerated and frozen foods.

Rickettsias: They are obligate parasites and like viruses they can be cultivated only on living cells. Rickettsia of public health concern is *Coxiella burnetii* which causes Q fever and can be excreted in milk in large quantities and can result in human infection. The pasteurization temperature of 62.8 ^{0}C (30 min.) in milk ensures its complete destruction.

Parasitic Worms: They are occasionally encountered in food and are mainly either flat worms, tape-worm or round worms. Flat worms, *Fasciola hepatica* are common in cattle and sheep, rare in humans and are caused by eating bovine liver. Tape worm *(Taenia Solium* can be present as contaminent in beef or pork. Round worms may be found in swines wild-game animals, under cooked fish etc. Thus cook fish, pork and beef thoroughly and avoid eating raw and under processed meat.

Protozoa: The parasitic infections transmitted by food are caused by *Entamoeba histolytica* which causes amebic dysentery and the source is water contaminated with sewage or faeces. Gardiasis is caused by *Giardia lamblia* which occurs in vegetables washed with contaminated water and can occur in tap water or ice-cream also. Toxoplasma gondil causes Toxoplasmosis and can be avoided by prevention of contamination of food and water from faeces.

Toxigenic algae: A number of algae (dinoflagellates, diatoms and cyanobacteria) can produce very toxic compounds which may be transported to shell fish and a number of distinct illnesses are known such as paralytic, neurotoxic and diarrhoeal shellfish poisoning. These toxins are generally unaffected by cooling.

Toxigenic fungi: These are some wild poisonous mushrooms like *Amanita phalloides, Coprinus atramentarius, Psilocybe cubensis* etc. which may cause gastro intestinal irritation, liver and kidney damage and sometimes even death.

Guidelines to keep food safe

1. Cooking/canning/heating should be adequate and the quality of raw ingredients added should be ensured.
2. There should not be a lapse of 12 hours or more between preparation or eating.
3. Keep meat, fish, poultry and dairy products refrigerated or frozen. Refrigerate leftover promptly.

4. Do not thaw frozen food at room temperature, use a microwave.
5. Never taste or eat raw meat or poultry.
6. Buy only clean, uncracked eggs and use them within 5 weeks of purchase. Do not eat raw or uncooked eggs.
7. Wash fruits and vegetables thoroughly even if you plan to peel them before eating them
8. Left over food should be used within 2-3 days. If in doubt throw it out
9. Contaminated dish cloth can house millions of bacteria after a few hours. One option is to use paper towel to clean and throw immediately.
10. Clean food and everything it touches thoroughly which includes washing food, utensils, counters, sinks etc.

14.10 FOOD ALLERGIES AND POISONING BY CHEMICALS

Food allergies may be caused by the following compounds:

i) Histamines

ii) Mono Sodium Glutamate (MSG)

iii) Nicotinic acid

Poisoning by consumption of chemicals is rather uncommon and symptoms appear within a short time. Antimony, arsenic, cadmium, chlorinated hydrocarbons, copper, cyanide, fluoride, nicotinic acid, lead and zinc in foods can come from utensils, insecticides or fruit sprays.

Ergotism is caused by a fungus, *claviceps purpurea.* This fungus is a parasite of some grasses and cereals and produces a tough, purple brown sclerotium called ergot. Ergots contain alkaloid metabolites which are toxic and cause constriction of blood capillaries.

14.11 ANTI-MICROBIAL AGENTS USED IN FOOD

The anti-microbial agents used in food are grouped into the following three categories:

i) Natural agents: They are not defined as such by law and include natural organic acid like lactic, citric and malic acids. They also include salts such as vinegar (acetic acid), sodium chloride, sugars, spices and their oils woodsmoke, carbondioxide and nitrogen.

ii) Agents generally regarded as safe (GRAS) for addition foods. They are listed in tables.

iii) Agents proved safe and approved by the Food and Drug administration.

