

**Paper No: 1**

**Paper Title: Food Chemistry**

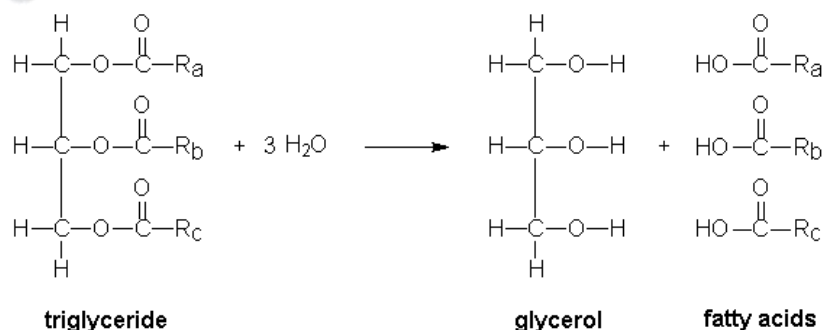
**Module No: 11: Fat Hydrolysis, Interesterification and Hydrogenation**

### 11.0 Introduction

Lipids are recognized as essential nutrients in both human and animal diets. Edible fats and oils (lipids) are derived from plant, animal and marine sources. Fats are solid at normal room temperature whereas oils are liquid under similar conditions. Hydrogenation, interesterification and hydrolysis are distinct processes that can be applied to modify the physical or chemical properties of fats and oils in order to improve their usefulness. These processes can be used alone or in combination with each other. By combining hydrogenation and interesterification with the simple blending of native and modified oils, it is possible to modify the characteristics of fats and oils for specific applications. Hydrogenation is used to convert liquid oils into products having different consistencies, melting points and textures. On the other hand, interesterification produces changes in physical properties by rearrangement or redistribution of fatty acids within and among the triacylglycerides of oils.

### 11.1 Hydrolysis

Hydrolysis of fats and oils refers to the splitting of their triacylglycerides into their constituent fatty acids and glycerol in the presence of water (Figure 11.1).



**Figure 11.1. Fat hydrolysis**

Three distinct routes are used for the hydrolysis of triacylglycerides (TAG) to free fatty acids (FFA) and glycerol: high-pressure steam splitting, alkaline hydrolysis (saponification), and enzymatic hydrolysis. In current industrial practices, fats and oils are hydrolyzed to produce free fatty acids or soaps and glycerol. For production of free fatty acids, fats and oils are hydrolyzed using steam in a countercurrent continuous process operated at high temperature (250°C) and pressure (50–60 bars). This is an efficient process giving an aqueous phase containing 10% glycerol and oil phase containing 97% free fatty acids. As a result of the high temperature used in this process, oils containing highly unsaturated fatty acids undergo decomposition yielding undesirable oxidation products. The products obtained by high-pressure steam splitting are usually dark-colored and, hence, require distillation. High-pressure steam splitting requires high capital investment and energy costs. Alkaline hydrolysis also consumes a large amount of energy and the resulting soap requires acidification to yield free fatty acids. Enzyme-catalyzed hydrolysis of fats and oils offers an alternative process for production of fatty acids and may be performed at ambient or moderate temperatures and pressures. Therefore, this process can be used to obtain fatty acids from oxidatively unstable oils that contain highly unsaturated fatty acids or conjugated fatty acyl residues. Lipase-catalyzed hydrolysis of fats for the production of fatty acids and glycerol appears to be less economical than conventional chemical processes mainly because of the relatively high cost of the lipases. Such processes, however, would be economically attractive for the production of specific value-added products. Hydrolysis of marine and fish oils by lipases for the production of n-3 polyunsaturated fatty acids is a possible application of enzymatic hydrolysis of oils for the preparation of products of reasonable commercial value.

