UNIT 4 FOOD CONSTITUENTS – PROTEINS, ENZYMES AND WATER

Structure

4.0 OBJECTIVES

After reading this unit, you will be able to answer:

- what are proteins and amino acids? How proteins are classified? Protein structure, their Denaturation and the role of protein and amino acids in non-enzymatic browning. Proteins of plant, seeds, marine and animal origin. Classification and properties of enzymes etc.; and
- you will also learn the importance of the state of water in foods in food preservation, freezing of water, water quality and standards and water analysis.

4.1 INTRODUCTION

Unlike the other two major nutrients viz., carbohydrates and lipids that are essentially energy sources, proteins constitute the main structure of the animal and human body. These constituents characterised by their nitrogen content are involved in many vital processes intricately associated with all living matter. Some proteins function as biocatalysts (enzymes). There are different types of enzymes in all living systems. They catalyse most of the biological reactions. As enzymes have high degree of specificity, mostly one enzyme can catalyse only one reaction. Several enzymes like amylase, invertase, glucose oxidase, Pectinases, proteases, find application in food processing.

Water is an essential constituent of foods. The state of water in foods has great bearing on food preservation. The physical, chemical and microbiological quality of water used in food processing operations should conform to certain minimum standards.

4.2 PROTEINS

Protein is one of the three major basic nutrients required for growth and development, the other two being carbohydrates and lipids. The word protein was coined from the Greek proteios, which means ‘of the first rank’. Proteins are very complex organic substances, constitute the main structure of the animal and human body. These macromolecules, characterized by their nitrogen contents are involved in many vital processes intricately associated with all living matter. Some proteins function as biocatalysts (enzymes) and hormones to regulate chemical reactions within the body.

4.2.1 Amino Acids

Amino acids are the building blocks of proteins. Proteins are polymers of some 20 amino acids joined together in different proportions and sequences. Most amino acids have general chemical structure as given in Figure 4.1.

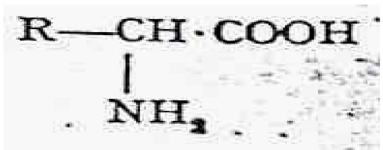

Figure 4.1: General chemical structure of amino acids where R= aliphatic, aromatic, heterocyclic etc. groups

They have both amino group and acidic carboxyl group. In proteins the amino acids are joined together by peptide bond (-CO-NH-) (Fig.2) i.e. The carboxyl group of one amino acid is linked with the amino group of the second amino acid with elimination of H_2O.

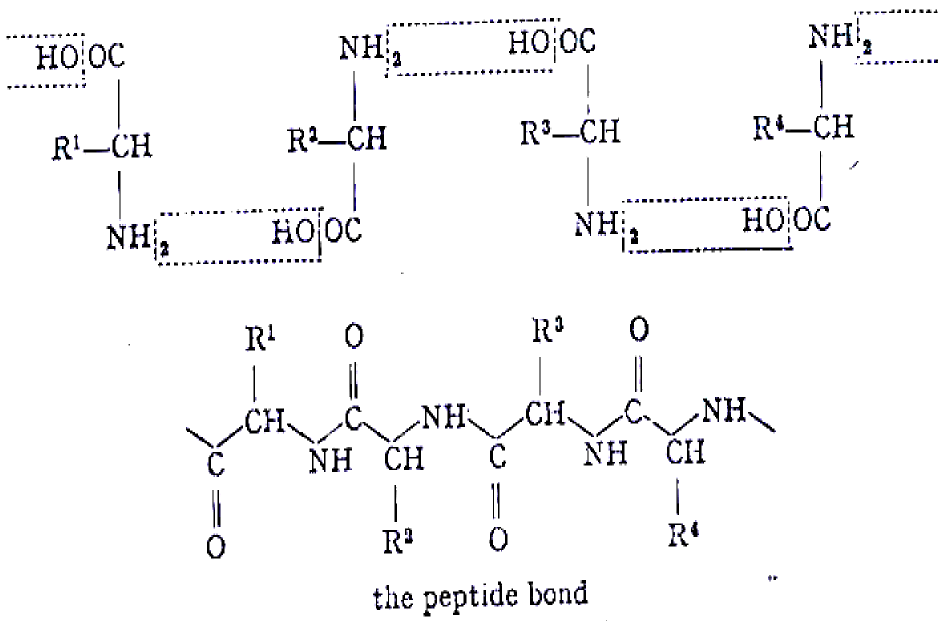

Figure 4.2: The peptide bond

Amino acids found in nature are classified into six groups: viz.,

1. Aliphatic mono amino mono carboxylic amino acids e.g. glycine, alanine, valine, leucine, isoleucine, serine and threonine.