The anti-microbial agents are added to food for inhibition or pilling of microorganisms and may be also classified according to their chemical composition, mode of action, specificity, effectiveness and legality. Sugar is effective because of its physical action while sodium benzoate due to its chemical reaction. While sodium chloride has a combination effect some anti-microbial agents are antiseptic in action rather than germicidal. Some are employed to treat wrappers or containers.

Table 1: Some GRAS anti-microbial agents.

Agent	Maximum Concentration allowed	Organisms affected	Foods
Benzoic acid/ benzoates	0.1%	Yeast and molds. It is most effective at 2.5-4.0 pH	Pickles, apple cider, soft drinks etc. Sodium benzoate is used extensively in jams, jellies, carbonated beverages, fruit juices etc.
Sorbic acid/sorbates	0.2%	Molds. It is most effective at low pH.	Used directly as an additive, as a spray, dip or coating on packaging material. Widely used in cheese, cheese products, baked goods, beverages fruit juices, pickles etc.
Propionic acid/ propionates	0.32%	Molds. They affect the cell membrane permeability.	Bread, cakes etc.
SO_2/Sulfites	200-300 ppm	Insects, microorganisms. It causes reduction of disulfide linkages and inhibition of respiratory mechanism	Wine making to reduce the normal flora of grape must, Molasses, dried fruits etc.
Ethylene/prophylene oxides	700 ppm	Yeasts, Molds. Ethylene oxide kills all microorganisms. They are strong alkylating agents attacking labile hydrogens.	Fumigant for spices nuts etc, Sterilants for packaging material and for cold sterlization of plastics, chemicals, syringes etc.
Sodium diacetate	0.32%	Molds	Bread, cheese spreads, treatment for wrappers used on butter.
Nisin	1%	Lactics, *Clostridia*	Certain pasteurized cheese spreads.
Sodium nitrite/ nitrates	120 ppm	*Clostridium botulinum*	Meat curing preparations.

Acetic acid: Acetic acid in the form of vineger is used in pickles, mayonnaise. It is more effective against yeast and bacteria than against molds and its effectiveness increases with a decrease in pH, which would favour the presence of the dissociated acid.

Sugar and salt: Both lowers the water activity and thus have an adverse effect on microorganisms. Sodium chloride is used in brines and curing solutions or is applied directly to the food. It causes plasmolysis of cells and reduces solubility of oxygen in the moisture. Sugars make water unavailable to organisms.

Alcohol: Ethanol, a coagulent and denaturizer of cell proteins is most between 70-95 % concentrations. The alcohol content in beer and wine etc. although cannot prevent spoilage but it, out limits the growth. Propylene glycol has been used as a mold inhibitor and as a spray to airborne microorganisms.

Woodsmoke: Woodsmoke adds desired flavours and number of volatile compounds that have a bacteriostatic (arrests growth of bacteria) or bactericidal effect (pills bacteria). Formaldehuge is most effective of these compounds.

Spices and condiments: They have an inhibitory effect on microorganisms. Cinnamon and cloves contain cinnamic aldehyde and eugenol respectively and are bacteriostatic. But the spices have to be treated to reduce their own microbial content. Onion and garlic have Acroleing and are bacteriostatic or germicidal.

Antibiotics: Many antibiotics have been tested on raw foods, chiefly meat, fish and poultry in order to lengthen the storage time at chilling temperature. Nisin, subtilin, bacitracin etc. has been tried. The FDA has approved chlortetracycline (Sppm) for preserving poultry.

14.12 ENZYME INACTIVATION

Enzyme inactivation prevents product deterioration during storage. Most of the food and microbial enzymes are destroyed at 79.4 ^{0}C with some exceptions. Some proteases and lipases retain their activity even after ultra high-temperature process and can spoil the processed product during long-term storage. Detection of enzyme bovine phosphatase in processed milk indicates that the milk was not properly pasteurized, so it acts as a monitor to check the quality of milk. Blanching is a heat process applied to foods especially fruits and vegetables in which after washing, fruits and vegetables are heated at <100 ^{0}C (86-98 ^{0}C) for 1-5 minutes to inactivate, enzymes, for expulsion of oxygen from tissues and reduction in microbial load.

