

UNIVERSITY OF SETIF 1 FERHAT
ABBAS

Faculty of Natural Science and Life

Department of Biotechnology

FOOD BIOCHEMISTRY

L3 Food biotechnology
student

Dr. BARGHOUT N.

General presentation and objectives

This course is intended for third-year undergraduate students majoring in Food Biotechnology, within the Department of Biotechnology, Faculty of Natural and Life Sciences, at Ferhat Abbas University Sétif 1. It is structured into six main chapters, each including an introductory phase, a learning process, and an assessment component through tutorials, designed to strengthen knowledge acquisition. At the end of this course, students will be able to:

- ✚ Know the different food constituents
- ✚ Understanding the importance of food constituents in terms of technological and functional properties
- ✚ To investigate the biochemical changes of the major constituents during technological processes.
- ✚ Describe the main functions of water in food systems
- ✚ Enumerate the properties of proteins in food
- ✚ Explain the chemical nature of lipids and carbohydrates in food
- ✚ Identify the components of the nutrient system
- ✚ Analyze the different mechanisms of alteration in food systems.

Semestre : 5

Unité d'enseignement Fondamentale 1

Matière 2: Biochimie alimentaire

Crédits : 6

Coefficient : 3

Objectifs de l'enseignement

L'enseignement de la biochimie alimentaire vise à décrire aux étudiants les grands constituants alimentaires, leur importance en matière de propriétés technologiques et fonctionnelles. Cette matière vise également l'initiation des étudiants aux principales évolutions (ou modifications) biochimiques des constituants majeurs en cours des procédés technologiques.

Connaissances préalables recommandées

Biochimie, chimie, physique, thermodynamique,...etc.

Contenu de la matière :

Chapitre 1 : L'eau - Généralités

1. Structure de l'eau
2. Propriété physique
3. Activité de l'eau
4. Comportement de l'eau des solutions lors de la congélation
5. Les isothermes d'adsorption
6. Phénomènes d'hystérésis des isothermes
7. Isotherme de sorption dans les I.A.A.

Chapitre 2 : Les systèmes protéiques

1. Propriétés physiques des protéines

2. Extraction des protéines alimentaires (méthodes, propriétés et utilisation des concentrations et isolats protéiques)

3. Les protéines de l'œuf : propriétés et utilisation

4. Les propriétés fonctionnelles des protéines laitières et amélioration

5. Les ingrédients protéiques

Chapitre 3 : Les lipides

1. Propriétés chimiques et physiques des lipides

2. Propriétés fonctionnelles de certains corps gras

3. Les besoins nutritionnels en corps gras

4. Conservation et altération

Chapitre 4 : Etude des polysaccharides

1. La cellulose et ses dérivés

2. L'amidon

2.1. Phénomène de gélification et rétrogradation

2.2. Comportement rhéologique

3. Propriétés fonctionnelles de l'amidon natif et amidons modifiés

4. Les enzymes amylolytiques et leur utilisation

5. Les fibres alimentaires

5.1. Cas des pectines

5.2. La gélification

Chapitre 5 : Systèmes alimentaires

1. Aspects généraux

2. Système alimentaire d'origine végétale

- 2.1. Métabolites primaires et secondaires
- 2.2. Céréales, légumineuses, fruits et légumes, algues
- 3. Système alimentaire d'origine animale
 - 3.1. Muscles
 - 3.2. Œufs
 - 3.3. Lait
- 4 Système alimentaire non conventionnelle (P.A. I.)
 - 4.1. Protéines
 - 4.2. Lipides,
 - 4.3. Biomasse

Chapitre 6 : Altérations alimentaires

- 1. Rôle de l'eau
- 2. Sources potentielles d'altérations
- 3. Altérations microbiologiques, enzymatiques et chimiques

Mode d'évaluation : (type d'évaluation et pondération)

Compte rendu et Examen semestriel.

Références bibliographiques (Livres et photocopiés, sites internet, etc) :

Citer au moins 3 à 4 références classiques et importantes.

Weinman, S., & Méhul, P. (2004). Toute la biochimie. Dunod.

Boeckel, T. P. V., Hounhouigan, J. D., & Nout, R. (2003). Les aliments: transformation, conservation et qualité. Technical Centre for Agricultural and Rural Cooperation.

Prerequisite

- ❖ Chemistry
- ❖ Biochemistry
- ❖ Structure and Types of Carbohydrates
- ❖ Structure and Types of Lipids
- ❖ Cellular Biology

Pretest

1. What structure types of sugars, fatty acids, and proteins do you know?
2. What is the difference between levorotatory and dextrorotatory in sugar?
3. What is the structure and the nature of water?

Table of Contents

General presentation and objectives.....	1
Prerequisite.....	5
Pretest.....	5
Chapter 1: Water.....	10
1.1 Structure of Water.....	11
1.2 Classification of water in food.....	12
1.2.1 Free water: The available water.....	12
1.2.2 Bound water: The tightly held moisture.....	13
1.2.3 Imbibed water: The absorbed moisture.....	14
1.3 Physical Properties.....	16
1.4 Water activity (aw).....	17
1.5 Water behavior in solutions during freezing.....	19
1.6 Relationship between Moisture Content and Water Activity.....	22
1.7 Sorption Isotherms.....	22
1.7.1 Types of Sorption Isotherms.....	23
1.8 Hysteresis Phenomena of Isotherms.....	24
1.9 Sorption isotherm in the Food Industry.....	26
Chapter 2: Protein systems.....	28
1.1 Protein Sources.....	28
1.1.1 Essential amino acids: There are nine, namely:.....	28
1.2 Physical Properties of Proteins.....	29
1.3 Functional properties of food proteins.....	31
1.4 Extraction of protein.....	31

1.4.1	Enzymatic methods	31
1.4.2	Physical methods.....	34
1.4.3	Chemical methods	37
1.5	Egg proteins	38
1.5.1	Main composition.....	39
1.5.2	Distribution and general characteristics	39
1.5.3	Digestibility of egg white proteins	40
1.6	Functional properties of milk proteins and improvement	40
1.6.1	General composition of milk proteins	40
1.6.2	Importance of milk proteins	41
1.6.3	Whey proteins	42
1.6.4	Biological activity and bioactive peptides proteins and bioactive fractions	42
1.6.5	Industrial applications	43
Chapter 3 : Lipids		45
1.1	Definitions	45
1.2	General classification	45
1.3	Lipid content in foods.....	46
1.4	Chemical and physical properties of lipids.....	46
1.4.1	Physical properties	46
1.4.2	Chemical properties.....	47
1.5	Functional properties of fats	49
1.6	Nutritional requirements for fats	50
1.7	Preservation and alteration of lipids	51
1.7.1	Factors influencing lipid stability against oxidation	51
Chapter 4: Study of Polysaccharides.....		53

1.1	Cellulose and its derivatives	53
1.2	Starch	54
1.2.1	Gelatinization and retrogradation phenomena	54
1.3	Functional properties of native and modified starches	55
1.4	Amylolytic enzymes	56
1.5	Dietary fiber: soluble fiber, pectins, and gelation	56
1.6	Case of pectins	57
Chapter 5: Food system		58
1.1	General aspect	58
1.2	Plant-based food systems	59
1.2.1	Primary and Secondary Metabolites	59
1.2.2	Cereals, legumes, fruits and vegetables, algae	62
1.3	Animal-Based Food Systems	68
1.3.1	Muscle	68
1.3.2	Eggs	69
1.3.3	Milk	71
1.4	Unconventional Food Systems	72
1.4.1	Alternative Protein Sources	72
1.4.2	Unconventional Lipid Sources	72
1.4.3	Biomass	73
Chapter 6: Food Spoilage		74
1.1	Role of water	74
1.2	Main Food Preservation Technologies	74
1.2.1	Heat preservation (thermal processes)	74
1.2.2	Low-temperature preservation	75

1.2.3	Reduction of water activity	75
1.2.4	Ionizing radiation	75
1.2.5	Chemical preservation.....	75
1.2.6	Packaging	75
1.3	Types of Microbial, Enzymatic, and Chemical Alterations	75
1.3.1	Microbial Alterations	75
1.3.2	Sensory Consequences	76
1.3.3	Enzymatic alterations in food spoilage	76
1.3.4	Chemical Deterioration in Food.....	77
References	79

Chapter 1: Water

Water is essential for basic survival, as it accounts for up to 60% of the adult human body. It is the most abundant constituent in most foods. Indicative values of water content in a number of food products are shown in Table 1

Classification of foods into three groups, according to their water content (high, intermediate, and low moisture foods), has been suggested.

- Fruits, vegetables, juices, raw meat, fish, and milk belong to the **high moisture category**.
- Bread, hard cheeses, and sausages are examples of **intermediate moisture foods**
- While the **low moisture group** includes dehydrated vegetables, grains, milk powder, and dry soup mixtures (Table 1).

Table 1: Water content of different types of food.

Food	Water (%)
Cucumbers	95–96
Tomatoes	93–95
Orange juice	86–88
Apples	85–87
Cow milk	86–87
Eggs, whole	74
Chicken, broiled	68–72
Hard cheese	30–50
White bread	34
Jams, preserves	30–35
Honey	15–23
Wheat	10–13
Nuts	4–7
Dehydrated onion	4–5
Milk powder	3–4

1.1 Structure of Water

- Water (H_2O) (Figure 1) is a polar molecule composed of two hydrogen atoms covalently bonded to one oxygen atom.
- The difference in electronegativity between the oxygen and hydrogens causes the oxygen to have a partial negative charge and the hydrogen to have a partial positive charge. This difference in charge causes polarity.
- The partial positive charge of the hydrogen of one water molecule attracts the partial negative charge of the oxygen of another water molecule. This attraction is called **hydrogen bonding**.
- Hydrogen bonding is weaker than the covalent bonds between the oxygen and hydrogens of the same molecule but causes many of water's unique properties. For example, more energy is required to break hydrogen bonds, so water has a higher melting and boiling point.

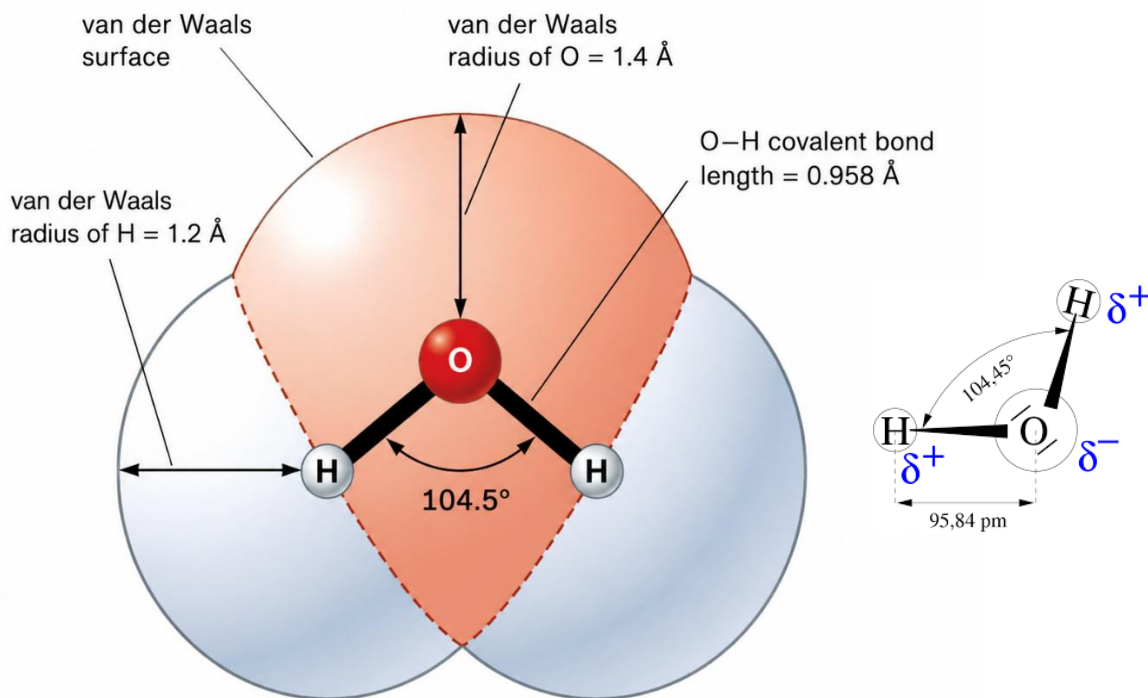


Figure 1: Structure of water.

1.2 Classification of water in food

Food scientists classify water in foods into three main categories: free water, bound water, and imbibed water. Each type behaves differently and serves unique functions within food systems.

1.2.1 Free water: The available water

Free water, also called available water, is the most abundant form of water in most foods. This is water that moves freely within the food matrix and isn't attached to any food components. Think of the water that drips from a freshly cut watermelon or the moisture that accumulates when you thaw frozen vegetables—that's predominantly free water.

Free water functions as a solvent for various nutrients, facilitating biochemical reactions essential for flavor development. However, it also has a downside: it provides the perfect environment for microbial growth, making foods with high free water content more susceptible to spoilage.

Free water has properties similar to pure water: it freezes at approximately 0°C, evaporates readily during drying, and can be easily removed through physical methods like pressing or draining. In food science, this type of water is often associated with a food's water activity (aw), a critical parameter used to predict food stability and safety.



Figure 2: role of water in food.

1.2.2 Bound water: The tightly held moisture

Bound water (Figure 3) is chemically or physically attached to food components like proteins, carbohydrates, and minerals through hydrogen bonding or other molecular interactions. This type of water is not available for microbial use and behaves quite differently from free water.

Bound water doesn't freeze at 0°C, often requiring temperatures as low as -40°C to solidify. It's also resistant to evaporation during conventional drying processes, which explains why even completely dried foods still contain some moisture—typically about 5-10% by weight.

The strong attraction between bound water and food molecules means this water cannot serve as a solvent for biochemical reactions or support microbial growth. Foods with higher proportions of bound water tend to have longer shelf lives even without preservatives.

Common examples include:

- **Dried legumes:** Even when completely dry to the touch, beans and lentils retain bound water molecules attached to their starch and protein components.
- **Wheat flour:** Contains about 14% moisture, much of which is bound water associated with starch granules and gluten proteins.
- **Hard cheese:** Despite their firm texture, aged cheeses like Parmesan still contain bound water molecules integrated with the protein matrix.

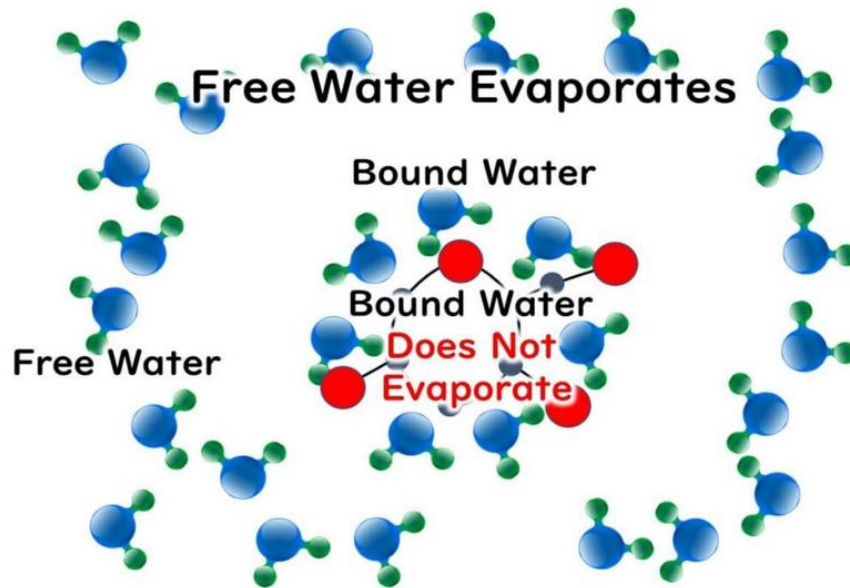


Figure 3: free and bound water

1.2.3 Imbibed water: The absorbed moisture

Imbibed water represents an intermediate state between free and bound water. This type of water is physically trapped or absorbed by hydrophilic (water-loving) substances but isn't chemically bound to them. The classic example is gelatin soaking up water to form a gel, or rice grains absorbing water during cooking.

Hydrocolloids (Figure 4,5) like pectin, agar, and starches have an exceptional capacity to imbibe water, which explains their widespread use as thickening and gelling agents in food processing. When these substances absorb water, they swell significantly while maintaining their structural integrity, creating the characteristic texture of products like jams, puddings, and gravy.

Imbided water behaves differently from both free and bound water: it doesn't flow freely but isn't firmly attached to food molecules either. It freezes at temperatures below 0°C but above the freezing point of bound water. Microorganisms can sometimes access imbided water, though not as readily as free water.