2. Sulphur containing amino acids e.g. cysteine, cystine and methionine.

3. Aliphatic mono amino dicarboxylic amino acids, e.g. aspartic acid and glutamic acid.

4. Aliphatic basic amino acids, e.g. lysine, arginine and histidine.

5. Aromatic amino acids, e.g. phenylalanine and tyrosine.

6. Heterocyclic amino acids, e.g. triptophan and proline.

Of the above amino acids, eight for adults and ten for children are considered essential or indispensable for the human diet. They are: lysine, triptophan, phenylalanine, threonine, valine, methionine, leucine and isoleucine. The amount of these amino acids present in a protein and their availability determine the nutritional quality of the protein. In general animal proteins are of higher quality than plant proteins. However, plant proteins can be upgraded nutritionally by judicious blending. That is why, traditionally people consume a variety of pulses. Egg protein is one of the best quality proteins and is considered to have a biological value of 100. It is widely used as a standard and protein efficiency ratio (PER) values are sometimes based on egg white as a standard. Cereal proteins are generally deficient in lysine and threonine. Soya is a good source of lysine but deficient in methionine. Cottonseed protein is deficient in lysine and groundnut protein in methionine and lysine. The protein of potato although present in small quantity is of excellent quality and is equivalent to that of whole egg.

4.2.2 Protein Classification

Proteins are divided into two main groups namely simple and conjugated and derived proteins.

Simple Proteins

Simple proteins yield only amino acids on hydrolysis and include the following classes:

a) Albumins: They are soluble in neutral salt free water. Usually these are proteins of relatively low molecular weights. E.g. egg albumin, lactalbumin and serum albumin in the whey proteins of milk, leucosin of cereals, legumelin in legumes.

b) Globulins: They are soluble in salt solutions and almost insoluble in water. E.g. Serum globulins and β-lacto globulin in milk, myosin and actin in meat, glycinine in soybean.

c) Glutelins: Soluble in very dilute acids or bases, insoluble in neutral solvents. E.g. Wheat glutelin and oryzenin in rice.

d) *Prolamins:* Soluble in 50-80% ethanol and insoluble in water. E.g. Gliadin in wheat, zein in corn and hordein in barley.

e) *Scleroproteins:* Insoluble in water and neutral solvents and resistant to enzymatic hydrolysis. These are fibrous proteins serving structural and binding purposes. Gollagen of muscle tissue, elastin of tendens, creatin of hair and fibroin of silk are examples.

f) *Histones:* Basic proteins containing a large number of basic amino acids like lysine and arginine. Soluble in water and precipitated by ammonia.

g) *Protamins:* Strongly basic proteins of low molecular weights. They are rich in arginine. E.g. Cupein from herring, and scombrin from mackerel.

Conjugated Proteins

Conjugated proteins contain an amino acid part combined with a non protein material such as lipids, nucleic acid, carbohydrates and others. Some of the conjugated proteins are:

1. *Phospho proteins:* They constitute an important group including many major food proteins. This group includes casein of milk and the phosphoprotein of egg yolk.

2. *Lipoproteins:* These are combination of lipids with proteins and have excellent emulsifying capacity. Lipo protein occurs in milk and egg yolk.

3. *Nucleoproteins:* These are combination of nucleic acids with protein. They are found in cell nuclei.

4. *Glycoproteins:* These are combination of carbohydrates with protein. Ovomucine of egg white is an example.

5. *Chromo protein:* These are proteins with coloured prosthetic groups. Hemoglobin, myoglobin, chlorophyll and flavo proteins are examples.

4.2.3 Protein Structure

The Primary structure of proteins is related to the peptide bonds between the component amino acids and also to the amino acid sequence in the molecule. A peptide chain may become involved in hydrogen bonding between amide nitrogen and carbonyl oxygen. These bonds may be formed between different areas of the same polypeptide chain or between adjacent chains. Such bonds establish the secondary structure of proteins, which may be of helical or sheet form.

The tertiary structure of protein is established when the chain are folded into compact structures stabilized by hydrogen bonds, disulphide bridges, etc. Large molecules may form quaternary structures by association of sub units.