14.13 TREATMENTS – PHYSICAL, THERMAL CHEMICAL, HIGH PRESSURE, COOLING, FREEZING, MICROWAVE, HEATING, DRYING, DEHYDRATION, IRRIADIATION ETC.

Physical treatment: Physical treatments are used to inhibit, destroy or remove undesirable microorganisms from food without involving chemicals. They include:

– Physical dehydration processes like drying and freeze-drying
– Cooling or cold storage
– Freezing and frozen storage
– Heating (thermal treatment)
– Microwave heat treatment
– UV or ionizing radiations
– Non-thermal methods like high hydrostatic pressure pulsed electric fields etc.

Physical dehydration processes: Food preservation by dehydration is based on the principle that microbial growth is inhibited if the available water required for the growth is removed, thus reducing the water activity (aw). It can be done by hot air, freeze-drying or freeze concentration in case of liquid foods, where

freezing is followed by mechanical removal of frozen water.

Drying: During hot air drying of foods, the air stream has two functions, it transfers heat to evaporate the water from the raw materials, and it carries away the vapour produced. In the course of drying, microorganisms on the raw material are affected by both the drying temperature and changes of **aw.** Wet heat is more lethal than dry heat. The **aw** of dried foods is usually considerably lower than the critical value for microbial growth and hence are stable microbiologically, provided the relative humidity of storage atmosphere less than 70%. The quality of raw materials used and their right handling should also be ensured.

Freeze Drying: Freeze drying (lyophilization) is gentle dehydration of the food in the frozen state through vacuum sublimation of its ice content, thus decreasing the **aw.**

Cool storage: Cool storage generally refers to storage at temperatures from 16 ^{o}C to minus 2 ^{o}C at which the rates of chemical reactions and growth of microorganisms are decreased. However psychrotropic microorganisms can grow at cold temperature such as *Yersinia enterocoitica*, *Vibrio parahaemolytics*, *Listeria monocytogenes* etc. and are important especially in minimally processed, extended shelf-life products. Variation of storage temperature should be avoided.

Controlled-Atmosphere Storage: (CAP) Controlled-atmosphere storage of certain fruits and vegetables is widely used in which there is reduced oxygen content (2-5 %) and an increased (8-10 %) CO_2 content in air tight chilled storage rooms. This modified storage atmosphere depresses the respiration, maintains the sensory and textural quality of perishables and also inhibits the growth of certain spoilage organisms. However in modified atmosphere packaging (MAP), which has a similar principle to CAP, one should ensure the absence of a pathogen, *Clostridium botulinum* which grows in the absence of oxygen.

Freezing and Frozen Storage: Freezing normally lowers the temperature of a food to -18 ^{o}C and the food is stored at this temperature or below. Many convenience food products are made by this technology. However, it is energy intensive. There are three basic freezing methods: (i) Cold air, (ii) indirect contact freezing (iii) submerging food or spraying the cold refrigerant liquid onto the food. At temperatures below about -8 ^{o}C there is practically no microbial growth. However, the thawed frozen products are vulnerable to microbial spoilage and refreezing of thawed products should be avoided.

Preservation by Heat Treatment: The most effective and most widely used method for destroying microorganisms and inactivating enzymes is heat treatment. The heat resistance of various microorganisms is very important from an industrial point of view. Pasteurization is a mild heat treatment aimed at inactivating enzymes and destroying 99-99.9 % of vegetative bacterial cells and to eliminte non-spore forming pathogenic bacteria, thus extending the shelf-life of food. Sterilization refers to complete destruction of microorganisms. Shelf-stable products may contain low number of viable but dorment bacterial spores and these products are called "Commercially Sterile". Commercial sterilization of food in hermetically closed cans or bottles is called canning or appertization. Exposure to ultra high temperature (140 ^{0}C), holding for several seconds and then rapid cooling leads to shelf-stable foods at ambient temperature. The use of higher temperature for short time process leads to less nutrient damage. Heat in the presence of water kills microorganisms

by coagulation of proteins. The destruction rate (D value) is defined as the time in minutes required to destroy 90 % of the microbial population. Thermal death time is defined as the time necessary to kill a given number of organisms at a specified temperature. The slope of the thermal death time curve is expressed by the Z value, which is the temperature increase required to reduce the thermal death time by a factor of 10. For the determination of heat processes for canned foods, F value is defined as the number of minutes at a specific number of viable cells with specific Z value. Thermal death point is the temperature necessary to kill all the organisms in 10 min.