## **11.2 Interesterification**

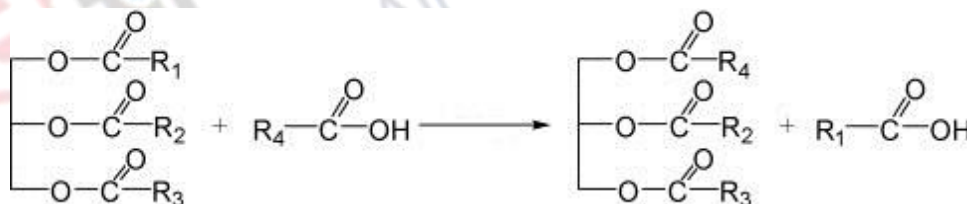
Chemical interesterification is one of the major reactions used for modification of natural fats and oils. It is a process that is used to modify the physical and functional properties of triacylglycerol mixtures in fats and oils. In its simplest

form, interesterification corresponds to an exchange of acyl groups between two triglycerides, resulting in the formation of new triglycerides that have chemical and physical properties, generally superior to those of the starting triglycerides and lead to substantial changes in lipid functionality.

### 11.2.1 Types of interesterification

The term “interesterification” is often used to describe reactions that involve the exchange of acyl residues between an ester and an acid (acidolysis), an ester and an alcohol (alcoholysis), or an ester with another ester (trans-esterification).

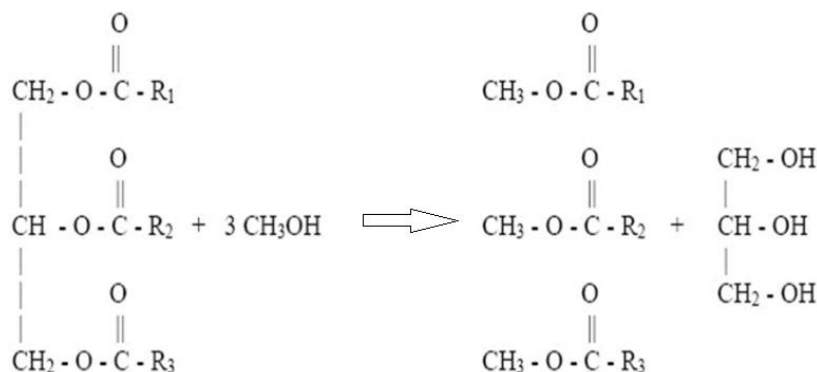
1. **Acidolysis:** Triacylglycerol reacts with organic acids, which is thereby introduced into triglyceride molecule. e.g. coconut oil and stearic acid can be reacted to partially replace the short chain fatty acids of coconut oil with higher melting stearic acid. Acidolysis is an effective means of incorporating novel fatty acids into triglycerides and may be used to incorporate eicosapentenoic acid (20:5, n-3; EPA) and docosahexenoic acid (22:6, n-3; DHA) into vegetable and fish oils to improve their nutritional properties.



**Figure 11.2. General scheme of acidolysis**

2. **Alcoholysis:** Reaction of an ester with an alcohol is known as alcoholysis. The reaction produces free glycerol, triglycerides of altered structure and also mono and diglycerides as well as esters of new alcohol and also partial esters of this alcohol, if it happens to be polyhydroxy. The reaction is commercially important as it offers a mean of preparing fatty acid esters other than glycerides and is more suitable and consistent than

other method of splitting the fat and reesterification. Alcoholysis of TGs with higher polyhydroxy alcohols converts frying oil to product of higher functionality for polymerization.



**Figure 11.3: General scheme of alcoholysis**

- 3. Tranesterification (Ester exchange):** Transterification is the most widely employed type of interesterification reaction used by the food industry. Transterification results in the “shuffling” of fatty acids within a single TAG molecule (intraesterification) and among TAG molecules (interesterification) until an equilibrium is achieved in which all possible combinations are formed. During the reaction, the first step involves the cleavage of ester bonds linking fatty acyl residues to the glycerol backbone. The newly liberated fatty acids form a fatty acid pool that are re-esterified onto a new position, either on the same glycerol (intraesterification) or onto another glycerol (interesterification) molecule. Once the reaction reaches equilibrium, a mixture of disaturated, monosaturated, triunsaturated, and trisaturated TAG components is obtained. Usually, intraesterification occurs at a faster rate than interesterification. It results in beneficial changes in physical characteristics of an oil or fat or blend of oils and fats.