4.2.4 Protein Denaturation

Denaturation is a process of change in structure of proteins without breaking covalent bonds. The process is peculiar to proteins and affects different proteins to different degrees. Denaturation can be brought about by a variety of agents of which the most important are heat, pH, salts and surface effects. The destruction of enzyme activity by heat is one of the most important operations in food processing. You are familiar with the coagulation or hardening of egg white on heating it. It is due to denaturation of egg albumin. Freezing can also cause protein denaturation as in the case of fish, which becomes tough on freezing and thawing. Milk protein casein and gelatin are examples of proteins, which can be boiled with out apparent denaturation.

Check Your Progress Exercise 1

Note: a) Use the space below for your answer.
b) Compare your answers with those given at the end of the unit.

1. Name the different groups of amino acids.

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2. Define albumins and globulins.

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3. What are phosphoproteins and lipoproteins?

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4.2.5 Non-Enzymatic Browning

Browning of several foods is familiar to all of us. Browning of potato chips, brown crust formation on bread and cakes, browning of evaporated milk, browning of jams, candies, fruit juice concentrates etc. are all examples of non-enzymatic browning. The browning reaction can be defined as the sequence of events, which begins with the reaction of the amino group of amino acids, peptides or proteins with a glycosidic hydroxyl group of sugars and terminates with the formation of brown nitrogenous polymers or melanoidins. While some browning reactions like brown crust formation on bread and cake are desirable, most others are undesirable and may accompany formation of off flavoured compounds.

Methods of preventing browning could consist of measures indented to slow the reaction rates such as control of moisture, temperature or pH or removal of an active intermediate. Generally it is easy to use an inhibitor. One of the most effective inhibitors of browning is sulphur dioxide. It is known that sulphur dioxide can combine with the carbonyl group of an aldose to give an addition compound thus blocking further transformations leading to formation of dark coloured compounds.

$$NaHSO_3 + R\text{-}CHO \rightarrow RCHOHSO_3Na$$

However, as sulphite can destroy the vitamin thiamine, it is not desirable to use it to inhibit browning in foods, which are good sources of this vitamin.

4.2.6 Protein from Different Sources

Human requirement of proteins is met from both animal and plant sources.

Proteins of Animal Origin

A typical adult mammalian muscle contains 18-20% protein. Muscle proteins are categorized on the basis of their origin and solubility as sarcoplasmic, myofibrillar and stroma proteins. Protein content of milk ranges from 3 to 4%. Buffalo milk has slightly higher level of protein. Milk proteins are grossly divided into casein and whey protein, the average ratio being 80:20. Egg contains on an average 11% shell, 31% yolk and 58% white. The yolk contains about 50% solids of which one third is protein and two third lipids. Egg white is essentially an aqueous solution containing about 12% protein.

Proteins of Marine Origin

Fish flesh contains on an average 10-21% protein. Fish muscle proteins, like those of mammalian muscle are generally classified as sarcoplasmic, myofibrillar and stroma proteins but their proportions differ.

Proteins of Plant Origin

The protein source of vegetarian diet is from cereals, pulses and oil seeds besides small quantities from vegetables.

Cereals like wheat and rice are important sources of protein because they are the staple foods of Indians. On an average wheat has 12-13% protein while rice has 7-9% protein. Gluten proteins are responsible for the unique bread making property of wheat. Wheat and rice proteins are generally deficient in the essential amino acid lysine.

Seed Proteins

Legumes (pulses) and oil seeds are major sources of vegetable proteins. The average protein content of the major pulses is given in Table 4.1.

Table 4.1: Protein content of major pulses

Legume	Protein (%)
Bengal gram dhal	20.8
Black gram dhal	24.6
Field bean, dry	24.9
Green gram dhal	24.5
Lentil (lens culinaris Medic)	25.1
Peas, dried	19.7

The average protein content of some of the oil seeds is given in Table 4.2.

Table 4.2: Protein content of some oil seeds

Oilseed	Protein (%)
Ground nut	26.7
Soybean	43.2
Sesame	18.3
Cotton seed	19.5
Sunflower seed	12.5

Check Your Progress Exercise 2

Note: a) Use the space below for your answer.
b) Compare your answers with those given at the end of the unit.

1. Explain protein denaturation.

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2. What are the major difference between proteins of animals and plants?

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3. Name four good sources of plant proteins.

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4 Which are essential amino acids?

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4.3 ENZYMES

Enzymes are called bio- catalysts. They are globular protein catalysts that accelerate several biological reactions. Enzymes are present in all biological systems both plant and animal. Enzymes show high degree of specificity. This property is very important in food processing where it is often desirable to modify only a single component in the process.

