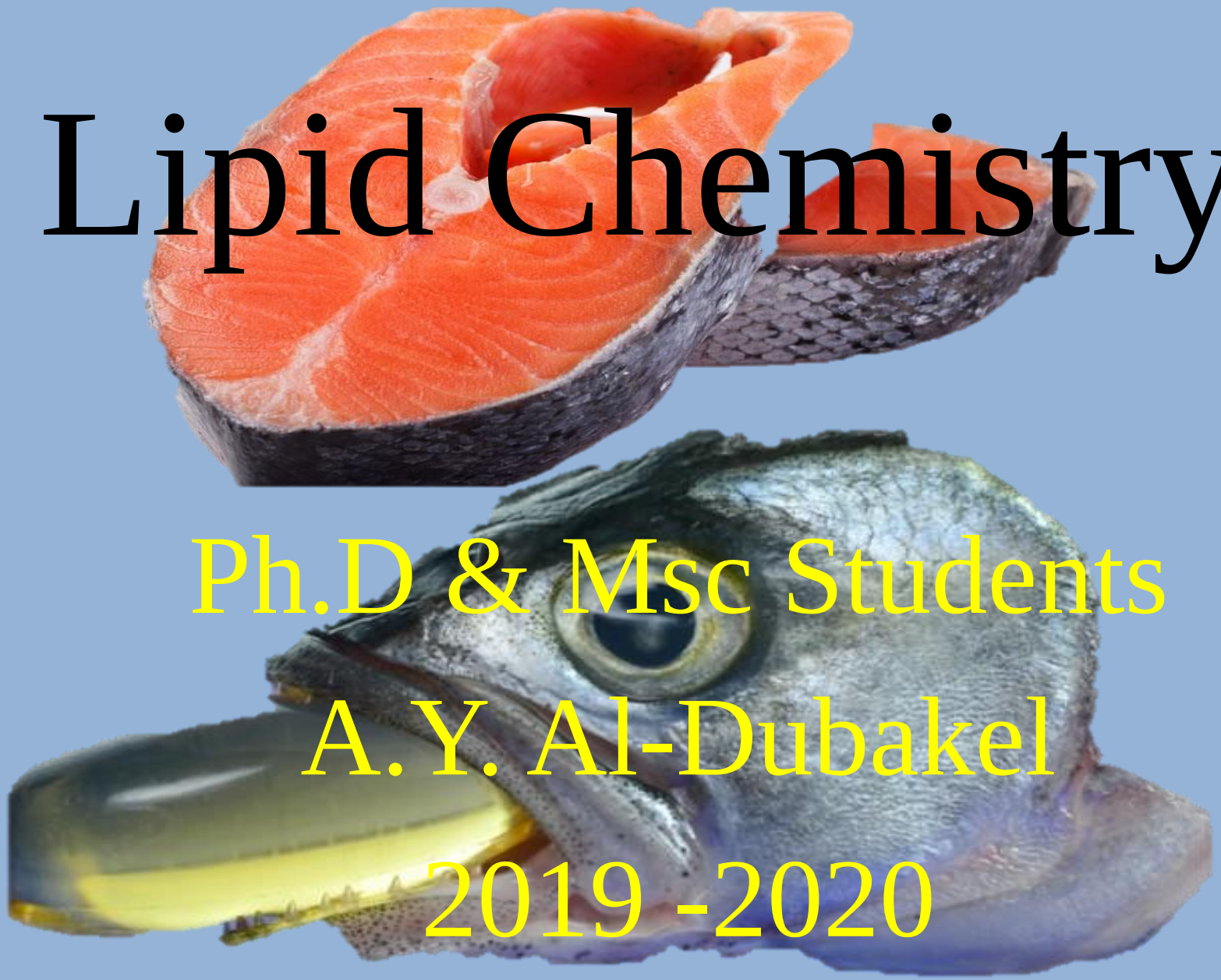
Lipid Chemistry



Ph.D & Msc Students

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b- Nonsolvent Liquid Extraction Methods

1) Babcock Method

2) Gerber Method

3) Detergent Method

c- Instrumental methods

1) Measurement of bulk physical properties

2) Measurement of adsorption of radiation

3) Measurement of scattering of radiation

Folch's method is one of the major contributions to the field of lipid biochemistry by the Catalan biochemist and prominent scientist in the field of neurochemistry Jordi Folch-Pi (1911–1979). Prior to Folch's method there was no effective way to quantitatively isolate lipids from biological tissue samples. The standard method for lipid extraction from tissues until Folch was Bloor's method that applied a number of successive extractions using ethanol, ether, and chloroform and/or petrolether .This method was very time consuming and the resulting lipid extracts were not free from non-lipid contaminants.

Folch’s extraction procedure is one of the most popular methods for isolating lipids from biological samples. It takes advantage of the biphasic solvent system consisting of chloroform/methanol/water in a volumetric ratio of 8:4:3 (v/v/v).

Comparison of Methods

Soxhlet extraction is most commonly methods this is mainly because it is fairly simple and is the officially recognized method for a wide range of fat content determinations. The main disadvantages of the technique are that a relatively dry sample is needed (to allow the solvent to penetrate), it is destructive, and it is time consuming. For high moisture content foods it is often better to use batch solvent or nonsolvent extraction techniques.

Many instrumental methods are simple to operate, rapid, reproducible, require little sample preparation and are nondestructive. Nevertheless, they are often expensive to purchase and can only be used for certain types of foods, i.e., where there is no interference from other components. In addition, calibration curves prepared for instrumental methods usually require that the fat content be measured using a standard method.