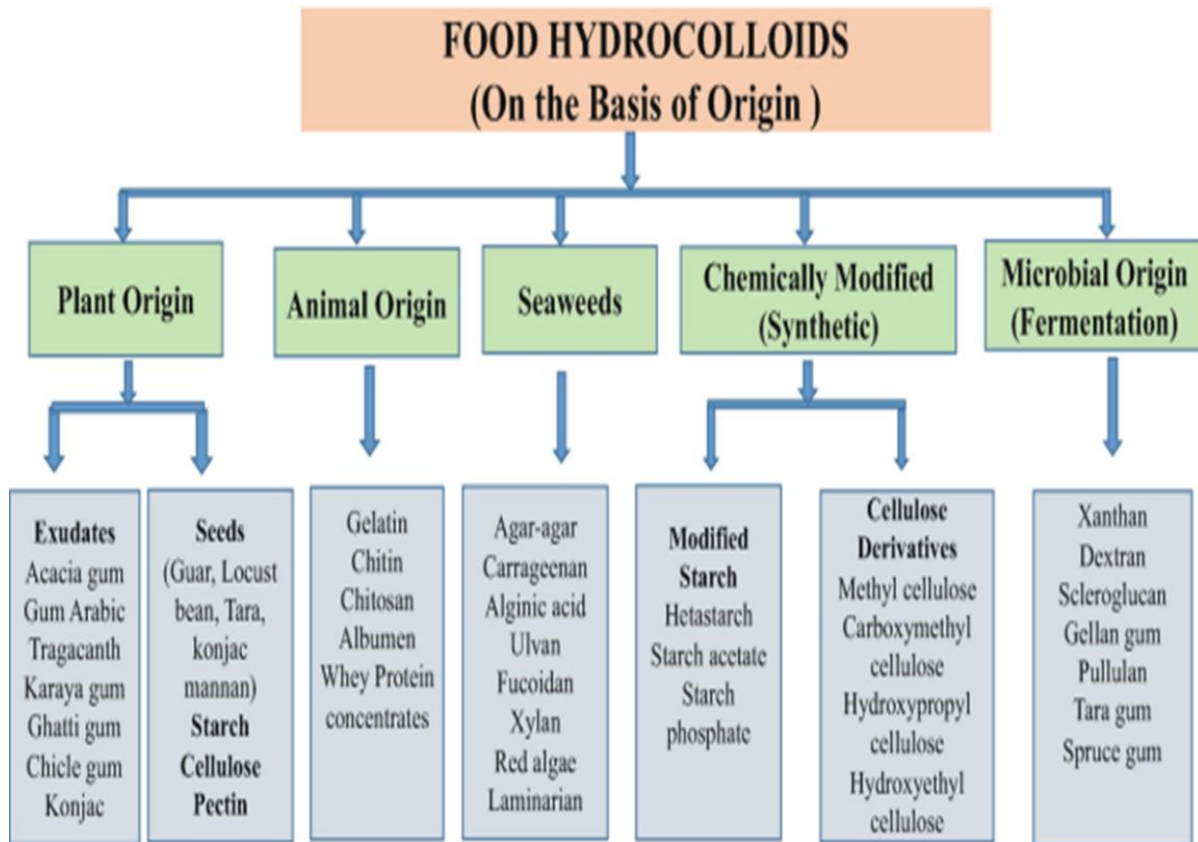


Figure 4: Origin and types of hydrocolloids

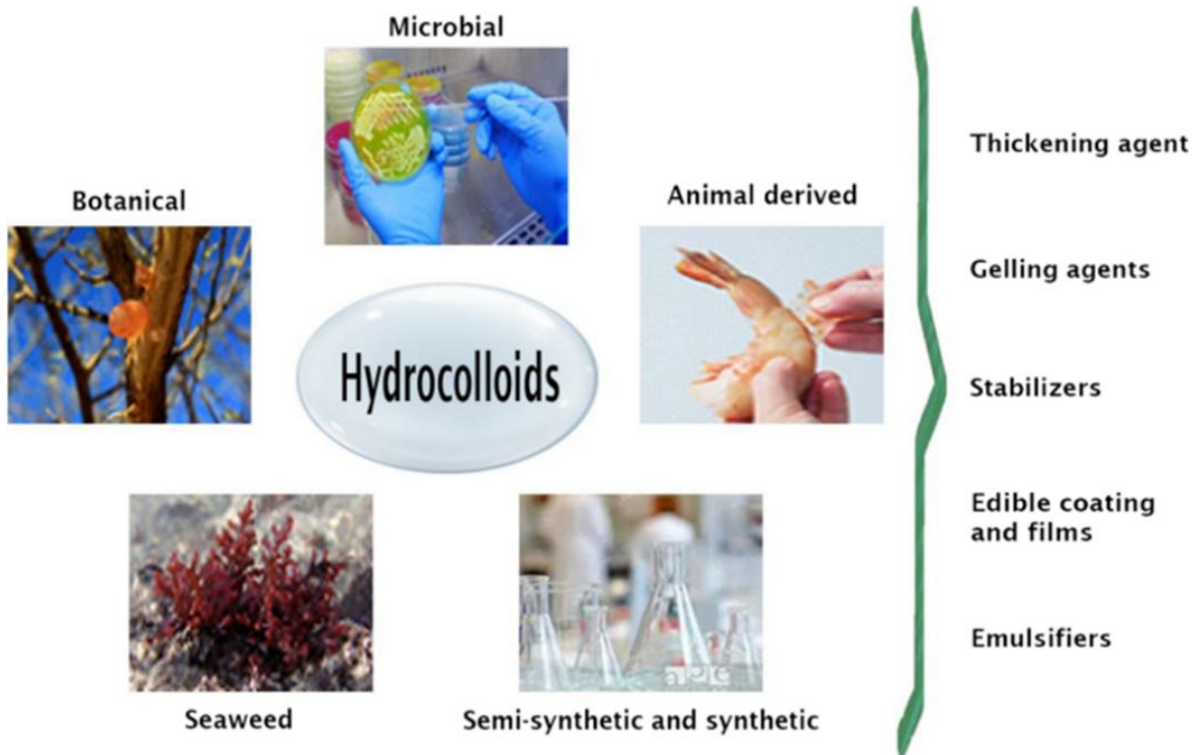


Figure 5: Hydrocolloids role and origin

1.3 Physical Properties

- **Melting and boiling points:** (figure 6) Water has a melting point of 0 °C and a boiling point of 100 °C at normal atmospheric pressure.
- **High specific heat capacity:** Water can absorb a large amount of heat before undergoing a significant change in temperature.
- **Latent heat of vaporization:** A considerable amount of energy is required to convert liquid water into vapor.
- **Density:** Water reaches its maximum density at 4 °C, which explains why ice floats on liquid water.
- **Surface tension:** Due to hydrogen bonding, water exhibits high surface tension, which plays an important role in capillarity within biological systems.

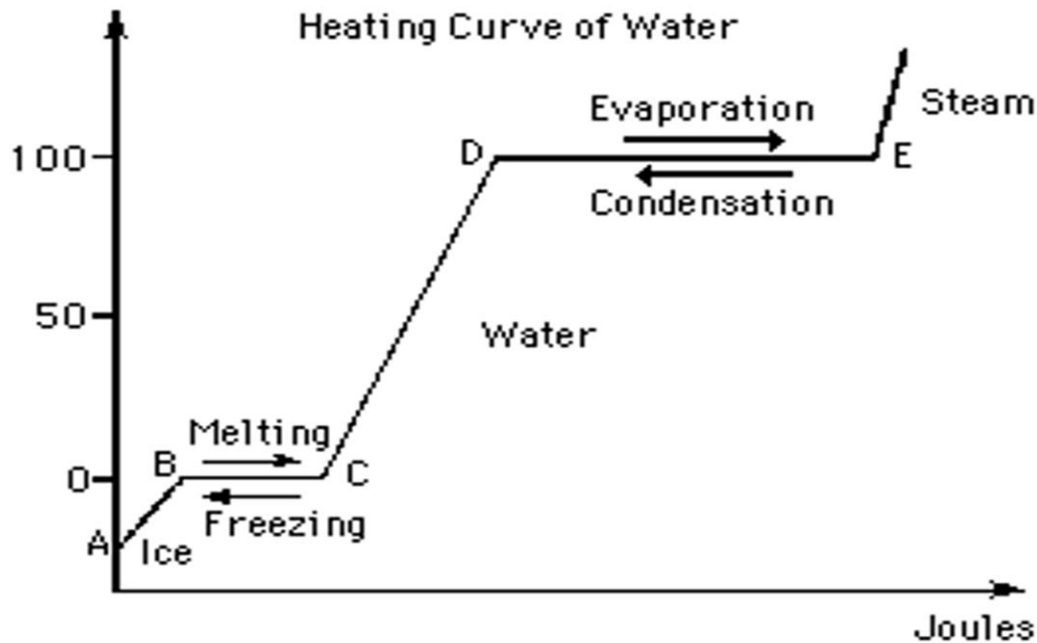


Figure 6: heating curve of water

1.4 Water activity (aw)

It is now well established that the effect of water on the stability of foods cannot be related solely to the quantitative water content.

- As an example, honey containing 23% water is perfectly shelf-stable while dehydrated potato would undergo rapid spoilage at a moisture content half as high. To explain the influence of water, a parameter that reflects both the quantity and the “effectiveness” of water is needed. This parameter is *water activity*.
- **Definition of water activity**

Water activity, a_w , is defined as the ratio of the water vapor pressure of the food to the vapor pressure of pure water at the same temperature.

$$\text{Formula : } a_w = \frac{P}{P_0} = \frac{HRE\%}{100}$$

where:

p = partial pressure of water vapor of the food at temperature T .

p_0 = equilibrium vapor pressure of pure water at temperature T .

The same type of ratio also defines the relative humidity of air, RH (usually expressed as a percentage):

$$\text{Formula : } RH = \frac{P'}{P_0} \times 100$$

where:

p' = partial pressure of water vapor in air.

If the food is in equilibrium with air, then $p = p'$. It follows that the water activity of the food is equal to the relative humidity of the atmosphere in equilibrium with the food. For this reason, water activity is sometimes expressed as the equilibrium relative humidity (Figure 7), ERH.

$$\text{Formula : } aw = \frac{P}{P_0} = \frac{HRE\%}{100}$$

The equilibrium relative humidity of a product, which is determined by its **partial water vapor pressure at the surface**, depends on the following factors: chemical composition; temperature; moisture content; storage environment; absolute pressure; and packaging.

- **Importance in food science:** A low water activity (aw) prevents microbial proliferation, which is essential for food preservation (table 2).

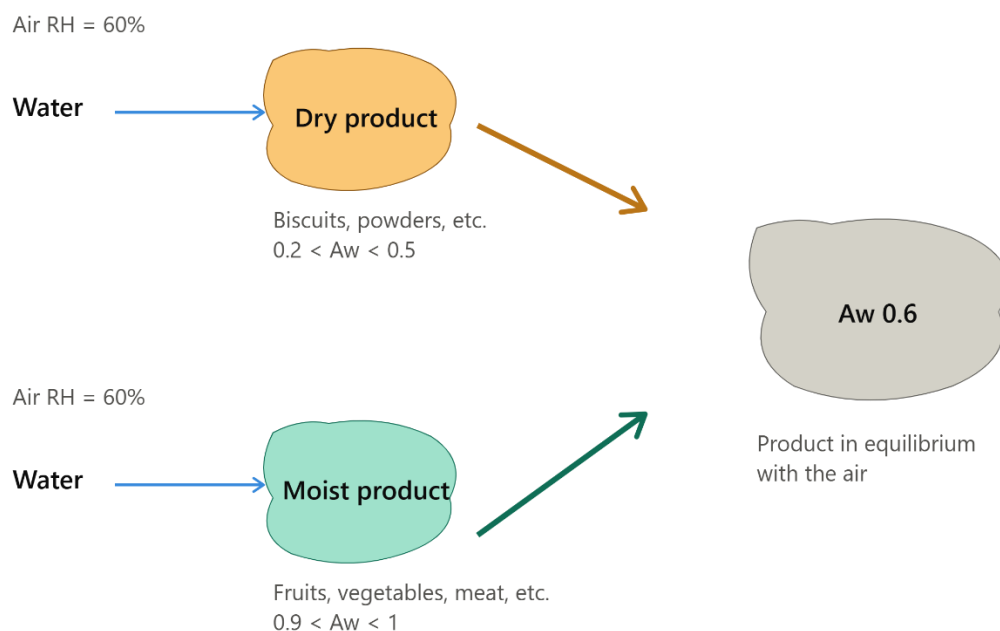


Figure 7: Relationship between water activity and the equilibrium relative humidity

Table 2: Typical water activities of selected foods

a_w range	Product examples
0.95 and above	Fresh fruits and vegetables, milk, meat, and fish
0.90–0.95	Semihard cheeses, salted fish, and bread
0.85–0.90	Hard cheese, sausage, and butter
0.80–0.85	Concentrated fruit juices, jelly, and moist pet food
0.70–0.80	Jams and preserves, prunes, dry cheeses, and legumes
0.50–0.70	Raisins, honey, and grains
0.40–0.50	Almonds
0.20–0.40	Nonfat milk powder
Less than 0.2	Crackers, roasted ground coffee, and sugar

1.5 Water behavior in solutions during freezing

- Freezing of pure water: At 0 °C, water crystallizes into ice.
- Aqueous solutions: Solutes lower the freezing point. As the solution freezes, water molecules crystallize first, thereby increasing the concentration of solutes in the remaining liquid phase.
- Supercooling phenomenon: Water can sometimes remain liquid below 0 °C before suddenly crystallizing.

Freezing of pure water: At 0 °C, water crystallizes into ice.

Depending on the pressure and temperature conditions, ice can exist in several crystalline forms (figure 8). At atmospheric pressure, ice type I has a hexagonal structure (one oxygen atom at each vertex). The O–H bonds form an angle of 109° and have a length ranging from 0.97 to 1.01 Å. Consequences: The crystalline structure is “highly open,” occupying a larger volume than liquid

water (1 L of water \rightarrow 1.098 L of ice at 0 °C). Consequently, ice is a solid with a lower density (0.92) than its liquid form (1).

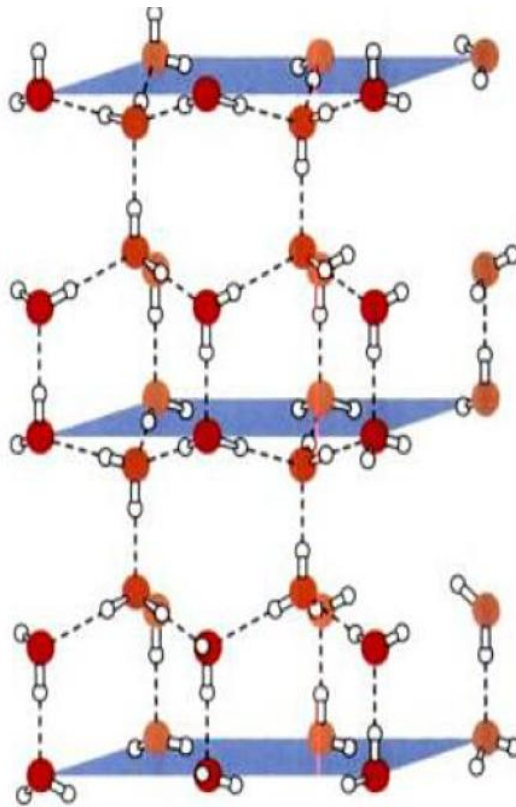


Figure 8: Ice structure

Freezing extends the storage life of foods by rendering them more inert and slowing down adverse reactions that promote food spoilage and limit shelf life. However, a number of physical and biochemical reactions can still occur, many of which are exacerbated when recommended handling, production, and storage practices are not followed. To ensure the quality and safety of frozen foods, temperature requirements are established for each critical step of the cold chain. It is recommended that stable food temperatures be maintained at $-18\text{ }^{\circ}\text{C}$ or below, although brief deviations are permitted during transport or local distribution, where $-15\text{ }^{\circ}\text{C}$ is acceptable. Retail display cabinets must be kept at $-18\text{ }^{\circ}\text{C}$ to ensure continuity with storage conditions, and not higher than $-12\text{ }^{\circ}\text{C}$.

Freezing can preserve the taste, texture, and nutritional value of foods better than most other long-term preservation methods. However, these qualities depend on the choice of raw materials, the application of appropriate pretreatments, the type of freezer and cold storage options used, as well

as the use of suitable packaging Freezing damage occurs as a result of several mechanisms, leading to quality loss in a product after thawing.

This quality deterioration may be directly observed (e.g., freezer burn, discoloration, and mechanical damage), but in many cases it does not become apparent until after thawing and cooking.

Most deterioration mechanisms are governed by the storage temperature, particularly the duration spent above its recommended value.

They are also exacerbated by temperature fluctuations. Ice and water can damage the food matrix in several ways, such as:

1. **Unfrozen water.** Even below $-18\text{ }^{\circ}\text{C}$, up to 10% of the water may remain unfrozen and participate in physical and biochemical reactions.
2. **Freezing damage.** The volumetric expansion of water as it crystallizes into ice can induce structural damage to the food. This often leads to the formation of large voids and excessive drip loss in frozen materials after thawing (Figure 9).

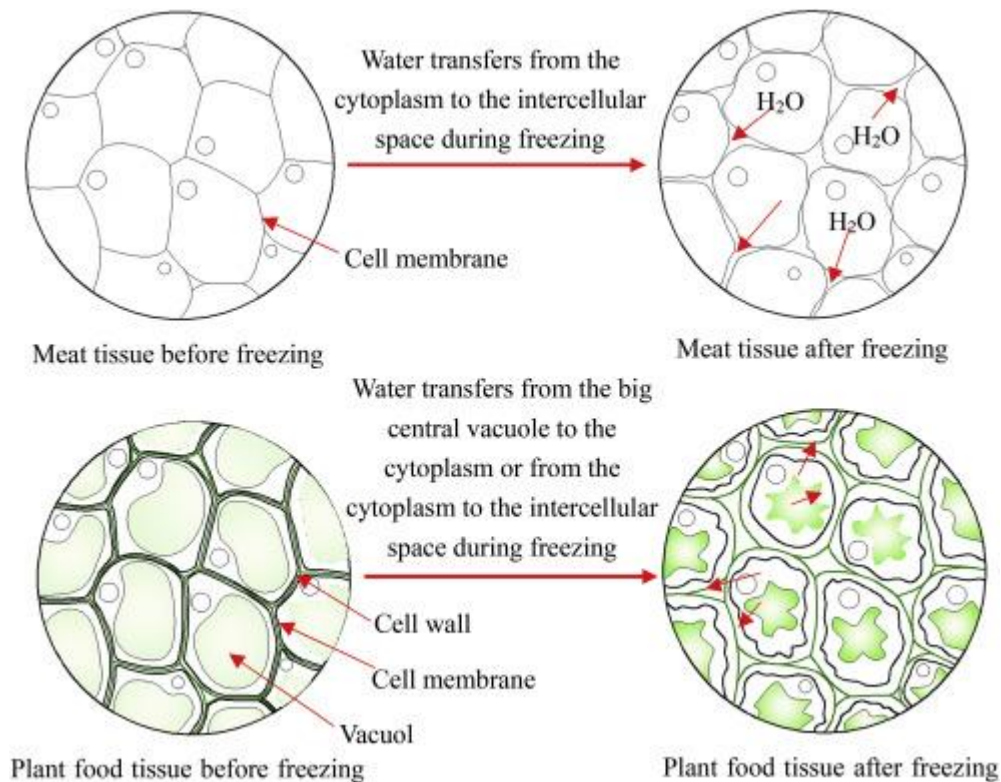


Figure 9: Behavior of water during freezing

1.6 Relationship between Moisture Content and Water Activity

The relationship between moisture content and water activity is represented by a curve known as the **sorption isotherm**. For each value of a_w , the isotherm provides the equilibrium moisture content (X_{eq}) of the product at a given temperature (figure 10).

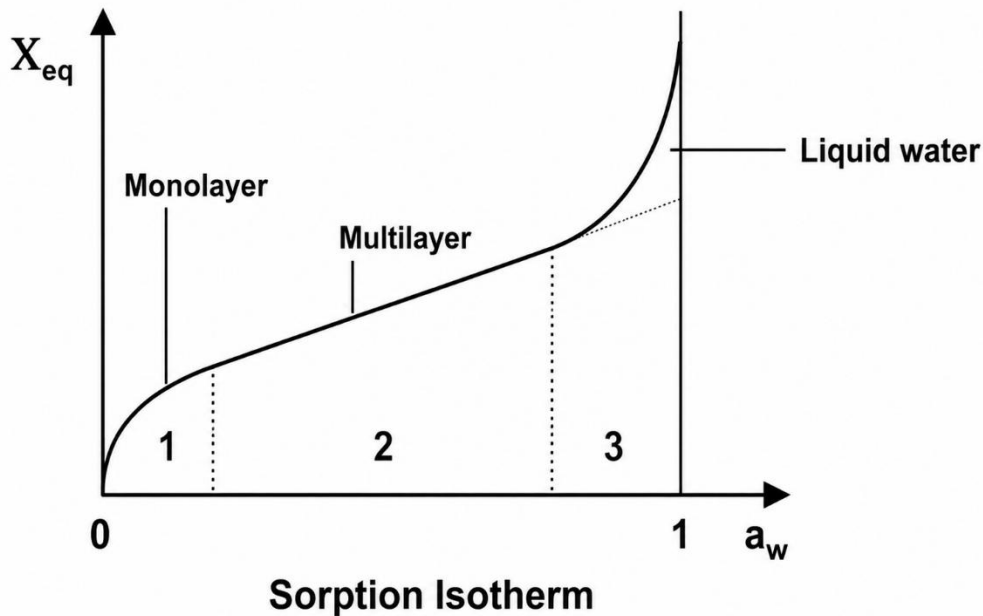


Figure 10: The sorption isotherm phenomena.

1.7 Sorption Isotherms

The **sorption isotherm** represents, at equilibrium and at a given temperature, the amount of water contained in a food as a function of its water activity (a_w) or the relative humidity of the air in equilibrium with the product.

A **sorption isotherm** describes the thermodynamic relationship between water activity and the equilibrium moisture content of a food product at constant temperature and pressure.

The knowledge and understanding of sorption isotherms are of great importance in food science and technology for the design and optimization of drying equipment, packaging design, quality and stability prediction, shelf-life determination, and for calculating moisture changes that may occur during storage.

The sorption isotherms are divided into three zones:

- Zone 1 ($a_w < 0.3$): **Strongly bound or constitutional water** (~3–10% water content in the product). This water is tightly associated with biochemical components through covalent bonds. It is **unavailable as a solvent or reactant**, corresponding to **the first (monolayer) layer surrounding the dry matter of the food**.
- Zone 2 ($0.3 < a_w < 0.7$): **Weakly bound water**, present in the form of polymolecular (multilayer) films partially covering the surface of the dry substrate. It is **moderately reactive and available both as a solvent and as a reactant**.
- Zone 3 ($a_w > 0.7$): **Free or bulk water**, which is held on the surface of the dry substrate only by hydrogen bonds. It is **readily available as both a solvent and a reactant**. This is the only form of water that can be utilized by microorganisms and that allows enzymatic reactions to occur.

1.7.1 Types of Sorption Isotherms

Two types of sorption isotherms are distinguished (figure 11):

- ❖ **Adsorption isotherm:** determined experimentally by starting from a dry product and progressively adding water.
- ❖ **Desorption isotherm:** determined experimentally by starting from a water-saturated product undergoing dehydration.