4.3.1 Nomenclature and Classification

Enzymes are classified into six major groups depending on the type of reactions they catalyze i.e. 1) oxido reductases, 2) transferases, 3) hydrolases, 4) lyases, 5) isomerases and 6) ligases

Over the years, thousands of enzymes have been isolated and identified. Quite often two or three enzymes differ only slightly from one another in their properties causing difficulty in naming them. Therefore, the International Union of Biochemistry on Nomenclature and Classification of Enzymes has assigned a code number of four numerals for each enzyme, which fully identifies an enzyme. However it is too cumbersome to follow this classification and therefore for routine purpose most of the enzymes have been given a trivial name, which is short and simple.

4.3.2 Properties of Enzymes

As already mentioned, all enzymes are proteins but all proteins are not enzymes. Some enzymes consist of protein only, but most enzymes contain additional non- protein components such as carbohydrates, lipids, metals, phosphates or some other organic moiety. The complete enzyme is called holoenzyme, the protein part apoenzyme and the non- protein part cofactor. The compound which is being converted in an enzyme reaction is called substrate, In an enzyme reaction the substrate combines with the holoenzyme and is released in a modified form as shown below.

$$\text{Enzyme} + \text{Substrate} \leftrightarrows \text{Complex} \leftrightarrows \text{Enzyme} + \text{Products}$$

Enzyme activity is affected by various factors like temperature, pH, chemicals etc. By far the most important is the requirement of optimum temperature and pH for their maximum activity. These properties serve both for obtaining maximum activity for an enzyme as well as for inhibiting the enzyme activity.

By far the largest group of enzymes important in food processing is the hydrolases. A few oxido reductases and isomerases are also encountered. Hydrolases catalyse the following general reaction.

$$AB + HOH \rightarrow HA + BOH$$

Typical examples of hydrolases are amylases, pectin esterase, poly galacturonase, proteases, lipases etc. Most of the enzymes used in industrial applications are now obtained from microorganisms. Table 4.3 gives examples of some of the important enzymes encountered in food processing.

Table 4.3: Some enzymes used in food processing

Enzyme	Food	Purpose/ action
Amylases	Baked products Brewing	Increase sugar content for yeast fermentation Conversion of starch to maltose for fermentation
Invertase	Artificial honey	Conversion of sucrose to glucose and fructose
Naringinase	Citrus juice	De-bittering of citrus juice
Pectinases	Fruit juice	Improve yield and clarity
Proteases	Brewing Meat and fish	Clarification, chill-proofing Tenderization
Lipases	Oils	Hydrolytic rancidity (deteriorative)
Glucose oxidase	Egg powder	Prevention of browning by removing glucose
Polyphenol oxidase	Fruits	Enzymatic browning (deteriorative)

4.3.3 Immobilized Enzymes

One of the most important recent developments in the use of enzymes in industrial food processing is the fixing of enzymes on water insoluble inert supports. The fixed enzymes retain their activity and can be easily added or removed from the reaction mixture. The use of immobilized enzymes permits continuous processing and repeated use of the enzyme. Another important use of immobilized enzymes is in analytical and medical fields.

Check Your Progress Exercise 3 ✍

Note: a) Use the space below for your answer.
b) Compare your answers with those given at the end of the unit.

1. What is the role of enzymes?

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2. Are all proteins enzymes?

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3. Explain immobilized enzymes?

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4.4 WATER

Water is an essential constituent of most foods. It may occur as an inter cellular and/or extra cellular component in vegetables and animal products, as a dispersing medium or solvent in a variety of products, as the dispersed phase in some emulsified products such as butter or margarine and as a minor constituent in other cases. The presence of water influences deterioration of food, either chemical or microbiological. Thus an understanding of its properties and its behaviour in foods is of great importance.

4.4.1 State of Water in Foods

Water is present in foods in different forms. They are grossly characterised as bound water and free water. Bound water is the water, which is bound to other constituents of foods like proteins and remains unfrozen. This water is unavailable as a solvent. The state of water in food is described by the relationship between the moisture content of the product and the relative humidity of the air surrounding it. This ratio is called water activity, which is an important characteristic of the system. The relative humidity corresponding to each specific moisture content of the product called equilibrium relative humidity (ERH) and the following relationship applies.

$$a_w = p/p_o = ERH/100$$

where a_w = water activity
p = partial pressure of water in food
p_o = vapour pressure of water at the same temperature
ERH = Equilibrium Relative Humidity in %

At high moisture contents, when the amount of moisture exceeds that of solids, the activity of water is close to or equal to 1.0. When the moisture content is lower than this amount, water activity is lower than 1.0. Below moisture content of about 50% the water activity decreases rapidly and the relationship between water content and relative humidity is represented by the sorption isotherm. The adsorption and desorption processes are not fully reversible;

therefore, a distinction can be made between the adsorption and desorption isotherms according to whether a dry product is subjected to increasing moisture levels or whether the moist product is gradually equilibrated with lower moisture levels and the product is being dried. Figure 4.3 shows the adsorption desorption isotherms.