Microwave Heating and Processing: This is becoming popular at the consumer level. Microwaves are electromagnetic waves between infrared and radiowaves at 500 MHz to 10 GHz. The heat produced by the excitation of food molecules results in microbial destruction.

Preservation by Irradiation

UV radiation: The microbiologically most destructive wavelength range of UV radiation is between 240-280 nm at which nucleic acids of cells are damaged. Inactivation is caused by the cross-linking of thymine dimers of the DNA, thus preventing repair and reproduction. Gram negative bacteria are most easily killed by UV, while bacterial endospores, molds and viruses are much more resistant. The UV sources are used mainly for disinfection of air and in aseptic filling of liquid food, in cheese ripening and for disinfection of water.

High-Intensity pulsed light: Decontamination of food surfaces with high-intensity xenon arc lamps is also used.

Ionizing radiation: Food irradiation is limited to gamma rays of ^{60}CO or X rays with energies upto 5 MeV – 10 MeV. The approximate dose to kill microorganisms varies from 0.5 – 200 KGy and for insects is 0.01 – 1 K Gy. The death of microorganisms by ionizing radiation is the result of damage to DNA.

Non-thermal physical preservation methods:

High hydrostatic pressure: High hydrostatic pressure of 300-1000 M Pa can be applied at ambient temperature and microbial inactivation occurs due to cell leakage caused by changes in proteins and cell membrane.

Pulse electric field (PEF): High voltage discharges can kill micro-organisms. An external PFE charges cells in such a way that they behave as small dipoles and an electric potential develops over the cell membranes. For vegetative bacteria the critical electric field strength is 15 KV/cm. The application of PEF has been tried in liquid foods and in meat industry.

14.14 HYGIENE AND SANITATION

Food safety should begin on the farm itself. From planting to consumption there are many opportunities for bacteria, viruses and parasites to contaminate the produce. On the farm, soil, manure, water, animals, equipments and workers may spread harmful microorganisms. Produce may be harvested on a farm, processed in one plant, repacked in another, then stored, displayed or served. Each of the steps is an opportunity for harmful microorganisms to enter the food supply. Clean soil, clean water, air and surfaces and hygiene among workers, can minimize the pathogen contamination. But there is no way to guarantee that every thing we grow

and consume is free of harmful microbial contamination. The risk can be reduced if preventive steps are taken. Animal manure and human faecal matter represents a significant source of human pathogens and thus should be closely managed to limit the potential for pathogen contamination. Risk assessment should be done, HACCP approach should be used to assess the risks, identify critical control points, appropriate preventive measures, and to record the plant hygiene and to determine corrective measures in case of non-conformity. Bacteriological monitoring and record keeping should be there. Thus keeping microbial loads at minimum levels is essential to provide safe food of high quality.

HACCP Plan for food-processing:

HACCP concept provides a systematic structured approach to assure the safety of a food product. However, there is no blueprint or universal formula for putting together the specific details of a HACCP plan. For it to work well, it should be a participatory endeavor at all levels of an organization, both in formulating and managing the plan. The integrity in the entire food chain cannot be guaranteed as a result of farm level vulnerability Hence farm level HACCP is also very important.