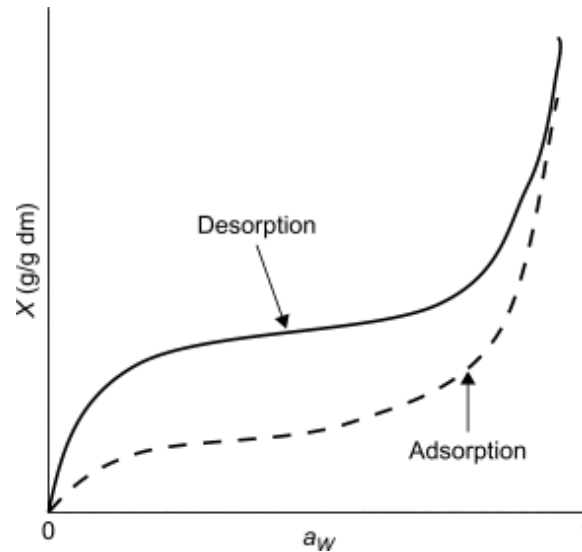


Figure 11: Types of Sorption Isotherms

1.8 Hysteresis Phenomena of Isotherms

The **desorption isotherm** does not coincide with the **adsorption isotherm** (figure 11).

- This phenomenon is known as **hysteresis** (figure 12). It is believed to be due to the condensation of water within the pores of the material. There exists a relationship between the partial pressure, the contact angle, and the pore diameter (as described by the Kelvin equation (eq)).

$$\ln \left(\frac{p}{p_0} \right) = - \frac{2 \gamma V_m \cos \theta}{r R T}$$

where:

- p = equilibrium vapor pressure above the curved liquid surface
- p_0 = saturation vapor pressure above a flat surface (pure water at the same temperature)
- γ = surface tension of the liquid ($\text{N}\cdot\text{m}^{-1}$)
- V_m = molar volume of the liquid ($\text{m}^3\cdot\text{mol}^{-1}$)
- θ = contact angle between the liquid and solid surface
- r = radius of curvature of the meniscus (m) or pore radius
- R = universal gas constant ($8.314 \text{ J}\cdot\text{mol}^{-1}\cdot\text{K}^{-1}$)
- T = absolute temperature (K)

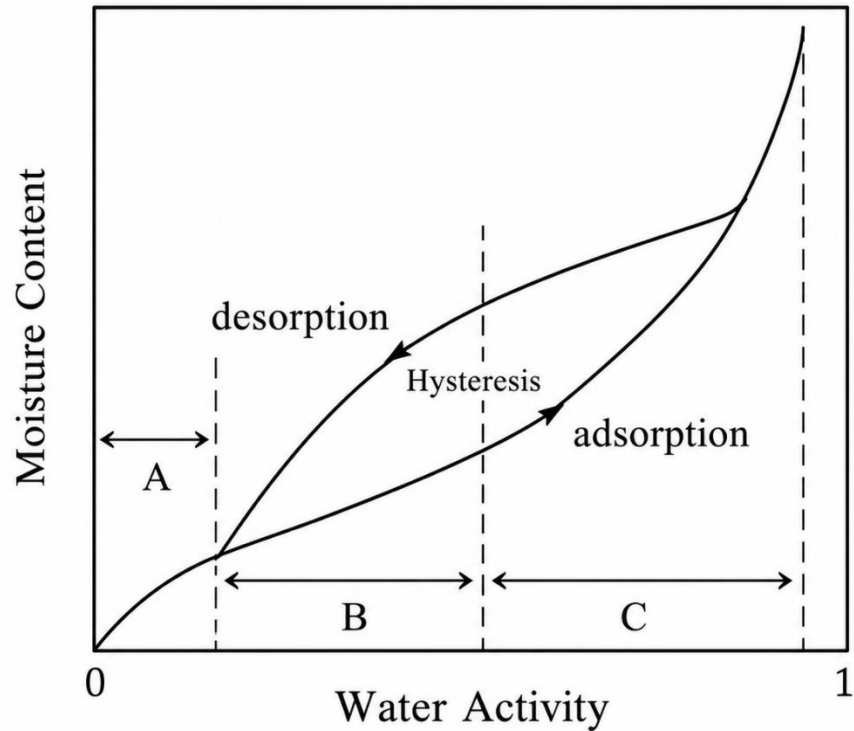


Figure 12: Hysteresis Phenomena of Isotherms

The **desorption isotherm** does not coincide with the **adsorption isotherm** (figure 13).

- This phenomenon is known as **hysteresis**. It is believed to be due to the condensation of water within the pores of the material.
- ✓ The **liquid–solid contact angle** is greater when the liquid wets a dry surface (adsorption) than when it recedes from a wet surface (desorption).
- ✓ Moreover, **the pore diameter** is often smaller at the pore entrance than deeper inside, which means that a higher partial pressure is required for pore filling.

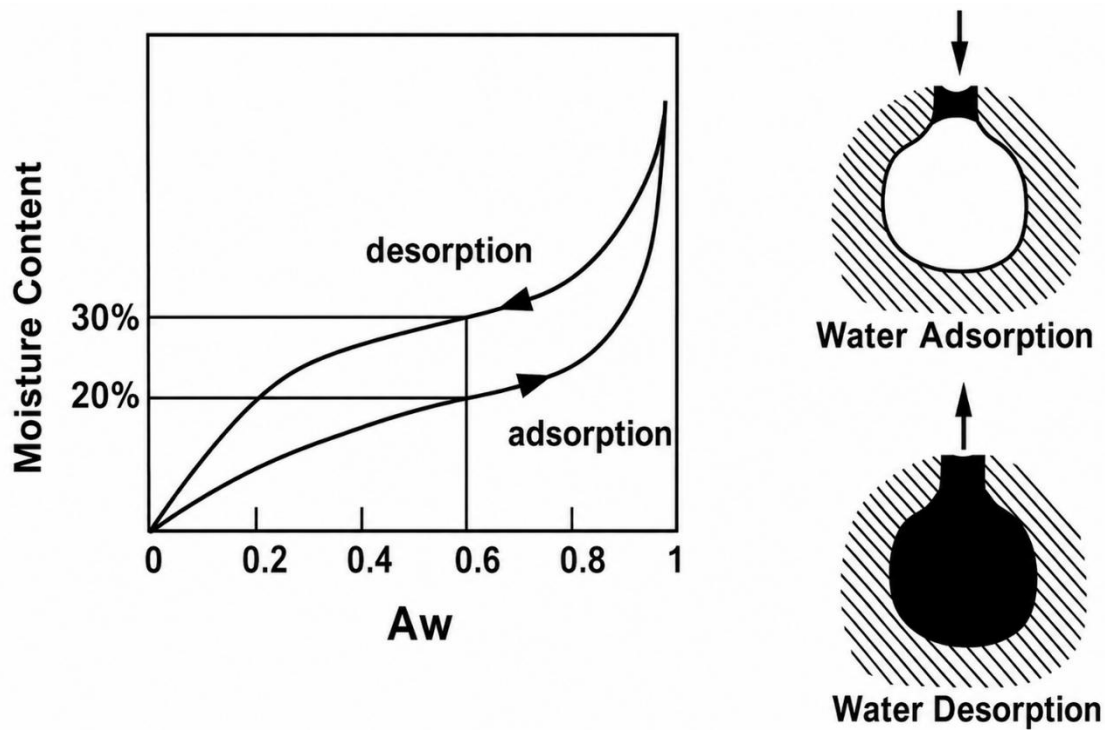


Figure 13: Hysteresis Phenomena of Isotherms

1.9 Sorption isotherm in the Food Industry

Sorption isotherms are used to predict and understand the behavior of a food product during storage. The first piece of information provided by the sorption isotherm is the **hygroscopicity** of the food. This hygroscopicity reflects the influence that a change in the surrounding relative humidity will have on the moisture content of the product when it is not protected by a moisture-proof (impermeable) packaging.

- ↪ **Water binding and the shape of sorption isotherms vary considerably depending on the nature of the chemical constituents.** *Example:* Starches and proteins retain more water in the lower region of the isotherms than sugars and lipids.
- ↪ **pH and ionic strength modify water retention due to electrostatic charges.** *Example:* The aggregation of protein chains within a gel expels both free and adsorbed water, which then flows out or evaporates—resulting in minimal water retention at the isoelectric point (pHi).
- ↪ **The physical state (crystalline, amorphous, etc.) is greatly influenced by processing, and depending on its nature, the isotherm may change.** *Example:* Heating starch

converts it from a crystalline, water-impermeable structure to an amorphous—i.e., gelatinized—state capable of retaining water.

Chapter 2: Protein systems

1.1 Protein Sources

- Red meat
- Poultry
- Fish
- Eggs
- Milk and dairy products
- Soy
- Legumes
- Seeds
- Plant-based foods
- Microorganisms
- **Comparative Bioavailability**
- ✓ **Animal proteins:** Generally more digestible and bioavailable.
- ✓ **Plant-based proteins:** Generally less digestible and have lower absorption rates.
- **Note:** If the protein intake is varied and sufficient, these differences are not significant from a nutritional standpoint (table 3).

Table 3: Protein content in food

food	Protein content
Fruits	< 1 %
Potato	2 %
Cow's milk	3,5 %
Seed	7 – 13 %
Hen's egg	12 %
Meat and fish	20 – 22 %
Cheese	24 %
Soy	40 %

1.1.1 Essential amino acids: There are nine, namely:

- Histidine

- Threonine
- Valine
- Tryptophan
- Leucine and Isoleucine
- Lysine
- Phenylalanine
- Methionine

Complete proteins contain all essential amino acids in sufficient quantities; this is the case for animal proteins and soy.

Incomplete proteins are deficient in one or more essential amino acids; this is typical of plant proteins (with the exception of soy).

1.2 Physical Properties of Proteins

- ✓ **Color and Taste:** Proteins are homogeneous and crystalline. It is observed that, regardless of their origin, proteins influence the **color, flavor, and texture** of a product.

These organoleptic properties are a determining factor in **consumer choice** and are, as such, **extensively considered by food manufacturers**.

- ✓ **Optical Activity:** All protein solutions rotate the plane of polarized light to the left, meaning they possess the L-configuration.

For instance, the specific rotation $[\alpha]_D$ for ovalbumin is close to -30° over the pH range of 3.5 to 11. However, at pH values outside this range, the rotation becomes more negative; for example, at pH 13, the $[\alpha]_D$ is approximately -60° .

The magnitude of this levorotation is further increased by subjecting the proteins to elevated temperatures.

- ✓ **Protein solubility** is strongly influenced by pH. Solubility is minimal at the isoelectric point and increases with increasing acidity or alkalinity.
- ✓ **Water Retention** According to their molecular skeletons and the charged side chains they contain, proteins can adsorb varying amounts of water. **This property depends on several factors, such as pH, salts, temperature, and other molecules.**

Thus, for a dry protein up to the point of swelling, the processes involved are **absorption, wettability, and water-holding capacity, cohesiveness, and adhesiveness**.

From swelling to dispersion or solubilization, the key properties brought into play are those of viscosity.

✓ **Colloidal Nature:** Due to their giant size, proteins exhibit numerous colloidal properties, such as:

- Their diffusion rates are extremely slow.
- They can produce significant light scattering in solution, resulting in visible turbidity (Tyndall Effect).

✓ **Denaturation:** Denaturation refers to changes in the properties of a protein. In other words, it is the loss of biological activity (figure 14).

*In many cases, the denaturation process is followed by a coagulation - a process where denatured protein molecules have tendency to form large aggregates and precipitate from solution.

*Denaturation can be caused by a variety of agents, both physical and chemical

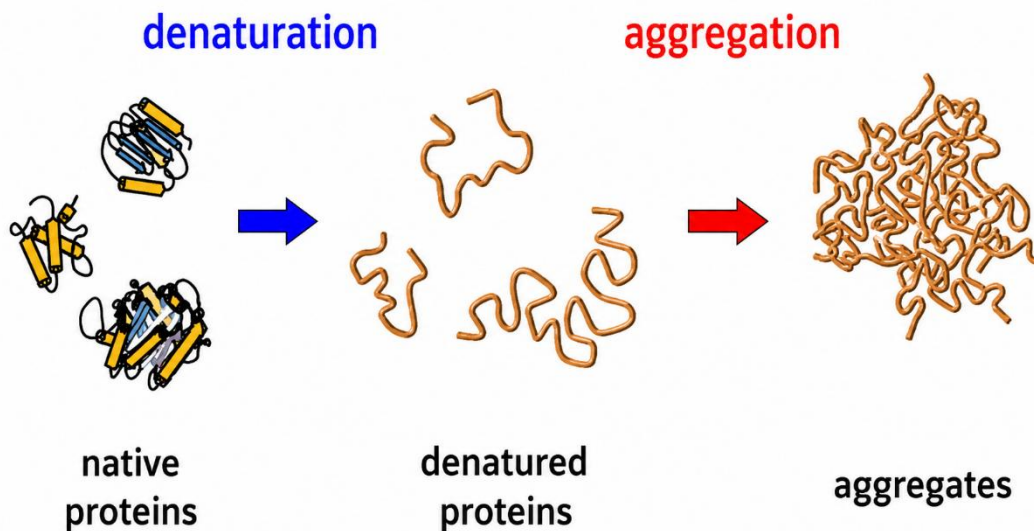


Figure 14: Denaturation of protein.

❖ **Chemical denaturation:**

- organic solvents (acetone, alcohol),
- aromatic anions (salicylates), certain anionic detergents (such as sodium dodecyl sulfate),

A common example of a protein easily denatured by solvent is milk casein.

❖ **Physical denaturation:**

- Mechanical actions (such as shaking),
- Thermal treatments,
- Cooling and freezing operations,
- Friction,
- High hydrostatic pressures (5,000 to 10,000 atm),
- Ultraviolet rays and ionizing radiation (such as X-rays, radioactive radiation, and ultrasonic waves).

A common example of a protein that is easily denatured by agitation or heat is egg white albumin.

1.3 Functional properties of food proteins

All functional properties of food proteins are summarized in the table 4

Table 4: Properties of proteins in food industries.

Category	Properties
Sensory properties	Flavor, odor, texture, color
Hydration properties	Solubility, dispersibility, gelation, viscosity
Textural properties	Viscosity, adhesion, aggregation, gelation
Other properties	Compatibility with other ingredients and processing conditions

1.4 Extraction of protein

For protein extraction, three types of techniques can be exploited, namely:

- Enzymatic methods
- Physical methods
- Chemical methods

1.4.1 Enzymatic methods

Enzymes are globular proteins from microorganisms, of plants, animals and humans, functioning as a catalyst. In dairy products (cheese, yogurt), bakery (production of bread) and meat processing,

enzymes are increasingly used. A range of food grade enzymes is also available in trade. Enzymes commonly used in industry at the moment current include **carbohydrase, lipase and mainly proteases.**

- **Advantages:** Replaces difficult chemical or physical conditions, preserves sensitive amino acids like glutamine and asparagine, and improves the extraction of food-grade proteins.

1.4.1.1 Types of enzymes used

- ↪ **Proteases** : Enzymes that catalyze the hydrolysis of proteins into smaller peptides and amino acids. They are the most widely used enzymes in protein extraction processes.
- ↪ **Carbohydrases:** Enzymes that catalyze the degradation of polysaccharides, thereby enhancing the release of proteins entrapped within carbohydrate matrices (e.g., pectin, starch, and cellulose).
- ↪ **Lipases:** Enzymes that catalyze the hydrolysis of lipids, facilitating protein extraction in systems where proteins are associated with fats.

1.4.1.2 Enzyme-assisted extraction process

Key steps:

- ✓ Cell rupture : Cell lysis releases proteins from the internal cellular compartments.
- ✓ Enzymatic hydrolysis : Allows complex proteins to be transformed into peptides or more soluble isolates (figure 15).

Extraction conditions :

- ✓ Essential parameters (pH, temperature, duration, enzyme/substrate ratio) to adjust according to the enzyme used.
- ✓ Importance of optimizing conditions to maximize yield extraction.

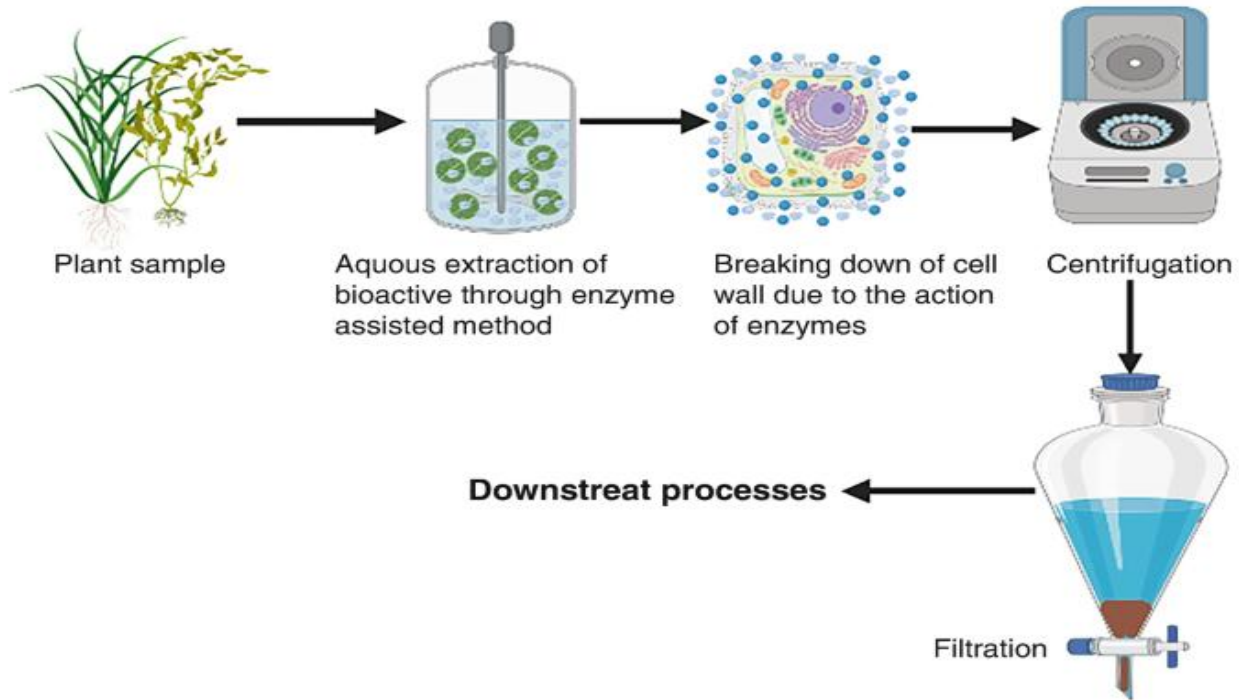


Figure 15: Enzyme-assisted extraction process

1.4.1.3 Industrial applications in the food sector:

- ✓ **Dairy products** : Use of enzymes for the extraction of proteins from of milk or derived products.
- ✓ **Legume cakes** : Extraction of protein isolates from plant residues.
- ✓ **Functional properties** : Protein isolates used as emulsifiers, foaming agents, and stabilizers in various food formulations

Limitations and challenges of enzyme-assisted extraction

- ✓ Commercial problems: High cost of enzymes on an industrial scale.
- ✓ Dependence of enzymes on specific environmental factors (temperature, pH).

Future improvements:

- ✓ Development of new, more specific enzymes.
- ✓ Cost reduction through the synthesis and purification of suitable enzymes.

1.4.2 Physical methods

1.4.2.1 Cavitation-assisted extraction

Cavitation-assisted extraction (CAE) shows interest in more environmentally friendly methods compared to conventional protein extraction methods, such as reflux, percolation and maceration with organic solvents.

Ecological and economic advantages: CAE can reduce solvent use and provide energy savings, making these processes more sustainable and less expensive.

Definition of cavitation

Cavitation is the process of bubble formation, growth, and implosion in a liquid under the effect of variations in pressure and temperature, leading to a release of energy and the breakdown of cellular structures (figure 16).