### 11.2.2 Catalysts used for interesterification

Interesterification can proceed without catalysts at high temperature (~300°C), however, the desired results are not obtained, because equilibrium is slowly attained and also other reactions i.e. isomerization, polymerization, decomposition may occur. The addition of catalyst significantly lowers reaction temperature and duration. The concentration of catalysts should be minimum. Catalysts can be divided into 3 groups:

1. **Acids:** Mineral acids, sulfonic acids, cation exchangers and zealites
2. **Metals, their oxides, hydroxides and salts:** Oxides of Zn, Mg, Ca; hydroxides and chlorides of Sn, Zn; acetates and stearates of Ca, Mg; aluminates of alkali metals and anion exchangers
3. **Organometallic compounds:** Acetylacetonates, titanium compounds and metal alkyl aluminates

### 11.2.3 Approaches used for interesterification

#### 1. Random interesterification

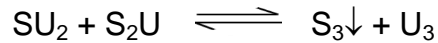
The process involves the formation of an equilibrium mixture of triglycerides formed by a random distribution of fatty acids on glycerol molecules. The process is therefore known as randomization or random esterification. This is usually performed at temperature above the melting point of highest melting point component in the mixture. Random interesterification is entropically driven, until an equilibrium is reached and does not lead to fatty acid selectivity. This may be of two types:

- a. Batch interesterification
- b. Continuous interesterification

#### 2. Directed interesterification

If the equilibrium of reaction mixture is disturbed i.e. interesterification is carried out at temperature below the melting point of highest melting component, the end result will be a mixture enriched in this component, it is then referred to as direct interesterification.

This process is essentially reverse of randomization as it tends to convert mixed triacylglycerides into highly crystallizable, fully saturated triacylglycerides and fully unsaturated TAG.



It is generally used to increase the solid fat content without affecting unsaturated fatty acids. This consists of two reactions which occur simultaneously.

- i. Trisaturate is produced by interesterification, it crystallizes and fall from solution.
- ii. To regain equilibrium, reaction equilibrium in the remaining liquid phase is pushed towards increased production of crystallizing trisaturate

The crystallization continues until all triacylglycerides capable of crystallizing have been eliminated from reaction phase.

### 3. Enzymatic interesterification

The selectivity of enzymatic catalysis can be exploited to generate tailored triacylglycerides that are difficult to obtain by conventional physical or chemical methods and the mild conditions used for enzymatic interesterification enable reactions with unstable material such as polyunsaturated fatty acids (PUFA) to be carried out without generation of by-products associated with use of more severe chemical procedures. Extracellular microbial lipases from bacteria, yeasts or molds are used as catalyst for the interesterification of oils and fats. The enzyme catalyzes the hydrolysis of acylglycerols and other fatty esters to provide nutrients for the microorganisms, but these reactions are easily reversible, and consequently lipases are also effective catalysts for various esterification reactions. The main criteria for selection of lipases are – specificity, activity and stability in microaqueous reaction system and acceptability for use in food processing.

*Rhizpos, Humicola, Rhizomucor, Aspergillus* – catalyze reactions at outer 1, 3 position of glycerol, 2 position remains unaffected



*Candida antarctica*, *Geotrichum candidum* – no significant regioselectivity, random distribution

Microbial lipases vary in their activity with  $a_w$ . Some immobilized lipases have good activity at low  $a_w$  (<0.2), whereas others express high catalytic activity only when  $a_w$  is raised to 0.5. For interesterification, *Rhizomucor miehei* lipase is most preferred.

#### **11.2.4 Applications of interesterification in food lipids**

Chemical interesterification is used industrially to produce modified edible fats and oils for shortenings, confectionery fats, margarines and spreads, reduced-calorie fats and oils and infant formula.

##### **11.2.4.1 Shortenings**

Lard and other animal fats as well as many types of vegetable oils can be used in shortening products. Interesterification is used to improve physical properties of lard. It halves the solid fat content (SFC) of lard at 20°C, prevents development of graininess (due to large proportion of palmitic acid at Sn-2 position; 64%) and improves the plastic range of fat, which makes it a better shortening.