Steps to control hygiene in food-processing:

- **Site and infrastructure of food-processing:** The site should be on cleared ground. It should be away from sources of insects/rodents/smells/compost piles/ wells/water source. There should be road access for bringing in raw materials. The surrounding area should be planted with grass, as short grass acts as a very efficient trap for airborne dust. The building should be painted and tiled. ceilings ,doors, working surface and walls should be smooth, non-toxic and easy to disinfect. At the bottom of the wall where it meets the floor, there should be a right angle so that the dust is not collected. The windows should be such so that they are easily cleaned.
- **Washing & toilet facilities:** They should be separated preferably in a separate building or two closed doors should be there between the toilet and the processing area to prevent insects/odours. The toilet facilities should be fly proof.
- **Drains:** The drains should be covered by a fine mesh and drainage should be proper, floors (slope) and should be self-drained.
- **Electric points:** They should be fixed at least 1.0-1.5 m high on the walls to keep them dry.
- **Building and equipment:** They must be kept clean at all times. Equipment should be made up of smooth, non-toxic material and should permit adequate cleaning. Workers must be trained to keep equipment clean through the day and to remove wastes from the building. All dust from high window ledges/ old sacks etc. should be removed so as to avoid breeding of insects.
- **Sanitization:** Sanitization can be done by the use of bleaching powder *i.e.* (30-40 g/litre) 1 %, 2 ml in 100 ml. (200 ppm), 1 ml in 100 ml for processing table, utensils and equipments for floors the concentration required is 200 ppm and for drains it is 250-500 ppm.
- **Processing room:** No food should be eaten in the processing room.
- **Water Supply:** The water supply should be potable/ WHO guidelines and cater to the standards specified for washing and drinking purpose. Non-potable water should not be allowed to mix into the potable water systems.
- **Air quality, ventilation & lighting:** All of them should be adequate as to

avoid food contamination. Natural or mechanical ventilation should be provided to control air-borne contamination of food and control of odours and humidity. Adequate natural or artificial light should be provided to ensure that food is not contaminated by breakages.

- **Storage:** Adequate storage facilities should be provided which allow effective cleaning and suitable environment to minimize deterioration of food. Raw material & processing area should be separate as to avoid cross contamination. All externally opening doors and should be fitted with automatic air curtains. There should be fly proofing of all windows, ventilators, drain outlets/holes should be netted. The fly catchers should be away from food handling points.
- **Personal hygiene:** The infected workers should be excluded from work and all workers should undergo periodic training on the important role of personal hygiene in the quality of the processed product. Adequate means of hygienically washing and drying of hands and suitable toilets should be provided.

Training

Food handlers should be trained in the basic concepts and requirements of food and personal hygiene as well as those aspects particular to the specific food-processing operation. The level of training varies depending on the type of operation and the precise job description of the employee, however some form of induction training with regular updating or refresher courses is an absolute minimum.

Training should give food handlers an understanding of the basic principles of hygiene, why it is necessary, and how to achieve it in practice. A core curriculum for any such course should emphasize:

1) Micro-organisms as the main cause of food spoilage and food borne illness and the characteristics of the common types of food poisoning.
2) How to prevent food poisoning through the control of microbial growth, survival or contamination.
3) Standards of personal hygiene required of food handlers. These are principally to avoid contamination of food with bacteria. The food handler may harbour as part of the body's flora, *e.g.* Staph, aureus, Salmonella or that they may bring in with them from the outside world, *e.g.* Listeria, B. cereus.
4) Some do's and don'ts associated with good personal hygiene are listed later
5) Principles of the handling and storage of foods such as the correct use of refrigerators and freezers, the importance of temperature monitoring, the need for stock rotation and the avoidance of cross-contamination.
6) Correct cleaning procedures and the importance of the 'clean-as-you-go' philosophy.
7) Knowledge of the common pests found in food premises and methods for their exclusion and control.
8) An introduction to the requirements of current food legislation.

Some do's and don'ts of personal hygiene for food handlers:

DO' S

Wash your hands regularly throughout the day and especially:

– after going to the toilet;
– on entering a food room and before handling food or equipment;

– after handling raw foods;
– after combing or touching the hair;
– after eating, smoking, coughing or blowing the nose;
– after handling waste food, refuse or chemicals.
– Keep fingernails short and clean
– Cover cuts, spots or boils with a waterproof dressing
– Keep hair clean and covered to prevent hair/dandruff entering the food
– Always wear clean protective clothing (including footwear) in food processing areas.