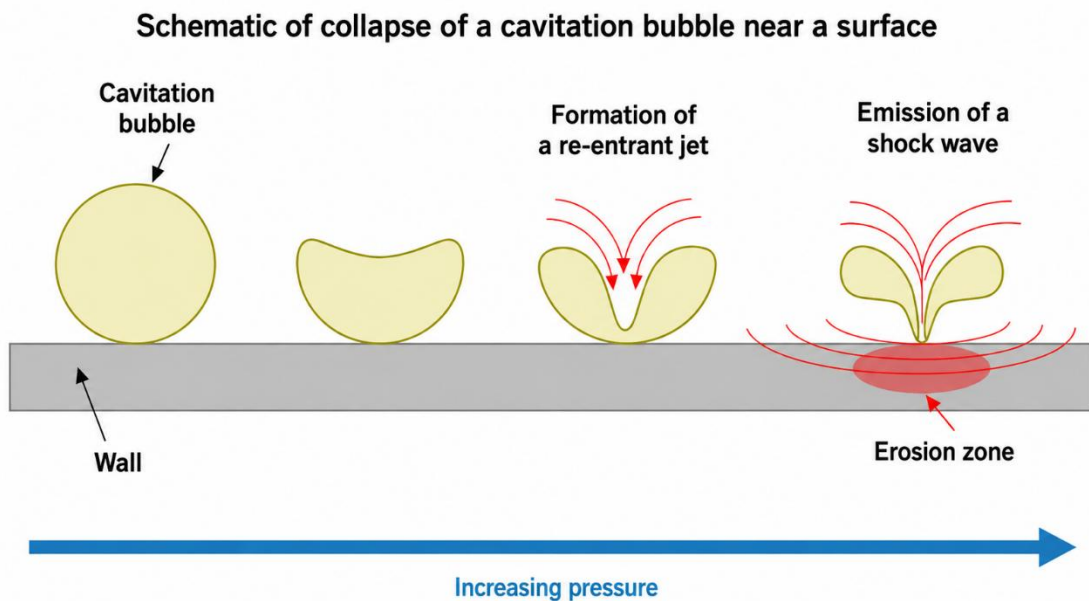


Figure 16: Mechanism of action of cavitation against a cell wall.

Mechanisms

- ↪ **The increase in temperature and pressure** resulting in a rate of high mass transfer. The two factors combined are responsible for a diffusion and implosion of bubbles, facilitates extraction by:
- ↪ **Mass transfer** : Acceleration of the transfer of compounds to the solvent.
- ↪ **Pore enlargement** : Formation of micro- and nanopores, improving the protein release.

↪ **Production of reactive free radicals** : Degradation of the cell wall, facilitating extraction.

Cavitation Techniques Used

↪ Ultrasound Assisted Extraction (UAE)

↪ Hydrodynamic Cavitation Extraction (HCE)

a. *Cavitation Techniques Used Ultrasound Assisted Extraction (UAE)*

Principle : ultrasonic waves generate cavitation bubbles which implode in the liquid, creating micro-currents and turbulence increasing extraction efficiency. Ultrasound is an acoustic wave whose frequencies range from 20 kHz to several gigahertz. Ultrasound-assisted extraction relies on cavitation effects which intensify mass transfer, and on a close interaction between the solvent and the biological matrix (figure 17). The collapse of cavitation bubbles at proximity to tissue surfaces produces micro-jets, which results in a tissue disruption and deep penetration of the solvent into the tissue matrix.

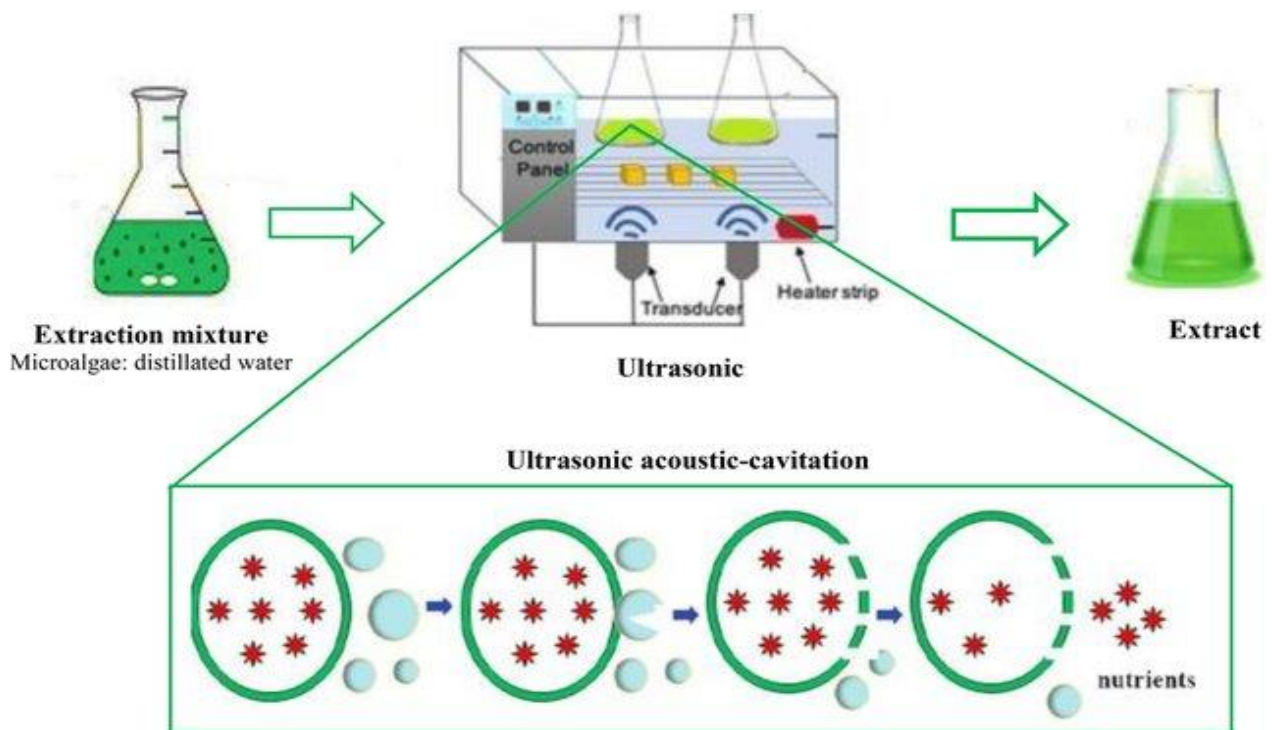


Figure 17: Processes of cavitation techniques used ultrasound assisted extraction.

Influential parameters

- **Temperature and solvent characteristics:** the polarity of the solvent and the temperature are important for the efficiency of protein diffusion.
- **Type of reactor (bath or probe) (figure 18), sonication frequency and power :** How these parameters affect the magnitude of cavitation and therefore the extraction yield.

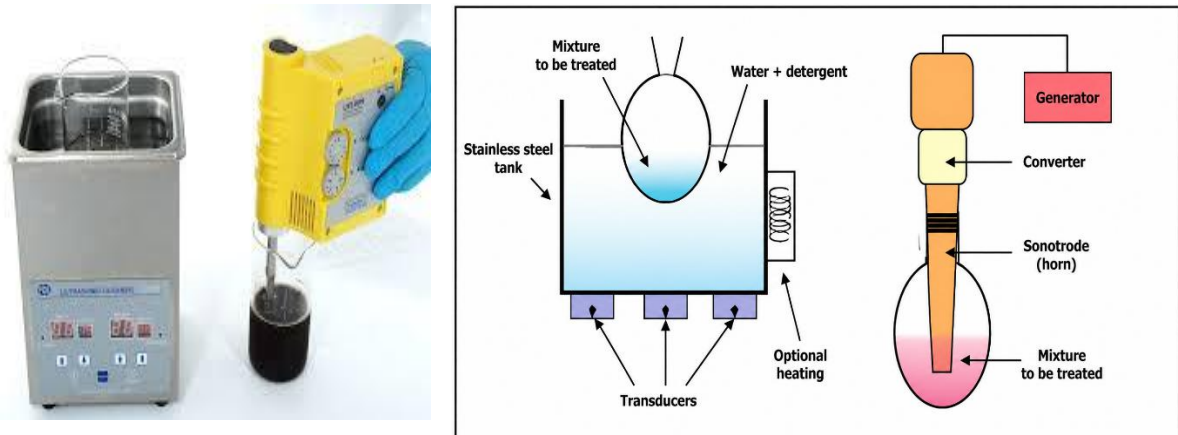


Figure 18: Type of reactor (bath or probe).

Preference for water : water is considered the solvent of choice for the CAE, due to its safety and ability to extract various compounds such as amino acids versus organic and other solvents inorganic.

Advantages and limitations of UAE : the technique is effective in extraction of proteins but also its scaling limitations due to emulsification, cell rupture and meat tenderization.

Cavitation Extraction assisted by Hydrodynamics (HCE)

Principle : cavitation is produced by forcing the liquid through an orifice under pressure, which generates bubbles in a flow continuous (figure 19).

Differences with UAE : Intensity and volume of cavitation bubbles : the intensity of the implosions (more low in HCE but more cavities generated).

Scaling advantages: HCE is easier to implement for large-scale continuous processes.

Common applications: use in emulsification, cell disruption and meat tenderization.

Advantages and limitations of HCE : Although easier to scale, the HCE may be less efficient in terms of cavitation force compared to UAE but allows for faster, high-volume extraction.

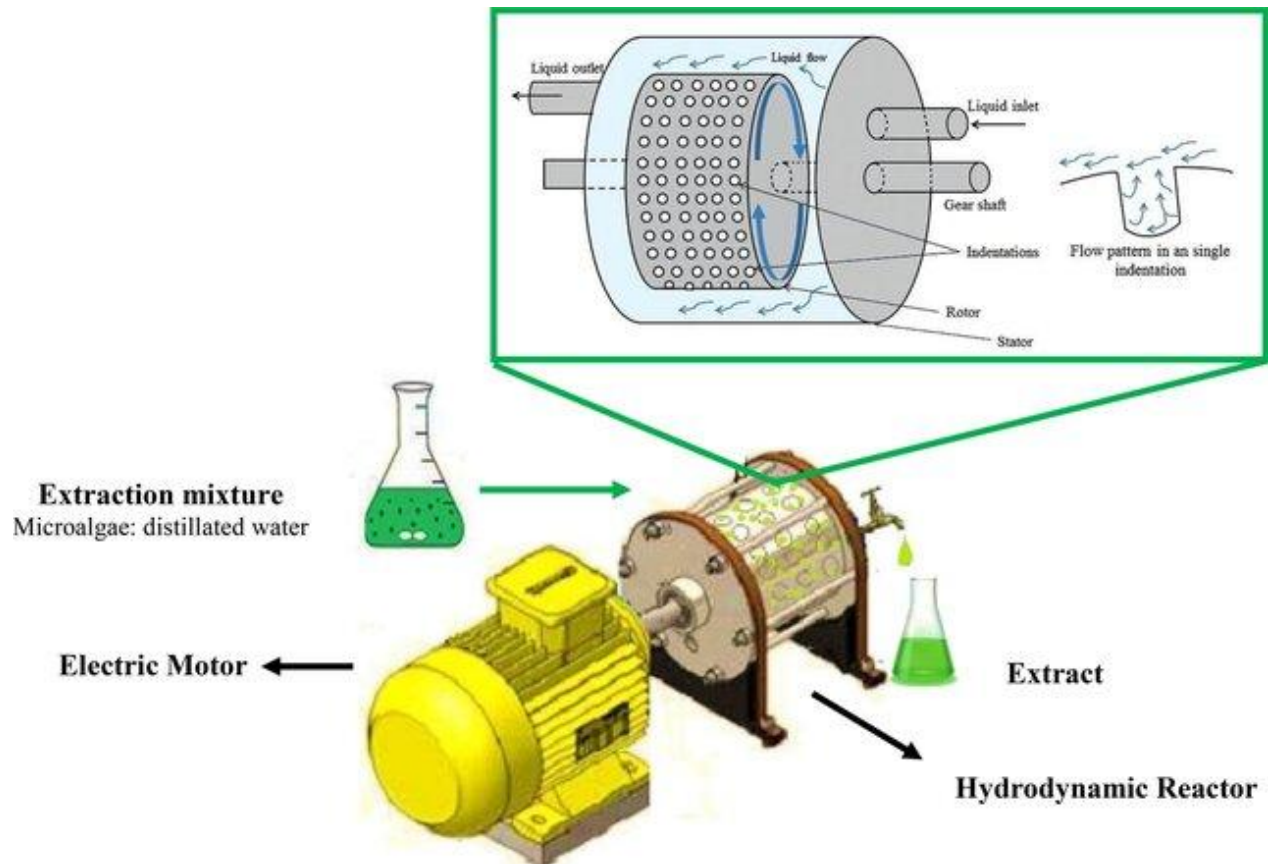


Figure 19: Cavitation Extraction assisted by Hydrodynamics

1.4.3 Chemical methods

1.4.3.1 Supercritical extraction process

Supercritical water is universally known as a powerful alternative against conventional protein extraction methods. This technique extraction uses high pressure hot water treatments (100 and 374 °C).

Ecological and economic advantages

When the water temperature increases to 250°C, it allows the dissolution of the hydrophobic complex. This is mainly due to the decrease of the relative dielectric constant from 80 to 27. Moreover, even in the absence of external catalyst, proteins and carbohydrates can be hydrolyzed in supercritical water.

1.4.3.2 *Extraction by solvent*

The use of aqueous acetone was carried out. The results of the study explained a relationship between solvent concentration and protein content, so that protein content increased with increasing concentration acetone up to 40% only.

- Acetone is sometimes used for protein extraction and precipitation in the context of certain methods of preparation and purification.
- Acetone acts as a water-immiscible solvent that can precipitate proteins due to its ability to dehydrate the solution.

When a solution containing proteins is added to acetone cold, this causes the precipitation of proteins, which can then be recovered by centrifugation.

Using acetone has **several advantages**, such as speed and ability to remove water-soluble contaminants, such as lipids and some sugars. However, it is important to work with acetone at low temperature (usually -20°C) to minimize denaturation proteins and preserve their functional activity.

1.5 **Egg proteins**

Egg can be defined as a low energy source of protein perfectly balanced and very well digestible lipids, ensuring elsewhere 20 to 30% of the daily human requirement in many minerals and vitamins (per 100 g of egg). The three main constituents of eggs are the eggshell (9-11%), the albumen, also referred to as egg white (60-63%), and the egg yolk (28-29%) (figure 20).

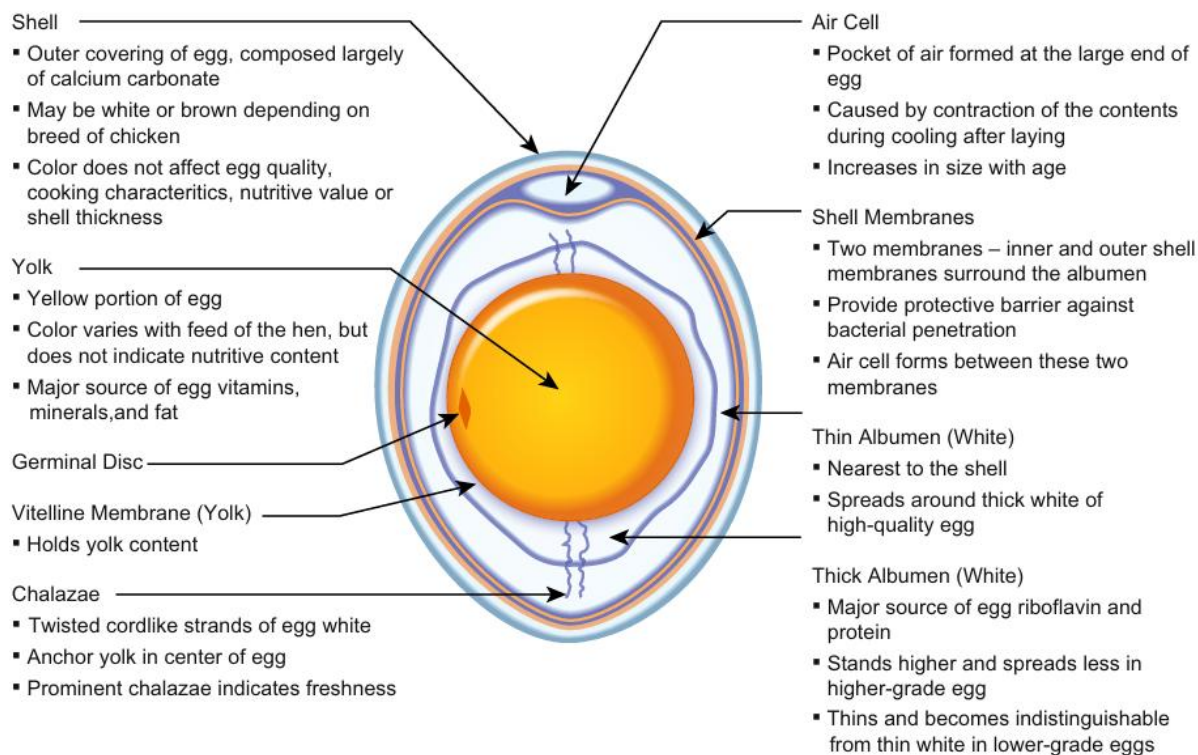


Figure 20: Structure of the hen's egg

1.5.1 Main composition

The main compounds in eggs are summarized in table 5. **Nutritional importance** : 20-30% of daily mineral requirements and vitamins.

Table 5: Composition of albumen, yolk and whole egg.

Egg component	% Protein	% Lipid	% Carbohydrate	% Ash
Albumen	9.7-10.6	0.03	0.4-0.9	0.5-0.6
Yolk	15.7-16.6	32.0-35.0	0.2-1.0	1.1
Whole egg	12.8-13.4	10.5-11.8	0.3-1.0	0.8-1.0

1.5.2 Distribution and general characteristics

The proteins are distributed equally between the egg white and the yolk. The proteins are the main component of egg white and predominantly include 54% ovalbumin, 12-13% ovotransferrin, 11% ovomucoid, 3.5% lysozyme, 2% G2 and G3 ovoglobulins, and 1.5-3% ovomucin. Some trace amounts of other proteins are also found in egg white, including ovostatin, ovoflavoproteins,

avidin, and enzymes such as α -mannosidase, β -galactosidase, and catalase. Ovalbumin is the most abundant constituent of egg white proteins. It is a phosphoglycoprotein with a molecular mass of 45kDa and is composed of 386 amino acid residues. Ovotransferrin and lysozyme have antimicrobial properties, while others give egg white interesting functional properties such as: viscosity, thermogelation, foaming properties and emulsifiers.

The protein content of liquid yolk is approximately 16%. Egg yolk protein constitutes mainly 16% high-density lipoproteins (HDL), 68% low-density lipoproteins (LDL), 10% livetins, 4% phosvitin and very low-density lipoproteins (VLDL). Because LDLs consist of apoproteins and phospholipids, they have amphiphilic properties and can be dispersed at the oil-water interface. Thus, LDLs are the essential component responsible for the emulsifying properties of egg yolk. However, egg yolk HDLs may have therapeutic potential including antioxidant and antimicrobial properties.

1.5.3 Digestibility of egg white proteins

Digestibility in raw state: low (51%) due to the antitrypsin activity of ovomucoid.

Effects of cooking:

- Destruction of antitrypsin activity by heat.
- Coagulation of proteins, promoting digestion (digestibility increased to 91-94%).
- **Factors influencing digestibility:** importance of cooking to optimize protein assimilation.

1.6 Functional properties of milk proteins and improvement

Milk and dairy products are important sources of animal protein, vitamins, minerals, and essential fatty acids for infants and young adult. Milk is defined as the normal lacteal secretion, practically free from colostrum, obtained by the complete milking of one or more healthy cows, containing not less than 8.25% milk solids-not-fat and not less than 3.25% of milk fat.

1.6.1 General composition of milk proteins

The major constituents of milk are water (86-88%), milk fat (3-6%), protein (3-4%), lactose (5%), and minerals (ash) (0.7%), with total solids of 11-14%. The composition of milk is affected by a variety of factors, including breed, stage of lactation, nutritional and health status of the cow, season (which can be both temperature and stage of lactation effects, if calving is not even throughout the year), and genetic factors.