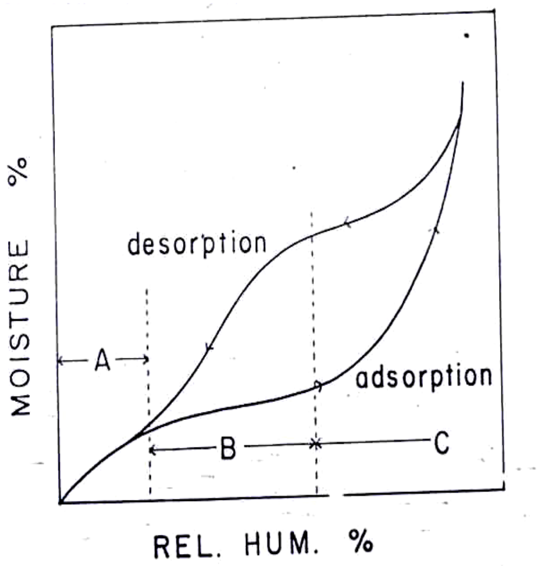

Figure 4.3: Adsorption desorption isotherms

Generally the isotherms are required for the observation of hygroscopic products and the desorption isotherms are useful for investigation of the process of drying. A steeply sloping curve indicates that the material is hygroscopic and a flat curve points to a product, which is not sensitive to moisture.

4.4.2 Water Activity and Food Spoilage

Moisture content and water activity are of major importance in affecting a progress of chemical and microbiological spoilage reactions in foods. Dried and dehydrated foods have very good microbiological stability though browning reactions take place. Such products have moisture content in the range of 5-15% and have low water activity. Intermediate moisture foods (IMF) have moisture content in the range of 20–40% and are fairly shelf stable. IMFs generally have water activity above 0.5. Adding/having soluble solids like sugar achieve this. Example of such products is jams, preserves, cakes, dry fruits, etc. Bacterial growth is practically nil below water activity of 0.90. Yeasts and moulds are usually inhibited between 0.88 and 0.80 water activity, although some osmophilic yeast can tolerate water activity as low as 0.65.

Most enzyme reactions are inhibited below water activity of 0.85. Non-enzymatic browning reactions are dependent on water activity showing maximum around 0.6–0.7. Since water activity is a major factor influencing the keeping quality of a number of foods, it is obvious that packaging can play an important role to maintain optimal conditions for long storage life. Packaging aspects will be dealt with in another section.

4.4.3 Freezing of Water

During freezing of water, the water molecules arrange themselves in a tetrahedral fashion. These results in a hexagonal crystal lattice which is loosely built and has relatively large hallow spaces resulting in high specific volume. This is the reason for increase in volume of water on freezing. You must have observed ice cubes floating on water. The density of ice at 0° C is only 0.9168.

Water can exist in three phases, viz., solid, liquid and gas. The conditions under which they exist are separated by three equilibrium lines (Figure 4.4).