##### **11.2.4.2 Margarines and spreads**

Margarine should have steep solid fat content and should crystallize as  $\beta'$  polymorphs. Chemically interesterified blends of 60% soybean oil and 40% beef tallow show similar properties like margarines. Interesterification has been applied to the production of high-stability margarine blends and hard butters that have desirable melting qualities. Interesterification affects the crystallization behavior of fats and oils. Oils that normally have  $\beta$  crystallization tendencies shift significantly toward  $\beta'$  after the interesterification process. The primary purposes of interesterification are to minimize trans-fatty acid content in margarines and spreads and; to a lesser extent, promote crystallization in the  $\beta'$  form.

##### **11.2.4.3 Cocoa butter alternative fats**

Cocoa butter alternative fats are substances with physical and functional properties similar to cocoa butter. The use of interesterification makes it possible to produce cocoa butter alternatives from other vegetable oils that better mimic the composition and properties of cocoa butter.

#### **11.2.4.4 Milk fat and milk fat substitutes**

Milk fat is a natural fat with unique physical, chemical and biological properties. Directed interesterification generates larger increase in melting point than random interesterification. This also increases the softening point of milk fat by 3.7-4.2°C due to higher proportion of high melting triacylglycerides (5-7%). Interesterified milk fat appears to be better suited for chocolate than non-interesterified milk fat, however, due to higher cost and loss of flavor, it does not have much industrial use for chocolate manufacture.

#### **11.2.4.5 Fat substitutes**

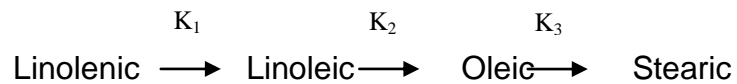
Chemical interesterification includes the production of low calorie fat substitutes such as salatrim and olestra. Salatrim (benefat) is chemically interesterified mixture of short chain and long chain fatty acid triglycerides. Olestra is acylated sucrose polyester obtained by interesterification of sucrose and edible oil methyl esters.

### **11.3 Hydrogenation**

Naturally occurring oils and fats are mainly composed of unsaturated fatty acids. The unsaturated acids contain from one to six double bonds. The number of double bonds in the fatty acids and the triglyceride composition are responsible for the physical state/nature of oils, e.g. liquid or solid at a given temperature and for its susceptibility to oxidation. Hydrogenation of edible fats and oils involves the addition of hydrogen, in presence of a catalyst, to the carbon-carbon double bonds present in the fatty acid chains, resulting in increase in the oxidative stability and melting point of the oil. Hydrogenation is a three-phase reaction in which solid catalyst, gaseous hydrogen and liquid oil interact. During the



hydrogenation of fats and oils, three important reactions occur simultaneously: (1) saturation of double bonds with hydrogen, (2) cis-trans isomerization of double bonds, and (3) migration of double bonds to new positions in the fatty acid carbon chain. A simplified reaction of hydrogenation is shown as:



where,  $K_1$ ,  $K_2$  and  $K_3$  all are reaction rate constants for the sequence. The selectivity ratio for linolenic (ln) to linoleic (l) acid is  $S_{ln} = K_1/K_2$  and for linoleic (l) to oleic (o) acid is  $S_l = K_2/K_3$ .

In a simple system, the reaction proceeds in a step-wise manner because  $S_{ln}$  is greater than  $S_l$ . However, in reality all the steps of the sequence take place simultaneously because of the complexity of triglyceride systems, impurities in the oil and the effect of processing conditions, and the selectivity of the reaction depends on the relative rate at which they occur. Reactions in which PUFA are hydrogenated in preference to MUFA, i.e. high selectivity, are required for improving the oxidative stability of an oil. Process conditions and their effect on isomerization and selectivity are shown in Table 11.1.