The lipid component of cow's milk (3-5%) is secreted by the lactating cow as small fat globules ranging in size from <1 to 15 μm in diameter. Each fat globule is surrounded by a special interfacial layer, or milk fat globule membrane, composed of a lipid bilayer and protein. This layer ensures that each lipid droplet is dispersed in the aqueous milk serum and is unable to aggregate with others. Lipids are now recognized to be predominantly polar, with a very small amount of neutral lipids including triacylglycerols, diacylglycerols, monoacylglycerols, and cholesterol and its esters.

The proteins of milk are a heterogeneous mixture and include two main groups: caseins and whey or serum proteins. These are composed of six major proteins: α₁-casein, α₂-casein, β-casein, κ-casein, b-lactoglobulin, and a-lactalbumin. Other protein fractions are also present but at very low levels, including bovine serum albumin (BSA), immunoglobulins, lactoferritin (LF), PP3, and ceruloplasmin. Caseins (figure 21) represent the major protein group and were defined as phosphoproteins precipitated from raw milk by acidification to pH 4.6 at 20°C, 80% caseins (micellar fraction). The residual proteins in the serum or whey after the removal of caseins were referred to as whey proteins 20% serum proteins (soluble fraction).

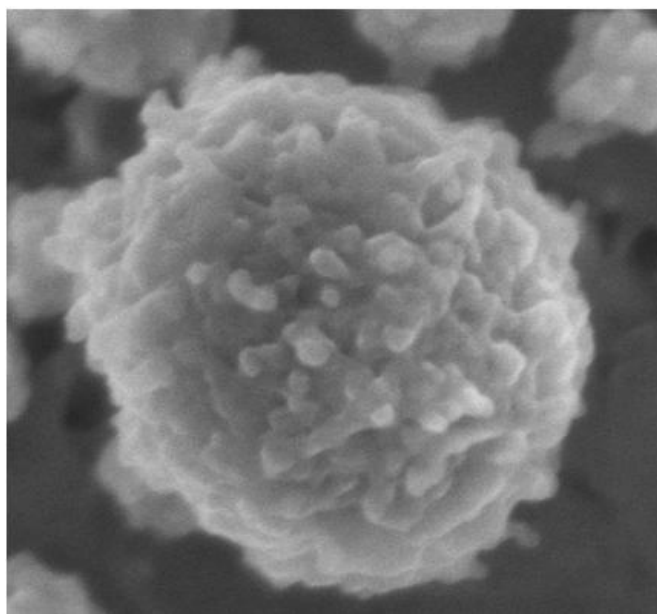


Figure 21: Image of a casein micelle from field-emission scanning electron microscopy.

1.6.2 Importance of milk proteins

- ↪ Nutritional role: complete source of essential amino acids.
- ↪ Industrial applications: functional and bioactive ingredients in food and health.

1.6.2.1 *Functional properties of caseins*

Formation of micelles stabilized by calcium phosphate. Ability to gel (enzymatic coagulation or acidification lactic acid). Examples: production of yogurts, cheeses. Emulsifying and film-forming properties (use in packaging biodegradable).

1.6.2.2 *Technological modifications*

- Microfiltration: production of native micellar casein.
- Enrichment or depletion of minerals (calcium, zinc, iron). Impact on digestibility and thermal properties.

1.6.3 **Whey proteins**

- Classification and characteristics

Soluble proteins after destabilization of caseins (pH 4.6 or action of chymosin).

- Main proteins:
 - lactoglobulin (absent in human milk). lactalbumin (major in human milk).
 - Bovine serum albumin, lactoferrin, and immunoglobulins.

1.6.3.1 *Bioactivity of whey proteins*

- ❖ Lactoferrin : antimicrobial activity, role in immunity.
- ❖ Immunoglobulins : immune defense.
- ❖ Technological properties : solubility over a wide pH range, retention water, thermal gelation.

Derivatives

- ↗ **Whey protein concentrates (WPC)** : 30-70% protein (DM).
- ↗ **Whey protein isolates (WPI)** : up to 90% protein (DM).
- ↗ **Purified specific proteins** : α -lactalbumin, β -lactoglobulin.

1.6.4 **Biological activity and bioactive peptides proteins and bioactive fractions**

- ↗ Enzymes : lactoperoxidase, lipase, lysozyme, xanthine oxidase, plasmin, acid and alkaline phosphatases.
- ↗ Hormones : insulin, prolactin, growth factor (lactoferrin.....etc).

Whose concentrations vary depending on lactation (colostrum vs. milk mature).

1.6.4.1 Bioactive peptides released by hydrolysis

Studies show the presence of active peptides in the milk protein sequence; this is for example the case: **casein glycomacropeptide**, **casein casomorphins**, peptides with antimicrobial activity or immunomodulatory peptides.

1.6.4.2 Protein/ water interactions

Solubility, water retention, swelling (important for hydrated products).

1.6.4.3 Protein/ protein interactions

-Gelling (example: production of yogurts).

-Texture and consistency dependent on gelling mechanisms.

1.6.4.4 Protein/ surface interactions

An emulsion or foam is stabilized by proteins, when these are found on the surface of oil droplets (emulsion) or gas bubbles (mousse) (figure 22). The adsorption properties of proteins on the surface of oil droplets or gas bubbles promoting the formation of films protecting the emulsions and foams against coalescence destabilization mechanisms

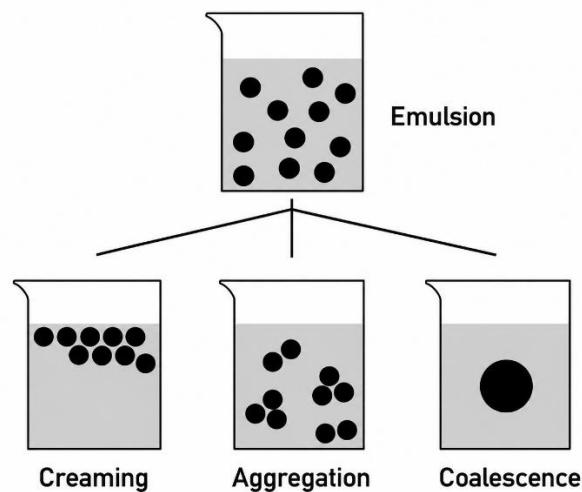


Figure 22: emulsion or foam aspect

1.6.5 Industrial applications

Caseinates are casein salts obtained by neutralizing casein with bases such as sodium hydroxide (NaOH), calcium hydroxide (Ca(OH)₂) or potassium (KOH).

Features :

*Soluble in water, unlike pure casein.

* Better digestibility and use in food formulations.

Exist in different forms depending on the cation used:

- ↳ **Sodium caseinate:** the most common, soluble and used for its functional properties (emulsifier, stabilizer).
- ↳ **Calcium caseinate:** used to enrich foods with calcium.
- ↳ **Caseinates :** have emulsifying, foaming properties (ability to give a large volume of foam), plasticizers (obtaining films, packaging biodegradable food), binding agents.
- ↳ **Serum proteins:** are soluble across the entire pH range. They exhibit water retention properties, foaming, gelling properties (thermally induced), etc.

Chapter 3 : Lipids

1.1 Definitions

Lipids, proteins, and carbohydrates make up the main structural building blocks of food. Lipids are broadly described as substances that dissolve readily in organic solvents such as ether or chloroform, yet remain largely insoluble in water. That said, no universally agreed-upon scientific definition exists for lipids, largely because certain molecules within this category exhibit partial water solubility, making a strict boundary difficult to draw.

Lipids vary considerably in their affinity for water. Triacylglycerols, for instance, are strongly hydrophobic, whereas di- and monoacylglycerols carry both water-attracting and water-repelling regions within the same molecule, allowing them to dissolve in moderately polar solvents. Short-chain fatty acids (C1–C4) go even further, mixing completely with water while remaining insoluble in nonpolar environments. Despite this variability, solubility remains the most widely accepted basis for defining lipids, a characteristic unique among macromolecules, most of which are classified according to shared structural features rather than solubility behavior.

As a group, lipids share certain common properties and compositional similarities. Among them, triacylglycerols, commonly known as fats and oils, represent the most abundant subclass. Although the terms lipid, fat, and oil are often used interchangeably, subtle distinctions apply: "lipid" refers broadly to all molecules meeting the solubility-based definition, while "fat" typically denotes lipids that are solid at room temperature and "oil" refers to those that are liquid. For regulatory purposes, the U.S. Food and Drug Administration (FDA) has defined total fat as the sum of fatty acids from C4 to C24, expressed as triglycerides, a practical standard designed to resolve any ambiguities in nutrition labeling.

1.2 General classification

Lipids found in food can be organized into three main categories:

- Simple lipids: Esters formed between fatty acids and an alcohol:
 - ↳ Fats: Fatty acid esters of glycerol, also called triacylglycerols
 - ↳ Waxes: Fatty acid esters of long-chain alcohols other than glycerol (e.g., myricyl palmitate, cetyl palmitate, vitamin A esters, and vitamin D esters)

- Compound lipids: lipids containing additional chemical groups beyond a basic fatty acid–alcohol ester:
 - ↪ Phospholipids: Glycerol-based structures esterified with fatty acids, phosphoric acid, and nitrogen-containing groups (e.g., phosphatidylcholine, phosphatidylserine, phosphatidylethanolamine, and phosphatidylinositol)
 - ↪ Cerebrosides: Molecules combining a fatty acid, a carbohydrate unit, and a nitrogen-containing component (e.g., galactocerebroside and glucocerebroside)
 - ↪ Sphingolipids: Compounds built from a fatty acid, a nitrogen moiety, and a phosphoryl group (e.g., sphingomyelins)
- Derived lipids: Substances obtained through the breakdown or transformation of simple or compound lipids. These retain the general characteristics of lipids and include fatty acids, long-chain alcohols, sterols, fat-soluble vitamins, and hydrocarbons.

1.3 Lipid content in foods

Foods may contain any combination of the lipid types described above. Bovine milk serves as a useful example of this complexity, harboring a diverse array of lipids that differ in both polarity and concentration. Across all food sources, triacylglycerols and phospholipids tend to be the most nutritionally significant. Triacylglycerols that remain liquid at room temperature are called oils, soybean and olive oil being common examples and are predominantly derived from plants. Those that are solid at room temperature are called fats, such as lard and tallow, which typically come from animal sources. It is worth noting, however, that the term "fat" can apply to all triacylglycerols regardless of whether they are solid or liquid under normal conditions. The lipid content varies widely across different foods.

1.4 Chemical and physical properties of lipids

1.4.1 Physical properties

1.4.1.1 Solubility

Butyric acid at 4°C is soluble in water, then the solubility of fatty acids decreases gradually. They are soluble in nonpolar organic solvents : benzene, chloroform

1.4.1.2 The melting point

The melting point increases with the number of carbon atoms and decreases when the number of double bonds increases.

1.4.1.3 Oxidation of double bonds

Oxidation by atmospheric oxygen leads to the rancidity of fats. Intracellular enzymatic oxidation of Arachidonic acid, via cyclooxygenase (cyclization + oxidation), leads to prostaglandins (figure 23), which are mediators were very active, but very quickly they deteriorated.

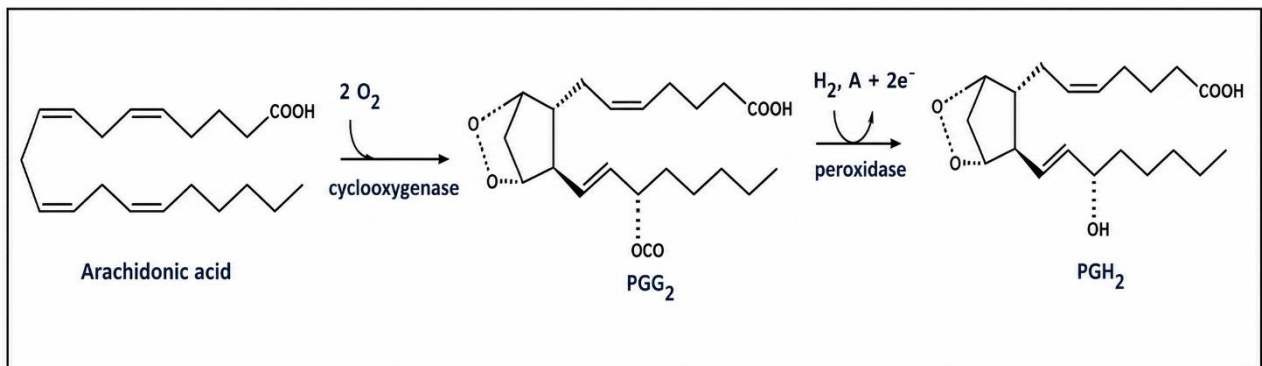


Figure 23: Intracellular enzymatic oxidation of Arachidonic acid

Prostaglandins are involved: (1) in the contraction of smooth muscles (intestine, uterus, blood vessels); (2) in the regulation of metabolism; (3) in platelet aggregation. Inhibition of platelet cyclooxygenase by Aspirin is useful in therapy (antiplatelet agent).

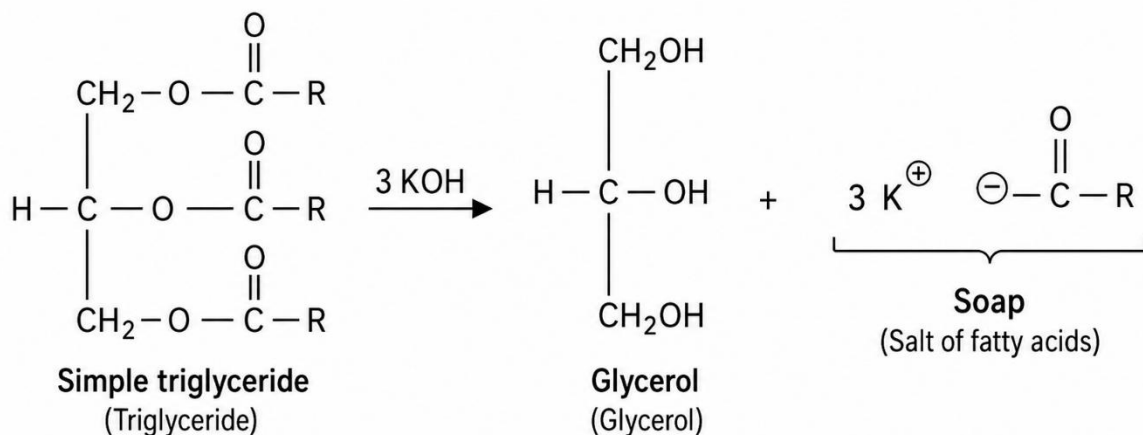
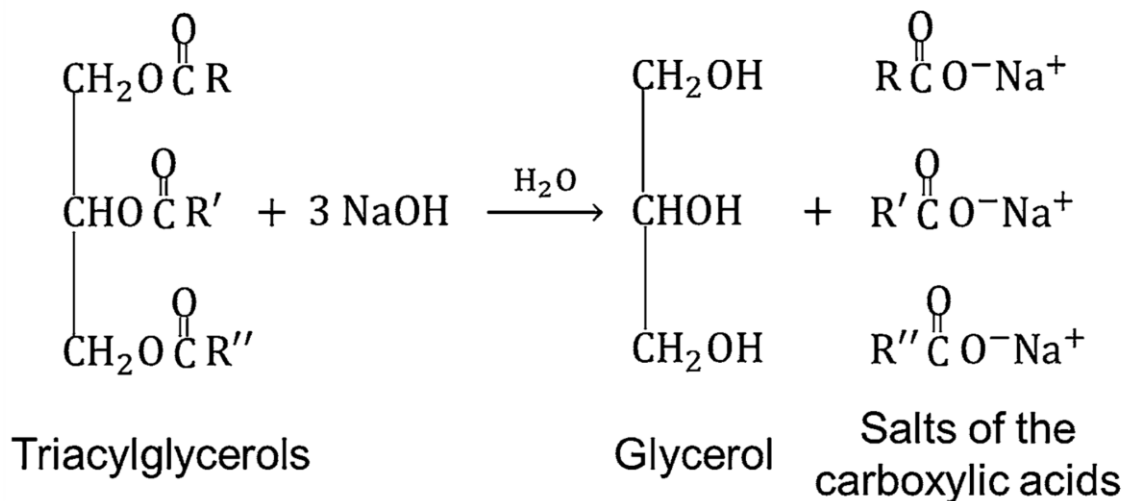
1.4.2 Chemical properties

The chemical properties of glycerides depend essentially on those of fatty acids that constitute them.

1.4.2.1 Hydrolysis and saponification

The hydrolysis of triglycerides releases one or several fatty acids. The reaction can be carried out by sulfuric acid or by way enzymatic.

Saponification is an alkaline hydrolysis by KOH or NaOH (figure 24), leading to the formation of soaps with monovalent or divalent cations: Natural soaps are sodium salts.



Reaction of a simple triglyceride with KOH (alkaline hydrolysis)

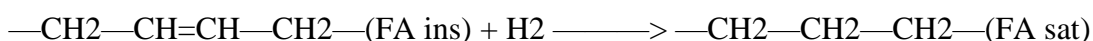
Saponification reaction of a simple triglyceride by KOH

Figure 24: Reactions of saponification

1.4.2.2 Hydrogenation

The hydrogenation takes place under 100 to 200 bar, 200 to 400 °C and in the presence of catalysts (finely divided nickel), by a stream of hydrogen.

The bonds of unsaturated fatty acids in triacylglycerols are saturated. This reaction is used in the food industry to improve fat preservation because it makes solid or semi-solid oils (margarines) less sensitive to oxidation (rancidity).



1.4.2.3 Esterification

In the food industry, this reaction is used to determine the fatty acid composition of oils and fats in order to determine the fraudulent mixtures (figure 25).

Fatty acid + Alcohol \rightleftharpoons Ester + Water

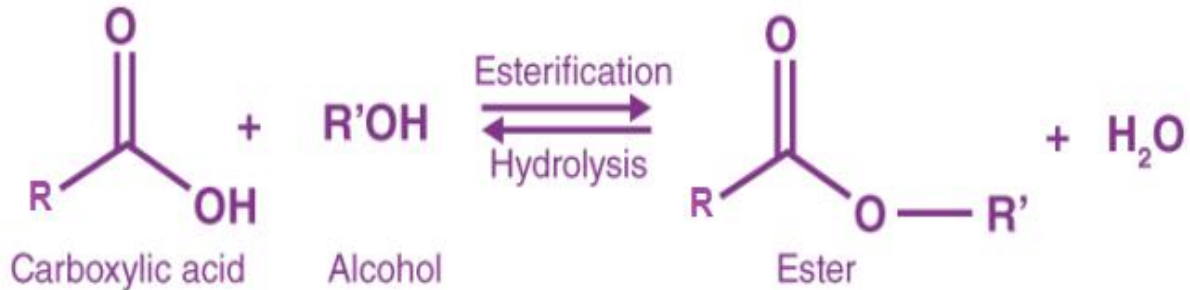


Figure 25: Esterification reaction

1.4.2.4 Organoleptic properties

Edible fats are liquid (oils) or solid (butter) products:

- ↗ Yellowish in color
- ↗ With a smooth, creamy feel
- ↗ Most often of a bland taste
- ↗ When properly stored, they should have virtually no acidity (neutral).

1.5 Functional properties of fats

- ↗ A structural role : e.g. phospholipids, constituent of membranes.
- ↗ A messenger role : AG precursors of several intra- and extra- messengers cellular.
- ↗ A role in transporting fat-soluble vitamins.
- ↗ In camels and dromedaries, lipids constitute a water reserve.

The main properties of fatty acids are linked to their unsaturation: the more unsaturation, the higher the melting point, the lower the melting point (increasing fluidity), the more their physiological interest increases, as does their chemical reactivity, which plays negatively impacting stability, particularly through increased susceptibility to oxidation. We have: Saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), and polyunsaturated fatty acids (PUFAs).

- **Importance of fats in food** : energy, functional, and nutritional.

- **Chemical structure** : Carboxylic group and hydrocarbon chain.
- **Presence and type of unsaturations**: cis/trans.
- **The geometry of the unsaturations also influences the behavior at the melting** : at equivalent chain lengths, a trans isomer of fatty acid will have thus a melting point (MP) intermediate between those of its cis isomer and of the corresponding saturated fatty acid (stearic acid, C18:0, PF=70 °C; acid oleic acid, C18:1 cis, PF = 13 °C; elaidic acid, C18:1 trans, PF = 45 °C).