DON'T'S

– Do not smoke; chew gum, tobacco, betel nut, fingernails or anything else.
– Do not taste food
– Do not spit, sneeze or cough over food
– Do not pick nose, ears or any other body site
– Do not wear jewellery when handling food.
– Do not wear protective clothing outside the production areas

Check Your Progress

Note: a) Use the spaces given below for your answers
b) Check yours answers with those given at the end of the unit.

1. List the factors which lead to food deterioration.

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2. What are the biochemical changes in food due biodeterioration and how they affect nutrition of the food?

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3. Name food borne diseases. Which is the most fatal bacterial food poisoning.

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4. How can enzymes be inactivated?

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5. How can food be preserved?

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6. Give HACCP concept.

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7. Give some factors which help maintain hygiene and sanitation.

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14.15 LET US SUM UP

The shelf life of any food depends on the physical state, structure of the food, chemical properties of the food and the environmental conditions of its production, handling, processing if any and storage. Food can be deteriorated by the following factors: growth of microorganisms, insects, pests, rodents, birds, action of enzymes of the plant or animal food etc. The chief factors which influence microbial activity in food are: hydrogen-ion concentration, moisture, oxidation-reduction potential, nutrients and the presence of inhibitory substances Every microorganism has a minimal, maximal and an optimal pH for growth. Microorganisms important in food are moulds, yeasts, bacteria, actinomycetes and viruses.

The significance of microorganisms in stored food commodities depends upon a number of conditions: the types and number of microorganism, food commodity and treatments. The organisms that cause deterioration of stored foodstuffs all need a suitable moisture content and a favourable temperature to grow and reproduce. The maintenance of cool and dry conditions in and around food commodities will minimize damage.

Numerous kinds of microorganisms can decompose by hydrolysis caused by microbial enzymes, which lead to a decrease in the nutritional value of the food. Food borne diseases result from the following: Food borne toxification by bacteria, food infections by bacteria, Mycotoxins, Viruses, Rickettsias, Parasitic worms, Protozoa etc.

Physical treatments are used to inhibit, destroy or remove undesirable microorganisms from food without involving chemicals. They include: Physical dehydration processes like drying and freeze-drying, Cooling or cold storage, freezing and frozen storage, Heating (thermal treatment), Microwave heat treatment, UV or ionizing radiations, Non-thermal methods like high hydrostatic pressure pulsed electric fields etc. Food can also be preserved by various anti-microbial agents.

Food safety should begin on the farm itself. Clean soil, clean water, air and surfaces and hygiene among workers, can minimize the pathogen contamination. But there is no way to guarantee that every thing we grow and consume is free of harmful microbial contamination. The risk can be reduced if preventive steps are taken. Risk assessment should be done, HACCP approach should be used to assess the risks, identify critical control points, appropriate preventive measures, and to record the plant hygiene and to determine corrective measures in case of non-conformity. Thus keeping microbial loads at minimum levels is essential to provide safe food of high quality.

14.16 KEY WORDS

Water activity	**:**	It is the vapour pressure of the solution (of solutes in water in most foods) divided by the vapour pressure of the solvent, usually water. For pure water aw is 1.0.
Fermentation	**:**	Acid and gas produced from carbohydrates
Putridness	**:**	Objectionable flavours and odours produced from protein breakdown.
Rancidity	**:**	Objectionable odours and flavours produced by fat hydrolysis and oxidation.
Bacteria	**:**	Are the smallest of the micro-organisms normally associated with food spoilage. They may be seen as slimy colonies on some spoilt commodities.
Actinomycetes	**:**	Are similar to bacteria. Actinomycetes will often impart a white, speckled apperance to the grain.
Fungi	**:**	Moulds and yeasts are part of larger group of micro-organisms called fungi. Moulds frequently produce a cottony or velvety appearance on spoilt commodities and are also often associated with discolouration of cereal products.
Viruses	**:**	Viruses are the smallest & simplest form of organisms. Viral pathogens are often transmitted by infected food handlers through contact with sewage. Viruses do not require potentially hazardous foods to survive.
Carcinogens	**:**	Aflatoxins, formed by fungi in the food.
Toxic chemicals	**:**	Produced by micro-organisms that may cause chronic or acute toxicity if eaten.
Transmission of disease	**:**	Due to the presence of pathogenic bacteria
Insect, mites and rodents	**:**	Insect and mite infestations cause weight loss and deterioration in quality by their presence and feeding activities.
Food borne toxification by bacteria	**:**	This refers food borne illness caused by presence of a bacterial toxin formed in the food.