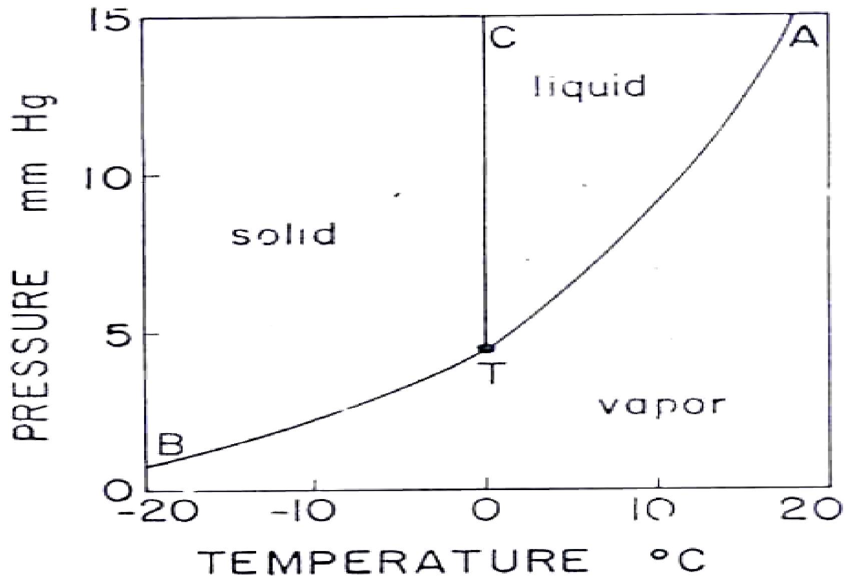

Figure 4.4: Phase diagram of water

The vapour pressure line TA, the melting pressure line TC and sublimation pressure line BT. They meet at the triple point T, where all three phases are in equilibrium. When ice is heated at pressures below 4.58 mm Hg it changes directly into vapour form (sublimation). This is the basis of freeze-drying.

Slow freezing will result in large ice crystal formation and rapid freezing in tiny ice crystal formation. Large ice crystals tend to damage the cell walls resulting in texture lose in frozen fruits, fish, meat, etc. During freezing of foods, water is transformed to ice with high degree of purity and solid concentration in the unfrozen liquid is gradually increased. This is accompanied by changes in pH, ionic strength, viscosity, osmotic pressure, vapour pressure and other properties. These changes along with the lower temperature are responsible for decrease in microbial activity and often on destruction of micro- organisms in frozen foods.

4.4.4 Water Quality and Standards

Water is used for different purposes in food processing. They include water used as ingredient in finished products, for generating steam, for cleaning raw materials, for cleaning plant and equipment, as heat exchange medium for heating and cooling etc. The water quality for different purposes varies.

In general, only potable water should be used in the preparation of food indented for human consumption. Potable water is that water which contains no bacteria capable of causing human intestinal diseases and is aesthetically satisfactory for drinking purposes, i.e. free from undesirable odours and flavours.

Potable water should have good clarity, colourless and free from objectionable odour and taste.

Hardness of Water

Calcium and magnesium salts cause hardness of water. Permanent hardness is due to chlorides and sulphates of calcium and magnesium and temporary hardness is due to bicarbonates of these ions. Hardness is expressed as ppm (parts per million i.e. mg per litre) of $CaCO_3$ (calcium carbonate) on which basis water is classified according to degree of hardness (Table 4.4).

Table 4.4: Classification of water based on hardness

ppm of $CaCO_3$	Condition
Less than 50	Soft
50 to 100	Slightly hard
100 to 200	Hard
Above 200	Very hard

Check Your Progress Exercise 4

Note: a) Use the space below for your answer.
b) Compare your answers with those given at the end of the unit.

1. Explain the relationship between water activity and food spoilage.

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2. Why quick freezing better than slow freezing?

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3. Explain hardness of water.

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Microbiological Quality

The bacteriological quality of water used throughout the plant should meet the standards required for drinking water. The fitness of water for drinking purposes with respect to bacterial content is determined by the presence or absence of the coliform group of bacteria including Escherichia and Aerobacter species which indicate the possibility of faecal contamination. Waters drawn from deep wells or those purified by artificial means seldom show the presence of E.coli in 100 ml.

The quality of water required varies for different process food industries. The essential microbiological parameters of the BIS standards for water meant for general purposes are given in Table 4.5.

Table 4.5: Bacteriological tolerances

Sl. No.	Characteristic	Tolerance
1.	Coliform bacteria, MPN index/100 ml.	Less than 1
2.	Standard plate count, per ml., Max.	50 (Note 1)
3.	Proteolytic and lipolytic organisms, combined count per ml. Max.	5 (Note 2)

4.4.5 Chlorination

As in the case of municipal water supply, chlorination of industrial water has become a common practice in food processing plants as a means of improving plant sanitation. Gaseous chlorine or calcium and sodium hypochlorites are used for chlorination of water.

Chlorine Demand of Water

When chlorine is added to water other than distilled water, a small amount, normally 0.25 to 0.75 ppm, reacts with impurities in the water. This quantity of chlorine is called the **chlorine demand** of the water. The impurities responsible for chlorine demand include compounds containing iron, manganese, nitrites and sulphides. The chlorine, which reacts with these compounds, has no germicidal properties and cannot be measured by the methods used for testing chlorine concentration.