1.5.1.1 *Physical and rheological properties of fats*

Melting and solidification:

Impact of fatty acids:

- ↷ Saturated fatty acids (solids at room temperature).
- ↷ Unsaturated fatty acids (fluid oils).
- ↷ Conditions influencing crystallization:
 - ↷ work (shearing, agitation).

1.5.1.2 *Physical and rheological properties of fats*

Hardness and consistency of fats depends of

- ↷ its fatty acid composition; from hardest to most fluid: SFA > MUFA > AGPI; Practical examples: butter, margarine, liquid oils.
- ↷ its triglyceride structure which influences the extent of polymorphism the solid state;
- ↷ conditions for its implementation:
 - ❖ temperature and thermal history (cooling rate, therefore of crystallization),
 - ❖ mechanical work (agitation, pressure or shearing during the crystallization).

1.6 Nutritional requirements for fats

1.6.1.1 *Energy and nutritional role*

Lipids provide the greatest amount of energy per gram. As well as their supply of essential fatty acids and fat-soluble vitamins.

- ✓ Protein: 1 g = 4 kcal
- ✓ Lipids or fats: 1 g = 9 kcal

- ✓ Carbohydrates or sugars: 1 g = 4 kcal

1.6.1.2 Essential fatty acid intake

Essential fatty acid intake necessary for development and healthy the functioning of the human body which cannot produce them, they participate in the prevention of cardiovascular diseases (anti-atherosclerotic, hypo-triglyceridemic) exp:

- ✓ Omega 3, the essential precursor of which is alpha-linolenic acid.
- ✓ Omega 6, the major representative of which is linoleic acid.

1.7 Preservation and alteration of lipids

Lipid oxidation is a series of chemical reactions involving the attack lipids are broken down by oxidizing agents, leading to their degradation.

Impact : Loss of organoleptic properties (taste, smell), decrease in nutritional value, and possible toxicity.

1.7.1 Factors influencing lipid stability against oxidation

1.7.1.1 Substrates of oxidation

a. Fatty acid composition

- ✓ Polyunsaturated lipids (e.g., linoleic and linolenic acids) are more reactive due to the presence of double bonds.
- ✓ Position of unsaturations: activated methylenes (CH between two double bonds connections) are particularly vulnerable.

b. Structure and location

Free lipids are more susceptible to oxidation than those complexed in structures such as cell membranes.

1.7.1.2 Oxidation Catalysts

- ↳ Metallic ions (Fe, Cu) : Promote the formation of free radicals by reactions from Fenton or Haber-Weiss.
- ↳ Heme proteins : The presence of heme groups can accelerate oxidation lipids.
- ↳ Pro-oxidant enzymes:
 - Lipoxygenase: catalyzes the formation of lipid hydroperoxides.

- Other metalloprotein enzymes.

1.7.1.3 Antioxidants

- ↪ Natural : Tocopherols, carotenoids, polyphenols.
- ↪ Mechanism : They act by trapping free radicals or by chelating ions metallic.

1.7.1.4 External Factors

- ↪ Temperature : An increase in temperature accelerates auto-oxidation.
- ↪ Light : Particularly in the presence of photosensitizers, it promotes photo-oxidation.
- ↪ Oxygen : The availability of oxygen determines the reaction rate.
- ↪ Water activity : Low humidity conditions can increase the exposure of lipids to oxidants.
- ↪ Storage : Duration, packaging, and modified atmosphere (nitrogen, CO₂).

Lipid oxidation (figure 26) can result from several reaction pathways depending on from the environment and the initiating agents:

- ✓ Auto-oxidation catalyzed by temperature, metal ions, radicals free;
- ✓ Photo-oxidation, initiated by light in the presence of photosensitizers
- ✓ Enzymatic oxidation initiated by lipoxygenase.

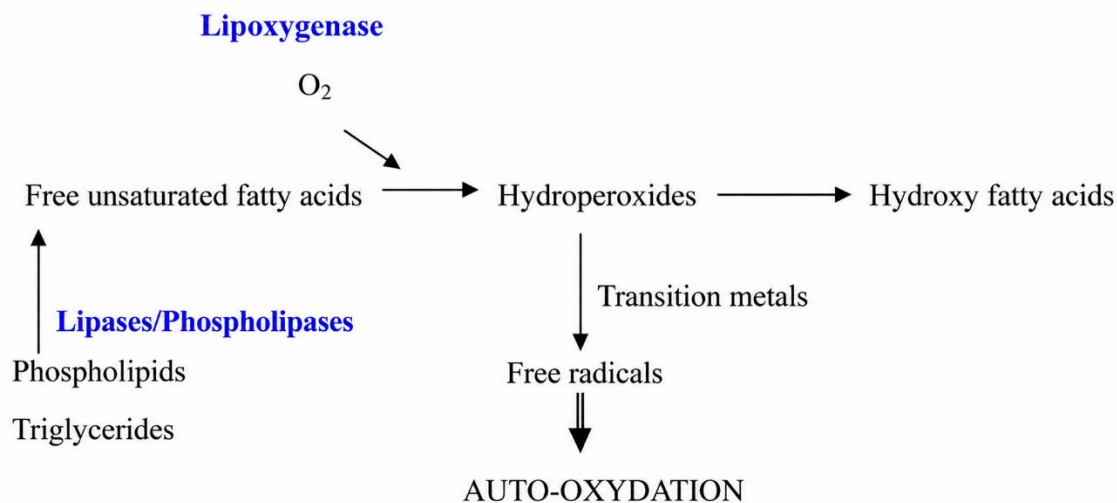


Figure 26: Enzymatic oxidation of fatty acid.

Chapter 4: Study of Polysaccharides

Carbohydrates are hydrates of carbon composed of carbon, hydrogen, and oxygen usually present in the ratio 1:2:1 and with a general formula of $C_n(H_2O)_n$. They are an important source of energy and provide 17 kJ energy per gram and are utilized before fats and protein in the body to meet the energy requirements. They are abundantly found in nature in plants, animals, and microorganisms in the form of simple sugars such as glucose, fructose, and sucrose and as complex sugars such as cellulose, pectin, and hemicellulose.

1.1 Cellulose and its derivatives

Cellulose and hemicelluloses represent the principal components of dietary fibre found in plant foods, particularly cereals. Defining these substances proves challenging due to the absence of clear boundaries, both in terms of chemical composition and nutritional function.

Cellulose is a major structural component of plant cell walls and the most abundant organic polymer. It consists of linear chains of at least 3000 β -1,4-linked glucopyranose units, which form a flat-ribbon structure stabilized by intra- and intermolecular hydrogen bonds. This highly ordered structure is responsible for its complete insolubility in most reagents and its resistance to mammalian digestive enzymes.

Hemicelluloses are a diverse group of structural polysaccharides (e.g., xylans, mannans) closely associated with cellulose. Unlike cellulose, they are often branched and more soluble (e.g., in alkaline solutions). Their branched nature prevents tight packing, allowing some solubility and making them a component of the "insoluble fiber" fraction.

Dietary fiber is defined (AACC, 2000) as the edible parts of plants or analogous carbohydrates that are resistant to digestion and absorption in the human small intestine, with partial or complete fermentation in the large intestine. It includes polysaccharides, oligosaccharides, lignin, and associated plant substances. Its physiological benefits include laxation and attenuation of blood cholesterol and glucose.

The classification of fiber has evolved. The Englyst method measures Non-Starch Polysaccharides (NSP) (cellulose, hemicelluloses, pectins, etc.), while the AOAC/Southgate method yields a "total

dietary fiber" value, which includes lignin and resistant starch. The internationally accepted AACC definition aligns with the AOAC methodology.

1.2 Starch

Starch is a polysaccharide which upon complete hydrolysis releases glucose. Most of the starches and starchy foods used in food preparation are obtained from cereals (rice, wheat, maida, sago, maize, barley), roots (cassava, tapioca, arrowroot) and tubers (potatoes, sweet potatoes). Starch is present in small particles known as granules. These granules are of various shapes and sizes. Starch granules present in the corn grain is of a different shape and size from that of a potato tuber (figure 27)

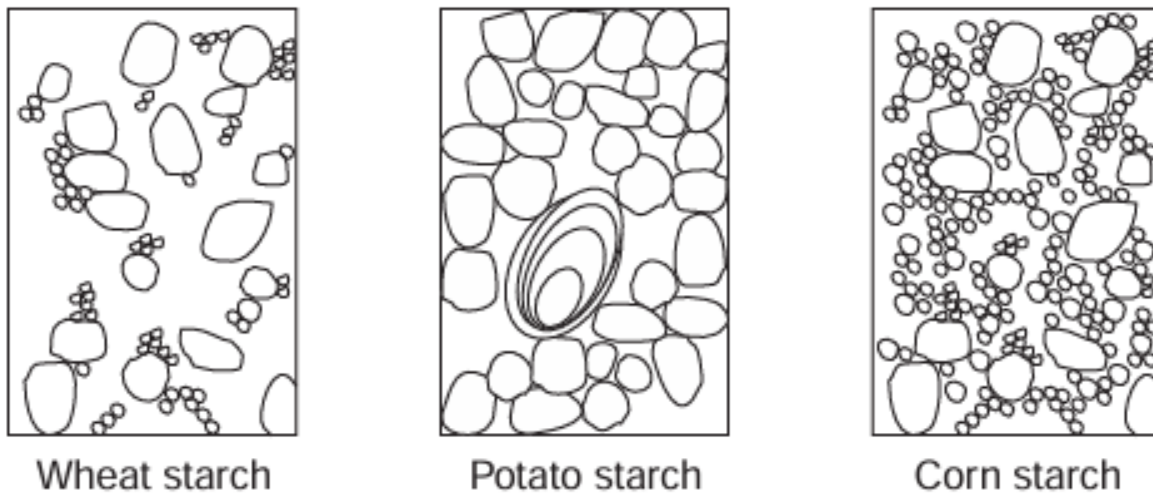


Figure 27: Structure of Wheat, Potato and Corn Starch Granules.

Starch is made up of two fractions **Amylose and Amylopectin**. The amylose fraction of starch is composed of straight-chain structure, while the amylopectin fraction has a branched chain configuration. The two possess different properties. Amylose contributes gelling characteristics to cooked and cooled starch mixtures. Amylopectin provides cohesive or thickening property but does not usually contribute to gel formation.

1.2.1 Gelatinization and retrogradation phenomena

The starch granule is completely insoluble in cold water. However, when a mixture of starch and water is cooked, a starch paste is formed. The starch granules absorb water, swell in size and as the temperature is increased, they burst (figure). Some pastes are opaque, some are clear, semiclear or

cloudy in appearance. In general, pastes made with cereal starches such as corn, or wheat, are cloudy in appearance, whereas those made from root starches such as potato, tapioca are clear. When some starch pastes are cooled, they become rigid and form a gel on standing, e.g., corn starch. However, some starch pastes do not form a gel.

Retrogradation: When the thickened mixture of starch is allowed to cool without disturbance, there is a tendency for reassociation of intermolecular hydrogen bonding. This is usually accompanied by increased viscosity and turbidity of pastes, exudation of water, and increased degree of crystallinity. The process is termed as retrogradation (figure 28). The development of hard and tough crystalline structure of retrograded amylopectin fraction of starch leads to undesirable changes in texture as in case of staling of bread and cakes. Retrogradation is desirable in some food applications due to the slower enzymatic digestion of retrograded starch and moderated release of glucose into the bloodstream such as food for geriatric and obese people.

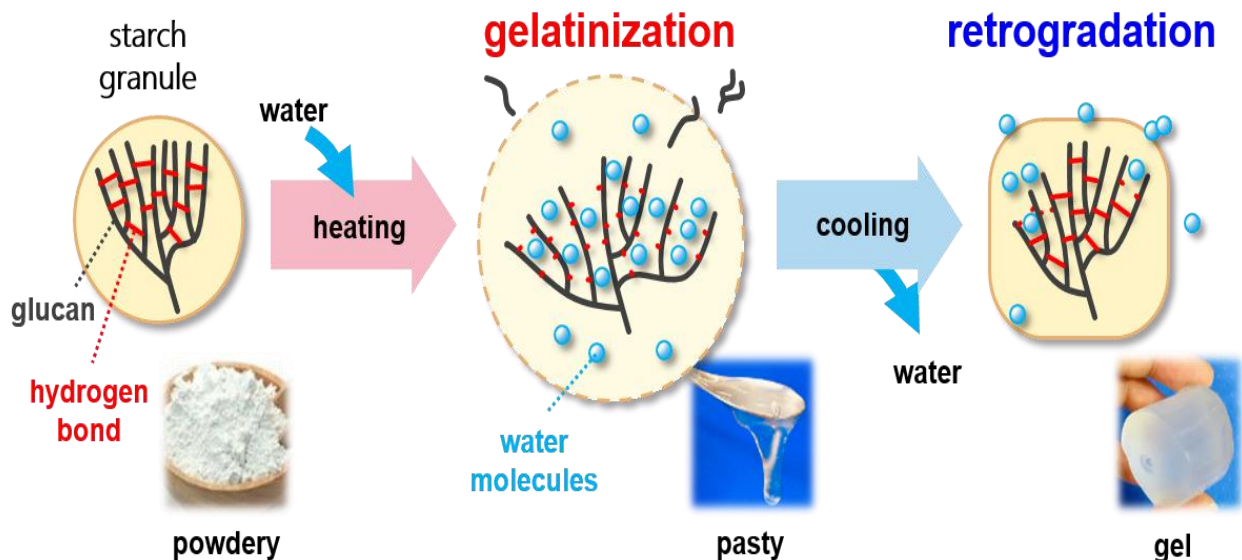


Figure 28: Gelatinization and retrogradation phenomena

1.3 Functional properties of native and modified starches

Native starches have specific functional properties (e.g., thickening, gelling, water-binding) that are influenced by their amylose/amylopectin ratio and granule structure. The gelatinizing property of starches is used in the food industry to develop thick gels and also to increase the viscosity of flowable foods such as soups. Properties of native starch can be altered to achieve variable paste

viscosity/consistency, clarity, and stability. For this purpose different chemical and physical treatments may be employed such as cross-linking, oxidization, depolymerization, and pregelatinization. Modified starches show increased solubility and stability, improved gelatinization and pasting characteristics, better freeze-thaw stability and paste clarity, and reduced gel syneresis.

1.4 Amylolytic enzymes

Amylases (e.g., α -amylase) are used to break down starch and proteins in the analytical determination of dietary fiber (e.g., in the Englyst and Southgate methods). These enzymes simulate the digestive processes of the small intestine, removing digestible components to isolate the indigestible fiber fraction.

1.5 Dietary fiber: soluble fiber, pectins, and gelation

Soluble Fibers include pectins, gums (e.g., oat β -glucan), and inulin. They are characterized by their ability to dissolve or form viscous solutions. They are particularly associated with lowering blood cholesterol levels.

Pectins are soluble fiber components found in plant cell walls that are known for their gelling properties, which are exploited in food products like jams and jellies.

Gelation in soluble fibers is linked to their health benefits. For example, the high viscosity of soluble fibers like oat β -glucan (a mixed-linkage β -glucan) is believed to impede the reabsorption of cholesterol and bile acids, thus lowering blood cholesterol.

Inulin, a fructan polymer of fructose, is a soluble fiber that resists digestion in the small intestine but is readily fermented by bacteria in the colon. This fermentation produces short-chain fatty acids (e.g., acetic, propionic, butyric) that contribute to human energy metabolism and promotes the growth of beneficial bacteria (e.g., bifidobacteria).

Fermentation of Dietary Fiber: Bacteria in the large intestine ferment undigested fibers, producing short-chain fatty acids (acetic, propionic, butyric), lactic acid, and gases (H_2 , CO_2 , methane). These fatty acids are reabsorbed and contribute to the host's energy supply, providing approximately 1.8 kcal per gram of original monosaccharide.

1.6 Case of pectins

Pectins are highlighted as a classic example of soluble fiber, distinct from insoluble cellulose and hemicelluloses in their chemical structure and physiological effects. They are a key component in the "soluble non-cellulosic polysaccharides" group and are known for their capacity to form gels.

Chapter 5: Food system

1.1 General aspect

Food systems have traditionally been understood as a sequence of interconnected activities spanning from agricultural production to end consumption, often framed through the lens of a value chain. Yet, the growing urgency around food security has substantially deepened and broadened how scholars and policymakers conceptualize these systems. Food security itself is a multidimensional challenge, encompassing availability, access, and utilization, while being shaped by a web of environmental, social, political, and economic forces.

A food system, in its most comprehensive sense, encompasses every component: environments, individuals, inputs, processes, infrastructure, institutions, and beyond, along with all activities tied to the production, processing, distribution, preparation, and consumption of food. Crucially, it also includes the outcomes generated by these activities, particularly their socioeconomic and environmental consequences.

Five broad categories of drivers shape how food systems evolve over time:

The first encompasses biophysical and environmental factors, including natural resources, ecosystem services, and the far-reaching effects of climate change.

The second involves innovation, technology, and infrastructure, which collectively determine the technical capacity of a food system to function and adapt.

The third category covers political and economic factors: policy direction, globalization, foreign investment, trade dynamics, food pricing and price volatility, land tenure arrangements, and the destabilizing effects of conflict and humanitarian crises.

The fourth group consists of sociocultural factors, such as cultural identity, religious practices, ritual traditions, and the degree to which women are empowered within food-related decision-making.

Finally, demographic factors: including population growth, aging trends, urbanization, migration, and forced displacement, exert continuous pressure on food system structures and demands.

Importantly, food systems do not operate in isolation. Their drivers, actors, and components are in constant interaction with one another and with adjacent systems, including those governing health,

energy, and transportation. These interconnections make food systems inherently dynamic, subject to ongoing adaptive cycles of growth, restructuring, and renewal.

1.2 Plant-based food systems

1.2.1 Primary and Secondary Metabolites

Metabolism can be broadly defined as the total sum of biochemical reactions carried out within a living organism. The molecules involved, both intermediate compounds and end products, are referred to as metabolites and are generally characterized as small-molecular-weight substances.

Primary metabolism encompasses those reactions and molecules that are absolutely essential to core cellular function, and therefore to the growth and development of the organism. This category includes well-conserved molecules such as carbohydrates, lipids, and nucleic acids compounds found consistently across all living organisms. Primary metabolites like amino acids, organic acids, and nucleosides represent some of the most industrially significant biotechnological products, commonly serving as raw materials in fermentation processes and chemical synthesis.

Secondary metabolism, by contrast, involves reactions and metabolites that are not strictly required for an organism's growth or survival. Secondary metabolites sometimes referred to as natural products tend to be taxonomically restricted, meaning they may be produced by only a limited number of species within a genus, order, or phylum. In some cases, biosynthetic capacity is confined to specific strains within a single species. Plants, bacteria (particularly Actinomycetes), and filamentous fungi are the primary producers of secondary metabolites. Yeasts, protozoa, and animals, on the other hand, produce them only under exceptional circumstances.

Over 2,140,000 secondary metabolites have been identified and catalogued. The major classes of plant secondary metabolites relevant to food biochemistry include: polyphenols, terpenoids, alkaloids.....etc

1.2.1.1 Flavonoids

These are glycosides containing polyphenolic compounds. Flavonoids include a number of compounds such as flavones, flavonols, flavanones (and isoflavones), and flavoxanthin (E161(a)). Yellow-coloured flavonoids such as kaempferol, quercetin, and myricetin are most widely present in all fruits and vegetables. Besides that, tea especially green tea is considered as the most

concentrated and convenient source of flavonoids. *Citrus* fruits are also a rich source of flavonoids. Soybean isoflavones are commercially available in stable isolated form.

1.2.1.2 Anthocyanins (E163)

Water-soluble pigment with various shades of color ranging from red to purple is exceptional characteristic of anthocyanins. The true flavonoids consist of the anthocyanins which are red, blue, and purple pigments; the anthoxanthins which are yellow; and the catechins and leucoanthocyanins which are colorless and readily change to brown color. The color of the anthocyanin results from the structure of the anthocyanidin which is combined with the monosaccharides—glucose, galactose, rhamnose, and occasionally with the pentose molecules. Anthocyanidins usually found in the plant tissue are cyanidin, delphinidin, and pelargonidin of which cyanidin is the most common. Acetylated anthocyanin has remarkable heat stability. However, instability at lower pH range limits its utilization as a colorant, especially in low acidic foods. Ascorbic acid and anthocyanins are also not compatible molecules in terms of stability, when coexist. Degraded product of ascorbic acid causes degradation of anthocyanins. Therefore, delivery of anthocyanins from ascorbate-rich foods is possible when stability and amount of ascorbic acid is assured to be on the higher side such as in Indian gooseberry (*Phyllanthus emblica* L.). Similarly, degradation products of sugars like furfurals also lead to the degradation of anthocyanins.