Parasitic Worms : They are occasionally encountered in food and are mainly either flat worms tape-worm and round worms

Protozoa : The parasitic infections transmitted by food are caused by *Entamoeba histolytica* , *Giardia lamblia and Toxoplasma gondii* .

Toxigenic algae : A no. of algae (dinoflagellates, diatoms and cyanobacteria) can produce very toxic compounds which may be transported to shell fish and cause a number of distinct illnesses such as paralytic, neurotoxic and diarrhoeal shellfish poisoning.

Toxigenic fungi : These are some wild poisonous mushrooms like *Amanita phalloides, Coprinus atramentarius, Psilocybe cubensis* etc. which may cause gastro intestinal irritation, liver and kidney damage and sometimes even death.

Anti-microbial agents : The anti-microbial agents are added to food for inhibition or killing of microorganisms and may be also classified according to their chemical composition, mode of action, specificity, effectiveness and legality.

Food safety : Clean soil, clean water, air and surfaces and hygiene among workers, can minimize the pathogen contamination done, HACCP approach should be used to assess the risks, identify critical control points, appropriate preventive measures, and to record the plant hygiene and to determine corrective measures in case of non-conformity.

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14.18 ANSWERS TO CHECK YOUR PROGRESS

1. **Food can be deteriorated by the following factors:** growth of microorganisms insects, pests ,rodents, birds, action of enzymes of the plant or animal food, purely chemical reactions i.e. those not catalyzed by enzymes

of the tissues or of microorganism by physical changes such as those caused by freezing, burning, drying, pressure etc.

2. **Fermentation:** Acid and gas produced from carbohydrates

   Putridness: Objectionable flavours and odours produced from protein breakdown.

   Rancidity: Objectionable odours and flavours produced by fat hydrolysis and oxidation.

   Nutrition decreases with biodeterioration.

3. Food borne toxification by bacteria, Food infections by bacteria, Mycotoxins, Viruses, Rickettsias, Parasitic worms, Protozoa .

   Clostridium botulinum

4. Enzyme inactivation prevents product deterioration during storage. Most of the food and microbial enzymes are destroyed at 79.4 ^{0}C with some exceptions. Some proteases and lipases retain their activity even after ultra high-temperature process and can spoil the processed product during long-term storage. Detection of enzyme bovine phosphatase in processed milk indicates that the milk was not properly pasteurized, so it acts as a monitor to check the quality of milk. Blanching is a heat process applied to foods especially fruits and vegetables in which after washing, fruits and vegetables are heated at <100 ^{0}C (86-98 ^{0}C) for inactivation of enzymes, expulsion of oxygen from tissues and reduction in microbial load.

5. They include: Physical dehydration processes like drying and freeze-drying, Cooling or cold storage, freezing and frozen storage, Heating (thermal treatment), Microwave heat treatment, UV or ionizing radiations, Non-thermal methods like high hydrostatic pressure pulsed electric fields etc. Food can also be preserved by various anti-microbial agents.

6. HACCP concept provides a systematic structured approach to assure the safety of a food product. However, there is no blueprint or universal formula for putting together the specific details of a HACCP plan. For it to work well, it should be a participatory endeavor at all levels of an organization, both in formulating and managing the plan.

7. Clean soil, clean water, air and surfaces and hygiene among workers, can minimize the pathogen contamination, HACCP approach should be used to assess the risks, identify critical control points, take appropriate preventive measures, and to record the plant hygiene and to determine corrective measures in case of non-conformity.