Break Point Chlorination

When chlorine is added to water, initially it is used up to satisfy the chlorine demand of the water. As additional chlorine is added, a free residual chlorine appears. At the same time, some chlorine loosely combines with nitrogenous matter present in water to form chloro-nitrogen compounds. The residual chlorine gradually increases until it reaches a concentration depending on the physical and chemical nature of the water, at which an oxidation reaction occurs between the free chlorine and the chloro-nitrogen compounds. The free residual nitrogen is decreased by the amount necessary to completely oxidise the chloro-nitrogen compounds. Further addition of chlorine beyond this point will result in a second rise in free chlorine concentration, which increases in almost direct proportion to the rate of chlorine application (Figure 4.5). The point after the first rise in concentration at which the free residual chlorine reaches its lowest level is known as the break point. Break point chlorination is defined as chlorination to a degree where a persisting residual chlorine of 2-10 ppm occurs. The residual chlorine in water exists either as free chlorine or chlorine which has loosely combined with other elements. The rate at which bacteria exposed to chlorine are killed is proportional to the amount of chlorine present as hypochlorous acid.

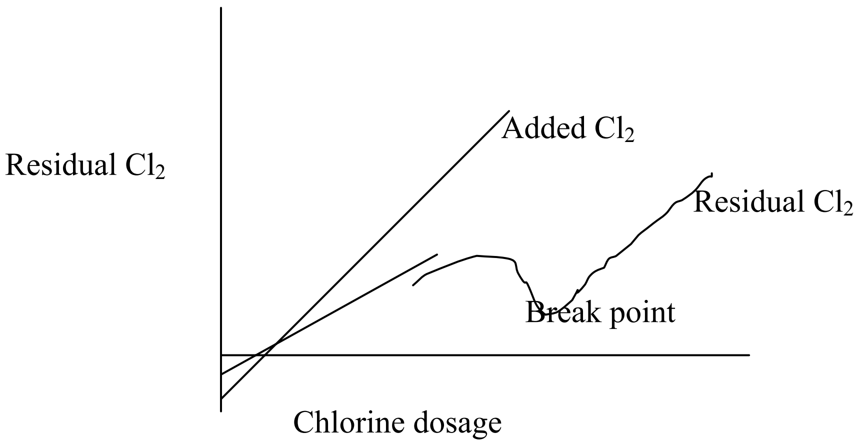

Figure 4.5: Break point chlorination of water

Chlorination of Water

In plant chlorination reduces bacterial count and clean up time and avoids odours. Chlorination of cooling water (for cooling canned products) prevents spoilage from recontamination. Residual chlorine of 5 ppm in the water is considered sufficient. Residual chlorine of 10-20 ppm is recommended for cleaning purposes. Chlorine concentration of 5 ppm has no effect on the flavour, odour or colour of canned products. For chlorination of water, hypochlorites are usually added in the form of stock solution containing 5000–10000 ppm of chlorine.

4.4.6 Packaged Drinking Water

As all of us know the quality of water supplied for drinking in many parts of our country is very poor and varies from place to place. Several diseases are spread through water. That is why today we find a booming packaged drinking water industry in the country. Water meant for producing packaged drinking water goes through a series of treatment and processes like filtration, reverse osmosis, ozonisation, etc., to obtain the required quality. PFA has laid down detailed specifications for packaged drinking water. The salient aspects of the specification are given in Table 4.6.

Table 4.6: Specifications for packaged drinking water

Sl. No.	Characteristics	Requirements
1.	Total soluble solids	Not more than 500 mg/litre
2.	PH	6.5–8.5
3.	Nitrates (as NO_3)	Not more than 45 mg/litre
4.	Nitrites (as NO_2)	Not more than 0.02 mg/litre
5.	Sulphide (as H_2S)	Not more than 0.05 mg/litre
6.	Manganese (as Mn)	Not more than 0.1 mg/litre
7.	Copper (as Cu)	Not more than 0.05 mg/litre
8.	Zinc (as Zn)	Not more than 5.0 mg/litre
9.	Fluoride (as F)	Not more than 1.0 mg/litre
10.	Barium (as Ba)	Not more than 1.0 mg/litre
11.	Nickel (as Ni)	Not more than 0.02 mg/litre
12.	Chlorides (as Cl)	Not more than 200 mg/litre
13.	Sulphate (as SO_4)	Not more than 200 mg/litre
14.	Calcium (as Ca)	Not more than 75 mg/litre
15.	Sodium (as Na)	Not more than 200 mg/litre
16.	Arsenic (as As)	Not more than 0.05 mg/litre
17.	Cadmium (as Cd)	Not more than 0.01 mg/litre
18.	Chromium (as Cr)	Not more than 0.05 mg/litre
19.	Mercury (as Hg)	Not more than 0.001 mg/litre
20.	Lead (as Pb)	Not more than 0.01 mg/litre
21.	Iron (as Fe)	Not more than 0.1 mg/litre
22.	Residual free chlorine	Not more than 0.2 mg/litre
23.	Yeast and mould counts	Absent in 250 ml
24.	E.Coli	Absent in 250 ml
25.	Coliform Bacteria	Absent in 250 ml
26.	Faecal streptococci and staphylococcus aureus	Absent in 250 ml
27.	Aerobic microbial count	Shall not exceed 100/ml.