1.2.1.3 Tannins

Tannins are complex mixtures of polymeric phenols and are also termed as tannic acid or gallotannic acid. The tannins can be divided into two categories: condensed tannins and hydrolyzable tannins. Tannins mainly contribute to the astringency and enzymatic browning reactions of foods. Tannins as anti-nutrient factor can also bind with iron divalent ions and reduces their bioavailability. Tannic acid has also been reported as mordant to color cellulosic fibers and natural dyes.

1.2.1.4 Betalains (E162)

Due to thermal and light sensitivity, betalains are relatively underexploited as bio pigments. They are often confused with anthocyanin pigments due to similar water-soluble nature and color tinge. Natural coexistence of both the pigments in same matrix has never been reported. Although, to date, several types of betalains have been identified, still these pigments have not been scientifically studied much. Interestingly, many plants accumulate betalains, but only two sources namely *Beta*

vulgaris L. and *Opuntia ficus-indica* are approved by the European Union to be used in food as natural colorants. Bio functionality of betalains is mainly governed by its two pigments: purple betacyanins (Latin Beta, beet; Greek kyanos, blue) and yellow betaxanthins (Latin Beta; Greek xanthos, yellow).

1.2.1.5 Quinones and Xanthonenes

Some of them are found in flowering plants while majority of these pigments are available in fungi, bacteria, and algae. Quinones are mainly used in the synthesis of artificial colorants/dyes. A derivative of quinoline yellow (E104) has been permitted as food colorant. However, the best example of xanthonenes is the pigment present in mango (*Mangifera indica*) known as mangiferin.

1.2.1.6 Carotenoids

Lipid soluble hydrocarbons are known as carotenoids which upon oxygenation get converted into xanthophylls. Similar to flavonoids, they are also widely present in plant kingdom. Along with plant carotenoids, they are also present in animal foods including milk, egg yolk, and some fish. Carotenoids are important, from the standpoint of human nutrition due to its bio-conversion into vitamin A. Beta carotene, found in milk, potato, carrot, and pumpkin is a precursor of vitamin A yielding two molecules of vitamin A. Lutein [E161(b)] of marigold, lycopene [E160(d)] of tomato and guava, and bixin in annatto color are the examples of carotenoids molecules. Trans-configuration of carotenoids is the stable form. A well-accepted pigment from saffron (*Crocus sativus*) known as “kesar” is chemically a carotenoid pigment, crocin, imparts a rich golden-yellow hue to dishes and textiles. Carotenoids are oxygen, light, and heat-sensitive therefore hot pulping/break of tomato is always recommended for better lycopene retention over cold pulping/break. Acidic environment at high temperature facilitates the conversion of respective trans form of carotenoids to cis form.

1.2.1.7 Curcuminoids/curcumin (E100)

Curcuminoids namely curcumin are used for the enhancement of color and additionally for prolonged storage period of food products due to its antimicrobial capacity. Curcuminoids has been used as a color enhancer for pineapple slices from turmeric (*Curcuma longa* L.). Several physiological active roles of curcuminoids have also been reported such as antioxidant, anti-inflammatory, antidiabetic, antibacterial, and anticancer properties.

1.2.2 Cereals, legumes, fruits and vegetables, algae

1.2.2.1 Cereals

Cereals are seeds of the grass family. The most commonly used cereals are: rice, wheat, maize (corn), and millets such as jowar, bajra, and ragi. Cereals are inexpensive and rich sources of carbohydrates. They contain approximately 65–75 per cent carbohydrates. Rice and vermicelli contain about 78 per cent carbohydrates. Cereal grains are the major staple food in many countries in the world. Composition and Structure As shown in Figure 29, whole cereal grains are composed of an outer bran coat, a germ, and a starchy endosperm.

The *outer chaffy* coat that covers the kernel during growth is eliminated when the grains are harvested. The outer layers of the kernel, which are called Bran, constitute 5 per cent of the kernel. During milling, the bran is discarded. The bran has a high content of fibre and minerals. It is also a good source of thiamin and riboflavin (vitamins of the B-complex group).

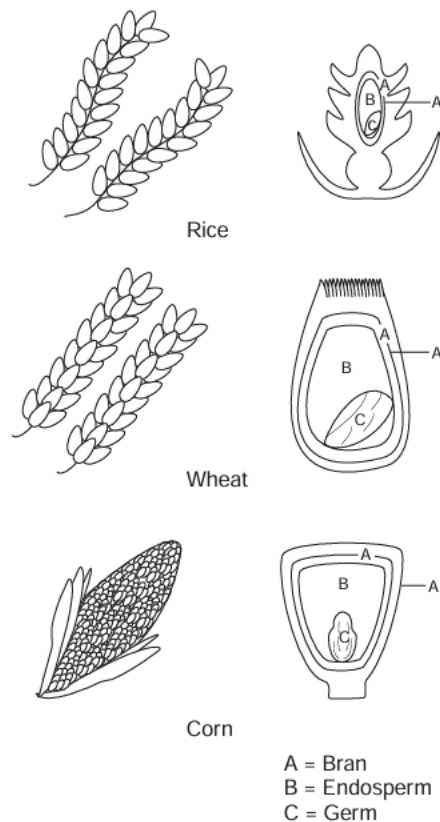


Figure 29: Structural Parts of Rice, Wheat and Corn Grains

The **aleurone** layer is a layer located just under the bran. These cells are rich in protein, phosphorus and thiamin. They also contain some fat. The aleurone layer makes up about 8 per cent of the whole kernel. This layer is also lost in the milling process along with the bran.

The **Endosperm** is the large central portion of the kernel and constitutes about 84–85 per cent of the kernel. It contains most of the starch and protein of the kernel, but very little mineral matter or fibre, and only a trace of fat. The vitamin content of the endosperm is low.

The **Germ** is a small structure at the lower end of the kernel. It makes up 2–3 per cent of the whole kernel. It is rich in fat, protein, minerals and vitamin. The germ serves as a store of nutrients for the seed when it germinates. During milling, some of the germ is lost along with the bran and aleurone layer.

1.2.2.2 Legumes

The term legume encompasses more than 13,000 different species, all of the family Leguminosae. Legumes play a dominant role in the diets of humans across the globe. Of the thousands of species, however, only relatively few are widely grown commercially: soybeans, peanuts, dry beans, peas, broadbeans, chickpeas, and lentils. Of these seven, soybean is by far the most widely produced. Many other species of legumes play an important role in local food production in various corners of the world, but they are too numerous to be discussed in this chapter. Legumes are perhaps best known for their high plant protein content, due to nitrogen fixation allowed by the symbiotic relationship with bacteria.

Despite great variation in the macronutrient composition of legumes, their basic seed structure is the same. Mature seeds contain three major components: the seed coat (testa), the embryo, and the endosperm. Most legume seeds, however, have very little endosperm at maturity, as the cotyledons of the embryo make up a majority of the seed weight and contain the necessary stores for growth. Thus, the cotyledons provide the great majority of the nutritional components of interest to food value, with the exception of fiber and calcium, of which a significant portion is found in the seed coat. The structure of a typical soybean seed is shown in Figure 30. Size, shape, color, and thickness of the seed coat vary among the different legumes, although the basic structure prevails.

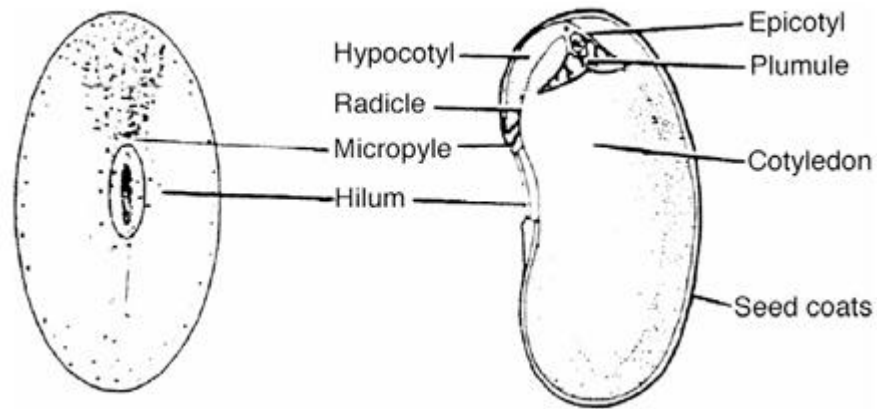


Figure 30: Structure of a soybean seed.

The protein content of legumes has been reported to range between 15% and 45%, with some soybean varieties containing as much as 50% protein. Carbohydrate content ranges from 24% to 68%, and appears to be inversely related to the lipid content. Legume seeds high in carbohydrates have low lipid content, and vice versa. A classical example is peanut, which has a very high lipid content (49.24%) and relatively low carbohydrate content (16.13%). Potassium is by far the most abundant mineral in most food legumes, with soybeans containing as much as 1.80 g/ 100g edible portion. Phosphorus, copper, iron, calcium, and magnesium are some of the important minerals found in significant amounts in legumes. Niacin and pantothenic acid account for the most quantitatively important vitamins in legumes, and most are also a good source of folate

1.2.2.3 Fruits

Fruits include both true fruits and spurious fruits, as well as seeds of cultivated and wild perennial plants. Fruits are commonly classified as pomaceous fruits, stone fruits, berries, tropical and subtropical fruits, hard-shelled dry fruits and wild fruits. Fruit composition can be strongly influenced by the variety and ripeness, thus data given should be used only as a guide. Table 6 shows that the dry matter content of fruits (berries and pomme, stone, citrus and tropical fruits) varies between 10–20%. The major constituents are sugars, polysaccharides and organic acids, while N-compounds and lipids are present in lesser amounts. Minor constituents include pigments and aroma substances of importance to organoleptic quality, and vitamins and minerals of nutritional importance. Nuts are highly variable in composition. Their moisture content is below 10%, N-compounds are about 20% and lipids are as high as 50%.

Table 6: Chemical composition and physicochemical characteristics of selected fruits.

Fruit	Dry Matter (%)	Total Sugar (%)	Titrateable Acidity (%)	Dietary Fiber (%)	Pectin (%)	Ash (%)
Apple	16.0	11.1	0.6	2.1	0.6	0.3
Pear	17.5	12.4	0.2	3.1	0.5	0.4
Apricot	14.7	8.5	1.4	1.6	1.0	0.6
Sour Cherry	14.7	9.9	1.8	1.0	0.3	0.5
Sweet Cherry	17.2	13.3	1.0	1.3	0.3	0.6
Peach	12.9	8.5	0.6	1.9	0.5	0.5
Plum/Prune	16.3	10.2	1.5	1.6	0.9	0.5
Blackberry	15.3	6.2	1.7	3.2	0.5	0.5
Strawberry	10.2	5.7	1.1	1.6	0.5	0.5
Red Currant	15.3	4.8	2.3	3.5	0.9	0.6
Black Currant	18.7	6.3	2.6	6.8	1.7	0.8
Raspberry	15.5	4.5	2.1	4.7	0.4	0.5
Grapes	18.9	15.2	0.9	1.5	0.3	0.5
Orange	14.3	8.3	1.1	1.6	0.5	—
Grapefruit	11.4	7.4	1.5	1.6	0.4	—
Lemon	9.8	3.2	4.9	0.5	—	—
Pineapple	15.4	12.3	0.7	1.0	0.4	—
Banana	26.4	20.0	0.6	1.8	0.9	0.8
Cherimoya	25.9	13.0	0.2	0.9	—	—
Date	80.0	65.1	1.3	8.7	1.8	—
Fig	20.0	13.0	0.4	2.0	0.6	0.7
Guava	17.0	5.8	0.9	5.2	0.7	0.7
Mango	19.0	12.5	0.3	1.7	0.5	0.5
Papaya	11.0	7.1	0.1	1.7	0.6	0.6

1.2.2.4 Vegetables

Vegetables are defined as the fresh parts of plants which, either raw, cooked, canned or processed in some other way, provide suitable human nutrition. Fruits of perennial trees are not considered to be vegetables. Ripe seeds are also excluded (peas, beans, cereal grains, etc.). From a botanical point of view, vegetables can be divided into algae (seaweed), mushrooms, root vegetables

(carrots), tubers (potatoes, yams), bulbs and stem or stalk (kohlrabi, parsley), leafy (spinach), inflorescence (broccoli), seed (green peas) and fruit (tomato) vegetables. The composition of vegetables can vary significantly depending upon the cultivar and origin. Table 7 shows that the amount of dry matter in most vegetables is between 10 and 20%. The nitrogen content is in the range of 1–5%, carbohydrates 3–20%, lipids 0.1–0.3%, crude fiber about 1%, and minerals close to 1%. Some tuber and seed vegetables have a high starch content and therefore a high dry matter content. Vitamins, minerals, flavor substances and dietary fibers are important secondary constituents.

Table 7: Average composition of vegetables (as % of fresh edible portion)

Category	Vegetable	Dry Matter (%)	Protein (N×6.25) (%)	Available Carbohydrates (%)	Lipids (%)	Dietary Fiber (%)	Ash (%)
Mushrooms	Champignon (cultivated <i>Agaricus arvensis/campestris</i>)	9.0	4.1	0.6	0.3	2.0	1.0
Mushrooms	Chanterelle	8.5	2.6	0.2	0.5	3.3	1.6
Mushrooms	Edible boletus (<i>Boletus edulis</i>)	11.4	5.4	0.5	0.4	6.0	0.9
Rooty Vegetables	Carrots	11.8	1.1	4.8	0.2	3.6	0.8
Rooty Vegetables	Radish (white fleshy root)	7.0	1.0	2.4	0.2	2.5	0.8
Rooty Vegetables	Viper’s grass (<i>Scorzonera</i>)	23.2	1.4	2.2	0.4	18.3	1.0
Rooty Vegetables	Parsley	16.1	2.9	6.1	0.6	4.9	1.6
Tuberous Vegetables	White (Irish) potato	22.2	2.0	14.8	0.1	2.1	1.1
Tuberous Vegetables	Celery (root)	11.6	1.6	2.3	0.3	4.2	1.0
Tuberous Vegetables	Kohlrabi	8.4	2.0	3.7	0.2	1.4	1.0
Tuberous Vegetables	Rutabaga	10.7	1.1	5.7	0.2	2.9	0.8
Tuberous Vegetables	Radish (reddish fleshy root)	5.6	1.1	2.1	0.1	1.6	0.9
Tuberous Vegetables	Red beet (beetroot)	13.8	1.6	8.4	0.1	2.5	1.1
Tuberous Root Vegetables	Sweet potato	30.8	1.6	24.1	0.6	3.1	1.1
Tuberous Root Vegetables	Cassava (manioc)	36.9	0.9	32.0	0.2	2.9	0.7

Category	Vegetable	Dry Matter (%)	Protein (N×6.25) (%)	Available Carbohydrates (%)	Lipids (%)	Dietary Fiber (%)	Ash (%)
Tuberous Root Vegetables	Yam	31.1	2.0	22.4	0.1	5.6	1.0
Bulbous Root Vegetables	Onion	11.4	1.2	4.9	0.3	1.8	0.6
Bulbous Root Vegetables	Leek	12.1	2.2	3.3	0.3	2.3	0.9
Bulbous Root Vegetables	Vegetable fennel	7.6	1.4	3.0	0.2	2.0	1.0
Stem (Shoot) Vegetables	Asparagus	6.5	1.9	2.0	0.2	1.3	0.6
Leafy (Stalk) Vegetables	Rhubarb	7.3	0.6	1.4	0.1	3.2	0.6
Leafy Vegetables	Endive (escarole)	5.6	1.3	2.3	0.2	1.3	0.8
Leafy Vegetables	Kale (curly cabbage)	14.1	4.3	2.5	0.9	4.2	1.5
Leafy Vegetables	Head lettuce	5.1	1.2	1.1	0.2	1.4	0.9
Leafy Vegetables	Brussels sprouts	15.0	4.5	3.3	0.3	4.4	1.2
Leafy Vegetables	Red cabbage	9.0	1.5	3.5	0.2	2.5	0.7
Leafy Vegetables	Spinach	8.5	2.6	0.6	0.3	2.6	1.5
Leafy Vegetables	Common (white) cabbage	9.6	1.3	4.2	0.2	3.0	0.7
Flowerhead (Calix) Vegetables	Artichoke	17.5	2.4	2.6	0.1	10.8	1.3
Flowerhead (Calix) Vegetables	Cauliflower	9.0	2.5	2.3	0.3	2.9	0.9
Flowerhead (Calix) Vegetables	Broccoli	10.9	3.6	2.7	0.2	3.0	1.1

1.2.2.5 Algae

More than 9 million tons (fresh weight) of macroalgae are processed worldwide each year. The largest market for seaweed, both in terms of economic value and production volume, is direct human consumption, which accounts for approximately 75% of global output. Seaweed has long been a staple food in many Southeast Asian countries, where its nutritional benefits have been recognized and utilized for generations.

The nutritional significance of seaweeds is largely attributed to their rich composition of dietary fibers, minerals, and proteins. In addition, they contain a wide range of bioactive compounds with

antioxidant properties, including carotenoids, polyphenols, vitamins, and polyunsaturated fatty acids, all of which contribute to their potential health-promoting effects.

Marine algae are particularly valued for their exceptional mineral content, as they absorb and concentrate numerous elements from seawater. The mineral fraction can account for up to 36% of their dry weight. These organisms provide a diverse array of essential minerals such as calcium, sodium, magnesium, potassium, phosphorus, iodine, iron, and zinc.

Protein levels in algae vary considerably among species. Some microalgae, such as *Spirulina*, may contain up to 70% protein on a dry-weight basis. Among macroalgae, several red seaweed species possess protein contents ranging from 30% to 40% of dry matter, values that are comparable to those found in many leguminous plants.

A distinctive feature of red and blue-green algae is the presence of phycobiliproteins, specialized pigments involved in light harvesting during photosynthesis. Important examples include phycocyanin from *Spirulina* and phycoerythrin from red algae. Beyond their role as pigments, these compounds exhibit strong antioxidant activity and are increasingly being investigated for their potential contribution to the prevention or management of oxidative stress-related disorders, including certain cancers, cardiovascular diseases, and eye conditions.

Red seaweeds are also recognized as valuable sources of long-chain polyunsaturated fatty acids. In species such as *Porphyra* and *Palmaria palmata*, the omega-3 fatty acid eicosapentaenoic acid (EPA) can represent nearly half of the total polyunsaturated fatty acid content.

Furthermore, most marine algae contain a unique group of polyphenolic compounds known as phlorotannins. These molecules are characterized by considerable structural diversity, which is associated with a broad spectrum of potential biological activities. Brown algae are particularly rich in phlorotannins, with concentrations generally ranging from 5% to 15% of their dry weight, making them one of the most important natural sources of these bioactive compounds.

1.3 Animal-Based Food Systems

1.3.1 Muscle

Meat is defined as the flesh of animals used as food. A more precise definition is provided by the US Food and Drug Administration (Meyer, 1964): meat is that derived from the muscles of animals closely related to man biochemically and therefore of high nutritive value. The more conventional

animal species include cattle, sheep, and the avian species chicken and turkey. In fish, however, it is often the white muscle that provides the main nutritional source. Seafood, particularly fish, accounts for approximately 20% of all animal proteins consumed worldwide. In the developing continents, Africa, Asia, and Latin America, the consumption of meat and fish is still extremely low or non-existent, as evidenced by the increasing incidence of malnutrition. This lack of high-grade proteins and the accompanying deficiency in essential amino acids still remains the world's most urgent problem. Following the death of the animal or fish, many chemical, biochemical, and physical changes occur leading to the development of postmortem tenderness. A greater understanding of these changes should make an important contribution to the production of high-quality meat or fish products. While the degradation of myofibrillar and cytoskeleton proteins is desirable for postmortem tenderization of mammalian and avian muscles, such changes can lead to unfavorable changes in fish muscle. Texture remains one of the most important quality attributes affecting consumer acceptance of meat and fish products.