**Table 11.1 Process conditions of Hydrogenation and their effects**

Condition (increase in)	Effect on sensitivity	Isomerization
Temperature	Up	Up
Pressure	Down	Down
Agitation	Down	Down
Catalyst concentration	Up	Up
Catalyst activity	Up	Down

In the process of hydrogenation, the oil is mixed with an appropriate catalyst and heated to a suitable temperature and hydrogen is then added at high pressure (up to 60 psi). At elevated temperatures and pressures, the solubility of hydrogen in the oil is increased, thus increasing the rate of the reaction. Only hydrogen, which is dissolved in the oil, is available for the reaction. After the hydrogenation process is completed, the catalyst is removed from the fat by filtration. During contact with the hydrogenation catalyst, isomerization takes place in which either some of the double bonds move to a new position or become transformed from cis to trans form. The trans fatty acids have higher melting points and thus contribute to the increase in melting point of the fat.

The purpose of hydrogenation process is two folds: (1) to convert liquid oils into semisolid or plastic fats for specific food applications, and (2) to improve the oxidative stability of the oil to maintain its organoleptic acceptability as a human food for an extended period of time.

### **11.3.1 Catalysts and hydrogen gas used for hydrogenation**

A number of active metals are capable of catalyzing this reaction, but industrially nickel is used. Since, the catalyst is slowly poisoned during the hydrogenation process, it needs to be reactivated after it has been used for some time. Impurities present in the oil may poison the catalyst i.e. get adsorbed on the surface of the catalyst; 1 ppm of S poisons 0.004% of Ni and 1 ppm of P poisons 0.0007% of Ni etc.

The purity of hydrogen is of great importance in the hydrogenation process. The standard of purity obtained in most cases is better than 99.5% but gases containing only 98.5% hydrogen can be usable depending upon the composition of the impurities in the gas. The impurities can be divided into two types, 1) those which will act as catalyst poisons, e.g. S, Cl and Co, which are adsorbed on the catalyst surface; and oxygen and moisture, the action of which on the oil produces compounds acting as catalyst poisons and 2) others are the inert

gases, e.g. Ne and Ar and methane which are inert in the reaction, however, by accumulating in the headspace of the container, can slow down and eventually stop hydrogenation by restricting the flow of hydrogen.

### **11.3.2 Applications of hydrogenation in food industry**

Main application of hydrogenation is for the processing of vegetable oils. Vegetable oils are generally derived from polyunsaturated fatty acids (containing more than one carbon-carbon double bonds). Hydrogenation converts liquid vegetable oils into solid or semi-solid fats, such as those present in margarine. Changing the degree of saturation of the fat changes some important physical properties such as the melting range, which is why liquid oils become semi-solid. Solid or semi-solid fats are preferred for baking because the way the fat mixes with flour produces a more desirable texture in the baked product. Hydrogenated fats are the predominant fats used as shortening in most commercial baked goods because partially hydrogenated vegetable oils are cheaper than animal source fats, are available in a wide range of consistencies and have other desirable characteristics (increased oxidative stability/longer shelf life).

### **11.3.3 Implications of hydrogenation**

During the process of hydrogenation or partial hydrogenation, some of the unsaturated fatty acids, which are normally found as the cis isomer are changed to their trans isomers. Trans fatty acids are of the same length and weight as the original cis fatty acids but they are shaped in a more linear form, as opposed to the bent forms of the respective cis isomers. Trans fat has both the benefits and drawbacks of a saturated fat. On the positive side, it has a longer shelf life than regular vegetable fat and is solid at room temperature. The major drawback is that trans fat tends to raise "bad" LDL- cholesterol and lower "good" HDL- cholesterol, although not as much as saturated fat. Elevated LDL cholesterol increases the risk of developing coronary heart disease. Trans fat is found in margarine, baked goods such as doughnuts and Danish pastry, deep-fried foods like fried chicken and French-fried potatoes, snack chips, imitation cheese, and

confectionary fats. It has long been recognized that monounsaturated (one double bond) and polyunsaturated fats (two or more double bonds) found primarily in vegetable oils tend to lower "bad" LDL cholesterol and saturated fats tend to increase the blood level of the "bad" LDL cholesterol.

### **Suggested Readings**

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