4.4.7 Water Analysis

In the examination of water supplies, the test will depend on the purpose for which the water is used. The initial examination of water, or the testing of supplies from a new source may consist of the following:

I. Sanitary Examination

1. Physical characteristics:

   i) Colour, ii) Odour and taste, iii) Turbidity

2. Chemical characteristics:

   i) Total solids,
   ii) Organic matter,
   iii) Hardness,
   iv) Alkalinity,
   v) Acidity,

vi) pH,
vii) Nitrogen as nitrates, nitrites, free ammonia and albuminoid ammonia,
viii) Chlorides,
ix) Sulphates,
x) Free CO_2,
xi) Oxygen absorption, and
xii) Heavy metals.

II. Microbiological Examination

1. Plate count
2. Coliform count
3. Faecal streptococci test
4. Clostridium welchii test.

Check Your Progress Exercise 5

Note: a) Use the space below for your answer.
b) Compare your answers with those given at the end of the unit.

1. Which are the microorganisms or concern in potable water?

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2. Define chlorine demand and break point in chlorination.

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3. Which are the components of water analysis?

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4.5 LET US SUM UP

Proteins are macromolecules built up of amino acids. They form the building block of animal and human body structure. Pulses, oil seeds, and animal and marine foods are good sources of protein. The quality of protein greatly depends on its essential amino acid makeup.

Enzymes are biocatalysts, which catalyse biological reactions. All enzymes are proteins but all proteins are not enzymes. Several enzymes find application in food processing.

Water is an essential component of foods. Its state in foods plays an important role in food preservation. Quality of water used in food processing operations and for drinking is of paramount importance. Water is treated in several ways to make it suitable for different purposes.

4.6 KEY WORDS

Amino acids : Nitrogen containing compounds having both carboxyl and amino groups.

Essential amino acids : Amino acids which cannot be synthesised by the human body.

Albumin : Proteins, which are soluble in neutral salt free water.

Denaturation : Change in the structure of protein without breaking covalent bonds.

Immobilised enzymes : Enzymes fixed on water insoluble inert supports.

Chlorine demand : The quantity of chlorine added which reacts with the impurities in water and which does not show up as free chlorine.

4.7 ANSWERS TO CHECK YOUR PROGRESS EXERCISES

Check Your Progress Exercise 1

Your answer should include the following points:

1. Aliphatic, aromatic, etc.
2. Different classes of simple proteins
3. Conjugated proteins

Check Your Progress Exercise 2

Your answer should include the following points:

1. Coagulation of egg albumin.
2. Deficiency of amino acids like lysine.
3. Pulses, oil seeds.
4. Valine, lysine, methionene, etc.

Check Your Progress Exercise 3

Your answer should include the following points:

1. Bio catalyst.
2. All enzymes are proteins.
3. Enzymes fixed on inert support.

Check Your Progress Exercise 4

Your answer should include the following points:

1. Relationship of water activity to microbial growth.
2. Large ice crystals damage cell structures.
3. Calcium and magnesium salts.

Check Your Progress Exercise 5

Your answer should include the following points:

1. Coliforms.
2. Reaction of added chlorine with impurities in water.
3. Depression in residual chlorine content.
4. Physical, chemical, microbiological.

4.8 SOME USEFUL BOOKS

1. Owen R. Fennema, (1976) Principles of food science, Part I- Food Chemistry, Marcel Decker Inc.; New York.

2. Meyer, L.H. (1969) Food Chemistry, Van Nostrand Reinhold Company, New York, Cincinnati, Toronto, London, Melbourne.

3. Braverman, J.B.S. (1963) Introduction to the Biochemistry of foods, Elsevier Publishing Company, Amsterdam, London, New York.

4. Ranganna, S. (2000) Hand book of Analysis and Quality Control for Fruit and Vegetable Products, Tata McGraw-Hill Publishing Co. Ltd., New Delhi.