Meat is a good source of proteins and fat. The proportion of nutrients in meat depend upon the kind of animal, the species and the type of cut. The protein content of meat decreases with an increase in fat content. The average protein content of meat ranges from 16 to 23 per cent and the average fat content ranges from 10 to 40 per cent. Meat is a good source of phosphorus, iron and some trace elements. Unless the cooking water is discarded, minerals and water soluble vitamins are not lost to a great extent. Meat provides us vitamin B-complex and some vitamin A, depending on the cut.

1.3.2 Eggs

Eggs have been a human food since ancient times. They are one of nature's nearly perfect protein foods and have other high quality nutrients. Eggs are readily digested and can provide a significant portion of the nutrients required daily for growth and maintenance of body tissues. They are utilized in many ways both in the food industry and the home. Chicken eggs are the most important. Those of other birds (geese, ducks, plovers, sea gulls, quail) are of lesser significance.

The egg (Figure 31) is surrounded by a 0.2-0.4 mm thick calcareous and porous shell. Shells of chicken eggs are white-yellow to brown, duck's are greenish to white, and those of most wild birds are characteristically spotted. The inside of the shell is lined with two closely adhering membranes (inner and outer). The two membranes separate at the large end of the egg to form an air space, the so-called air cell. The air cell is approx. 5mm in diameter in fresh eggs and increases in size during

storage, hence it can be used to determine the age of eggs. The egg white (albumen) is an aqueous, faintly straw-tinted, gel-like liquid, consisting of three fractions that differ in viscosity. The inner portion of the egg, the yolk, is surrounded by albumen. A thin but very firm layer of albumen (chalaziferous layer) closely surrounds the yolk and it branches on opposite sides of the yolk into two chalazae that extend into the thick albumen. The chalazae resemble two twisted rope-like cords, twisted clockwise at the large end of the egg and counterclockwise at the small end. They serve as anchors to keep the yolk in the center. In an opened egg the chalazae remain with the yolk. The germinal disc (blastoderm) is located at the top of a clubshaped latebra on one side of the yolk. The yolk consists of alternate layers of dark- and light-colored material arranged concentrically. The average weight of a chicken egg is 58g. Its main components are water (~74%), protein (~12%), and lipids (~11%). The proportions of the three main egg parts, yolk, white and shell, and the major ingredients are listed in Table 8.

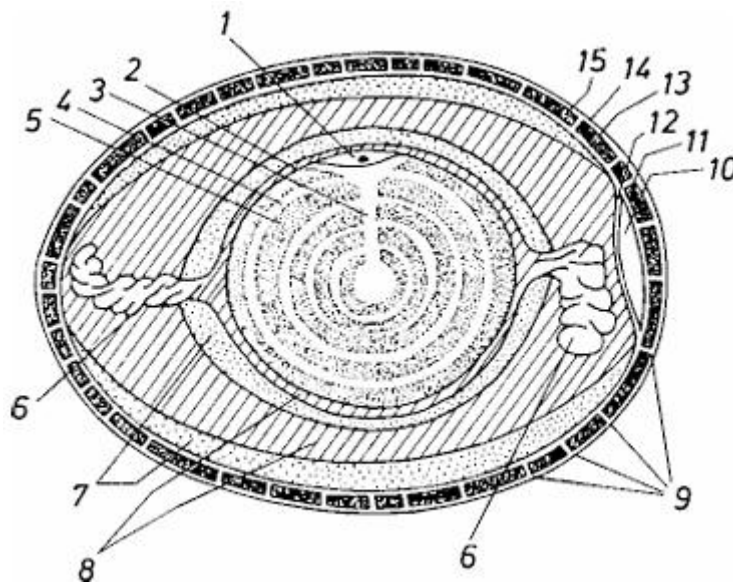


Figure 31: Cross-section of a chicken egg– a schematic representation. Egg yolk: 1 germinal disc (blastoderm), 2 yolk membrane, 3 latebra, 4 a layer of light colored yolk, 5 a layer of dark colored yolk, 6 chalaza, 7 egg white (albumen) thin gel, 8 albumen thick gel, 9 pores, 10 air cell, 11 shell membrane, 12 inner egg mem brane, 13 shell surface cemented to the mammillary layer, 14 cuticle, and 15 the spongy calcareous layer.

Table 8: Average composition of chicken eggs

Fraction	Percentage of Total Weight (%)	Dry Matter (%)	Protein (%)	Fat (%)	Carbohydrates (%)	Minerals (%)
Shell	10.3	98.4	3.3	0.0	0.0	95.1
Egg White	56.9	12.1	10.6	0.03	0.9	0.6
Egg Yolk	32.8	51.3	16.6	32.6	1.0	1.1

1.3.3 Milk

Milk is the fluid secreted by female mammals for the purpose of nourishing their offspring. Milk is considered as nearly complete human food which can be generally consumed without further processing. Although it is very essential for infant feeding, milk and milk products are important in our diet throughout life. Milk may be defined as the whole, fresh, clean, lacteal secretion obtained by complete milking of one or more healthy milch animals, excluding that obtained within 15 days before parturition and 15 days after calving or such periods as may be necessary to render the milk practically colostrum-free and containing the minimum prescribed percentages of milk fat and milk solids-not-fat (SNF). Milk is a complex, nutritious product that contains more than 100 substances that are either in emulsion, suspension, or solution in water. Various factors like genetic factors, stage of lactation, health status of the animal, and environmental factors are responsible for wide variation in milk compositions. The various fermented products (e.g., sour milk, yogurt, curd, dahi, lassi, shrikhand, kefir, kumiss, cheese), concentrated milks (e.g., evaporated milk and sweetened condensed milk), dried milk, fat-rich dairy products (e.g., cream, butter, ghee), ice cream, heat-desiccated products (e.g., khoa, rabdi, basundi), and heat-acid coagulated products (e.g., paneer and chhana) are some of the typical milk products having different chemical compositions (table 9).

Table 9: Gross composition of milks of selected species

Species	Water (%)	Fat (%)	Protein (%)	Lactose (%)	Ash (%)
Buffalo	84.20	6.60	3.90	5.00	0.70
Cow	86.30	4.90	3.40	4.80	0.70
Sheep	83.70	6.00	5.00	4.50	0.80
Goat	86.50	4.50	3.20	4.70	0.80
Camel	87.61	5.38	3.75	4.90	0.70
Human	87.43	3.50	2.98	6.98	0.20

1.4 Unconventional Food Systems

Unconventional foods are produced either from chemical compounds (carbohydrates, organic chemicals) through microbial, enzymatic, or chemical synthesis, or from existing natural products modified physically, chemically, or biologically. To be viable, they must offer nutritional value, functional food properties, and meet regulatory safety standards — free from toxins, mutagens, and pathogens

1.4.1 Alternative Protein Sources

Traditional protein sources face growing pressure, driving interest in alternatives such as single-cell organisms, aquatic species, and insects — all offering amino acid profiles comparable to conventional animal proteins, with digestibility ranging from 77.9% to 98.9%.

- ↳ Microorganisms (bacteria, yeasts, algae, molds) can yield biomass with 20–80% protein content by dry weight. They are rich in lysine and B vitamins, and minerals like zinc, phosphorus, magnesium, and selenium. However, they are deficient in sulfur-containing amino acids (methionine, cysteine), and their nucleic acid content (2–18%) poses a risk of gout if consumed in excess.
- ↳ Algae and microalgae are sustainable, non-competing protein sources containing protein levels comparable to meat, eggs, soy, and milk. They are also rich in amino acids, minerals, and vitamins, with negligible fat and cholesterol, offering benefits such as blood pressure reduction and stroke prevention.
- ↳ Krill (notably Antarctic krill, *Euphausia superba*) provides 60–65% protein by dry weight, making it a high-quality complete protein source.
- ↳ Fish protein concentrates (FPC) and isolates (FPI) must contain a minimum of 75% and 90% protein respectively, with very low fat and moisture. Their amino acid profiles are excellent, comparable to whole egg, with the exception of tryptophan and lysine.

1.4.2 Unconventional Lipid Sources

Several non-traditional oilseed plants offer promising nutritional and functional properties:

- ✓ *Moringa oleifera* : Contains ~40% oil, with over 70% oleic acid, making it resistant to oxidative degradation and a viable substitute for olive oil. It also offers cardiovascular and anti-inflammatory health benefits.
- ✓ *Nigella sativa* : Contains 32–53% oil (85% unsaturated fats), widely used in Middle Eastern and Indian cuisines.
- ✓ Avocado (*Persea americana*): About 70% of its oil consists of oleic acid (omega-9), supporting cardiovascular health by raising HDL cholesterol and reducing triglycerides and LDL levels. It also enhances carotenoid absorption.

1.4.3 Biomass

Biomass refers to all matter of living origin. Lignocellulosic biomass (agricultural, forestry, and industrial residues) is increasingly valued as a renewable, cost-effective resource to reduce dependence on petroleum. Beyond energy production, biomass can yield valuable bio-based products including:

- ↪ Sugar alcohols (e.g., xylitol — a low-calorie, anticariogenic sugar substitute)
- ↪ Lactic acid: used in food, pharmaceuticals, and biodegradable polymers
- ↪ Ethyl lactate: a biodegradable solvent replacing petroleum-derived volatile compounds
- ↪ Bioplastics, resins, cellulose derivatives, and carbon-based materials

Chapter 6: Food Spoilage

1.1 Role of water

Water is essential for microbial cells, serving key functions such as maintaining cellular structure, transporting metabolites, and eliminating waste. In food products, water exists in two forms: free water, which supports microbial growth, and bound water, which sustains biological functions.

Microorganisms regulate their internal water levels through a process called osmoregulation, which is critical for cell growth. When free water in the environment is reduced by adding solutes like salts or sugars water inside the cells moves outward to restore equilibrium. This leads to plasmolysis and osmotic shock, ultimately inhibiting microbial growth. Even a slight decrease in water activity (a_w) of just 0.005 (from 0.955 to 0.95) can reduce intracellular water by ~50% and cell volume by ~45%, highlighting how sensitive microbes are to small a_w changes.

Some microorganisms have developed adaptive mechanisms to survive these conditions. They counteract plasmolysis by absorbing solutes such as amino acids and polyols from their surroundings to restore turgor pressure, allowing them to tolerate reduced a_w levels to a certain extent.

Water activity has its major practical relevance in forecasting the development of bacteria, molds, and yeasts. For a food product to encompass a functional shelf life devoid of depending on refrigerated storage space, it is essential to manage either its acidity level or the value of (a_w) or an appropriate arrangement of the above two properties. This might successfully augment the product's constancy and compose its potential to envisage the shelf life under recognized ambient storage space surroundings

1.2 Main Food Preservation Technologies

1.2.1 Heat preservation (thermal processes)

High temperatures destroy microorganisms and inactivate enzymes. However, heat also triggers chemical reactions that alter texture, flavor, color, and nutritional value. Desirable changes constitute "cooking"; undesirable ones are called "thermal damage." Understanding thermal kinetics is essential for optimizing these processes.

1.2.2 Low-temperature preservation

Cold slows microbial activity and chemical reactions without fully destroying microorganisms or enzymes — their activity simply resumes once temperature rises again.

1.2.3 Reduction of water activity

Microorganisms cannot grow below a minimum water activity threshold. Drying, concentrating foods, and adding solutes like salt or sugar are all based on this principle.

1.2.4 Ionizing radiation

Capable of destroying microorganisms and inactivating enzymes, this powerful technique holds significant potential for solving distribution and production challenges in the food industry.

1.2.5 Chemical preservation

Salting and smoking are among the oldest methods, using chemical compounds to inhibit microbial growth. Acidification (via vinegar, citric acid, lactic acid, or fermentation) prevents pathogen growth. Synthetic preservatives such as sulfur dioxide, benzoic acid, and sorbic acid are also used, subject to food safety regulations.

1.2.6 Packaging

Packaging acts as a protective barrier against contamination and controls the exchange of moisture, oxygen, and odorous substances between food and its environment. **Active packaging** containing embedded preservatives like antioxidants is a recent and growing innovation.

1.3 Types of Microbial, Enzymatic, and Chemical Alterations

1.3.1 Microbial Alterations

Most foods naturally carry microorganisms (with few exceptions, such as the interior of fresh eggs). Some are beneficial to food production; others are harmful or even dangerous to human health (table 10). Microbial contamination can cause:

- ✓ **Infectious diseases** — pathogens multiply within the host (e.g., typhoid fever, cholera, dysentery).
- ✓ **Toxi-infections** — microorganisms produce specific toxins in large quantities before consumption.
- ✓ **Intoxications** — caused by exotoxins already produced in the food; the microorganism itself need not be present in the body.

Key factors influencing microbial growth include **pH, temperature, water activity, and redox potential**. Spoilage becomes noticeable at counts above 10^7 bacteria/g or 10^5 yeasts/g.

Table 10: Biochemical changes in food

Component	Alteration Mechanism	Effects
Carbohydrates	Polysaccharide hydrolysis, fermentation	Texture changes, production of acids, alcohols, ketones, off-flavors
Proteins	Hydrolysis, decarboxylation, deamination	Texture degradation, amine and ammonia formation, putrefaction
Lipids	Lipolysis, oxidation	Release of fatty acids, rancidity

1.3.2 Sensory Consequences

Microbial degradation progressively alters:

- ✓ **Odor** — due to compounds such as trimethylamine, hydrogen sulfide, ammonia, and butyric acid.
- ✓ **Color** — colored zones appear on surfaces due to pigment synthesis or destruction of natural pigments (carotene, myoglobin, polyphenols).
- ✓ **Taste** — typically characterized by increased sourness.
- ✓ **Texture and structure** — breakdown of macromolecules like pectin, hemicellulose, and proteins, or synthesis of new ones (e.g., dextran).
- ✓ **Impact on Nutritional Value**

Microbial spoilage can either improve or, more commonly, **diminish nutritional value** through:

- ✓ Reduction in caloric content (negative)
- ✓ Synthesis of biologically active molecules (variable)
- ✓ Destruction of toxic or anti-nutritional compounds (positive)
- ✓ Putrefaction (negative)

1.3.3 Enzymatic alterations in food spoilage

Foods naturally contain a variety of compounds, simple sugars, amino acids, fatty acids, and organic acids that microorganisms eagerly exploit. To do so, bacteria and other microbes release

intracellular enzymes that break down these compounds, pulling them into their cells and triggering visible food spoilage in the process.

One often-overlooked problem is what happens after microbial cells die. When cells lyse (burst open), they spill their enzymes into the food. Even if heat treatment kills all the microorganisms, many of their enzymes particularly from psychrotrophic bacteria are heat-stable enough to survive and keep working. Proteinases, lipases, phospholipases, and amylases are the main culprits here, quietly breaking down nutrients during storage long after the bacteria themselves are gone.

Beyond microbial sources, enzymes naturally present in food can also drive spoilage. A classic example is orange juice: an enzyme called pectin methyl esterase demethylates pectin, causing the juice to separate something manufacturers want to avoid. The tricky part is that heating the juice to knock out PME also damages its delicate flavor, so it's not a simple fix.

The two main enzyme families responsible for food degradation are hydrolases, which break chemical bonds using water, and oxidoreductases, which drive oxidation-reduction reactions. Amylases, for instance, chop up starch into smaller sugars, while lipoxygenases trigger the oxidative rancidity that gives vegetable oils and fats that unpleasant off-flavor when they go bad.

To keep these enzymes in check, several methods are used heat treatment, vacuum packaging, acidification (vinegar or citric acid), sulfites, blanching, and even irradiation each targeting enzymatic activity in a different way.

1.3.4 Chemical Deterioration in Food

Chemical deterioration in food happens through two main pathways: reactions driven by enzymes, and reactions that occur without enzymes at all. Generally speaking, enzyme-driven reactions tend to dominate in raw, unprocessed plant and animal tissues, while non-enzymatic reactions take over once food has been properly processed or cooked.

That said, both types can sometimes work side by side. A good example is the surface discoloration of red meat. The color change itself is a non-enzymatic reaction oxygen oxidizes myoglobin (which contains iron in its Fe^{2+} form) into metmyoglobin (Fe^{3+}), turning the meat brown. At the same time, enzymatic activity at the tissue surface consumes oxygen through cellular respiration, which actually encourages more of that non-enzymatic oxidation to happen. Some muscle tissue even

contains an enzyme called metmyoglobin reductase, which works in the opposite direction — converting metmyoglobin back into myoglobin and restoring the red color.

When food has been heat-treated or otherwise processed in a way that destroys or deactivates enzymes, non-enzymatic reactions become the main driver of spoilage. The two most significant ones are the Maillard browning reaction and lipid oxidation, both of which can seriously affect the appearance, smell, and nutritional value of food during storage.

How fast these reactions happen depends on a range of factors: pH, temperature, the concentration of reacting compounds, the presence of catalysts, the physical state of the food, and even how freely the reacting molecules can move around. Of all these variables, temperature tends to have the greatest influence on how quickly chemical changes unfold in food.

References

1. Anklam, E. (2005). H.-D. Belitz, W. Grosch, P. Schieberle: *Food Chemistry*.
2. Belhachemi, M. H. (2022). *cours de biochimie nutritionnelle: 3 année lmd biochimie*.
3. Boeckel, T. P. V., Hounhouigan, J. D., & Nout, R. (2003). *Les aliments: transformation, conservation et qualité*. Technical Centre for Agricultural and Rural Cooperation.
4. Chauhan, O. P. (2022). *Advances in Food Chemistry*. Singapore: Springer. <https://doi.org/10.1007/978-981-19-4796-4>.
5. Coultate, T. P. (2009). *Food: the chemistry of its components*. Royal Society of Chemistry.
6. Damodaran, S., Parkin, K. L., & Fennema, O. R. (Eds.). (2007). *Fennema's food chemistry*. CRC press.
7. Dilmi Bouras A. (2020). *Protéines et lipides alimentaires In: Biochimie alimentaire 2, Support du cours pour le Master 1 Sciences alimentaires et Nutrition humaine*. Université de Chlef. 188p. <https://fr.scribd.com/document/595957487/biochimie>
8. Eskin, N. M., & Shahidi, F. (2012). *Biochemistry of foods*.
9. Hadri, Z. (2022). *Biochimie alimentaire*. Université Ahmed Zabana de Relizane.
10. Jamshidi-Kia, F., Saeidi, K., Lorigooini, Z., Samani, B. H., & Barzegar, R. (2024). Optimization of extraction of liquid extract from microalgae *Chlorella vulgaris* via cavitation-based techniques. *Journal of Food Process Engineering*, 47(8), e14665.
11. Jayakody, M. M., Kaushani, K. G., Vanniarachchy, M. P. G., & Wijesekara, I. (2023). Hydrocolloid and water soluble polymers used in the food industry and their functional properties: a review. *Polymer Bulletin*, 80(4), 3585-3610.
12. Li, D., Zhu, Z., & Sun, D. W. (2018). Effects of freezing on cell structure of fresh cellular food materials: A review. *Trends in Food Science & Technology*, 75, 46-55.
13. Maled, S. B., Bhat, A. R., Hegde, S., Sivamani, Y., Muthuraman, A., & Elayaperumal, S. (2024). Enzyme-assisted extraction. In *Bioactive extraction and application in food and nutraceutical industries* (pp. 173-200). New York, NY: Springer US.
14. Mehta, B. M. (2015). Chemical composition of milk and milk products. *Handbook of food chemistry*, 511-553.
15. Mudambi, S. R., Rao, S. M., & Rajagopal, M. V. (2006). *Food science*. New Age International.

16. Nishad, J., Wahi, P., Kumari, S., & Negi, R. (2025). Hydrocolloids as Potential Additives. In *Advances in Pasta Technology* (pp. 239-256). Cham: Springer Nature Switzerland.
17. Rezak H.Y. (2023). *Les corps gras alimentaires analyse physico-chimique*. Polycopie
Cours Hydro-Bromatologie 5eme Année Pharmacie.
<https://fr.scribd.com/document/724335678/les-corps-gras-alimentaires>