Extraction techniques more accurate and more generally applicable and are therefore the standard methods for official analysis of many food materials (e.g., for labeling or legal requirements). Instrumental methods are most useful for rapid measurements of fat content in quality assurance laboratories of food factories where many samples must be measured rapidly.

The aim of all extraction procedures is to separate cellular or fluid lipids from the other constituents, proteins, polysaccharides, small molecules (amino acids, sugars...) but also to preserve these lipids for further analyses.

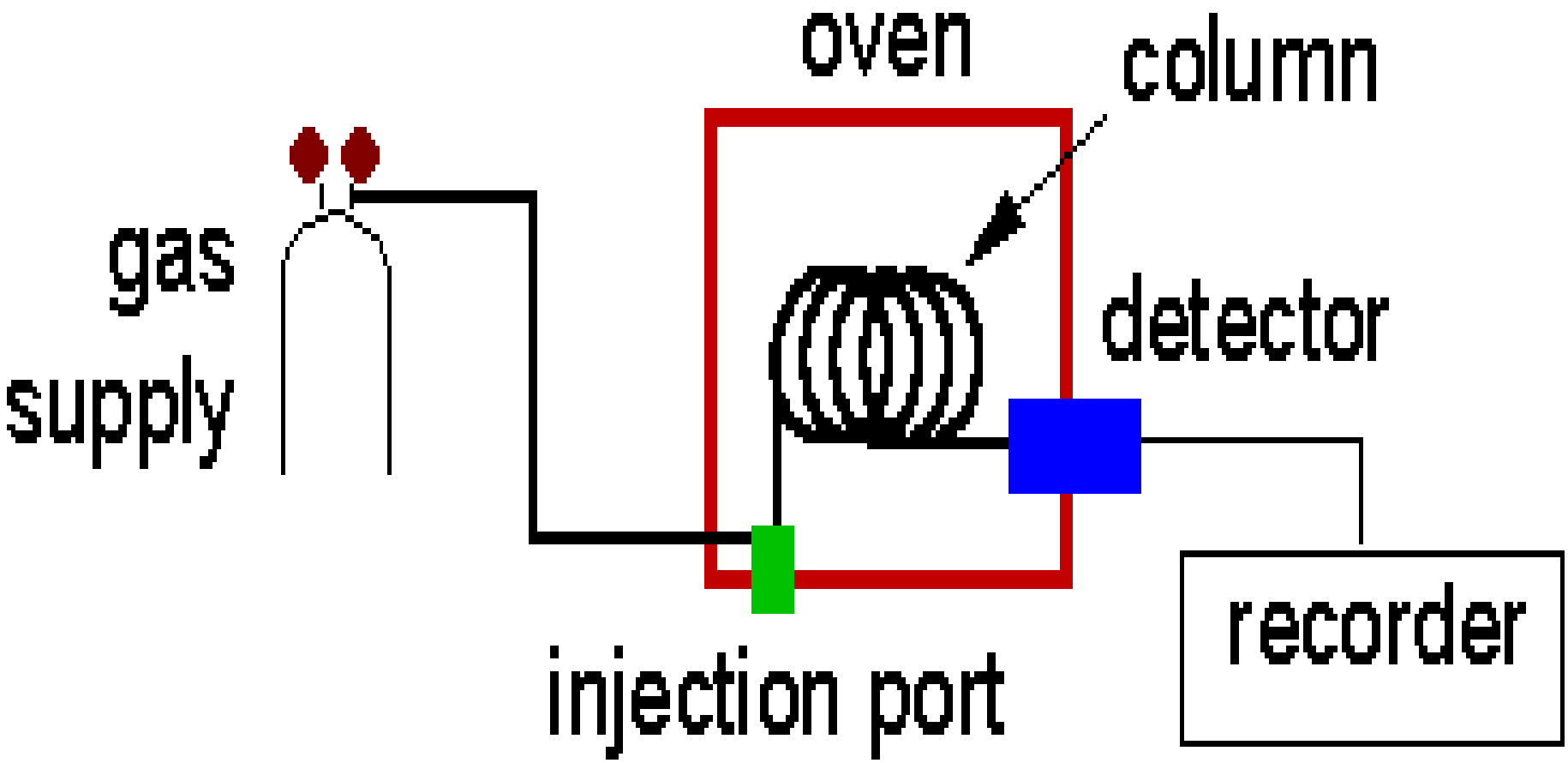
There is a great diversity of methodologies because biological tissues are not similar when considering their structure, texture, sensitivities and lipid contents. The ideal solvent for lipid extraction would completely extract all the lipid components from a sample, while leaving all the other components behind. In practice, the efficiency of solvent extraction depends on the polarity of the lipids present compared to that of the solvent.

Polar lipids (such as glycolipids or phospholipids) are more soluble in polar solvents (such as alcohols), than in non-polar solvents (such as hexane). On the other hand, non-polar lipids (such as triacylglycerols) are more soluble in non-polar solvents than in polar ones. The fact that different lipids have different polarities means that it is impossible to select a single organic solvent to extract them all. Thus the total lipid content determined by solvent extraction depends on the nature of the organic solvent used to carry out the extraction: the total lipid content determined using one solvent may be different from that determined using another solvent. Ethyl ether and petroleum ether are the most commonly used solvents, but pentane and hexane are also used for some foods.

Fatty Acid analysis by gas chromatography

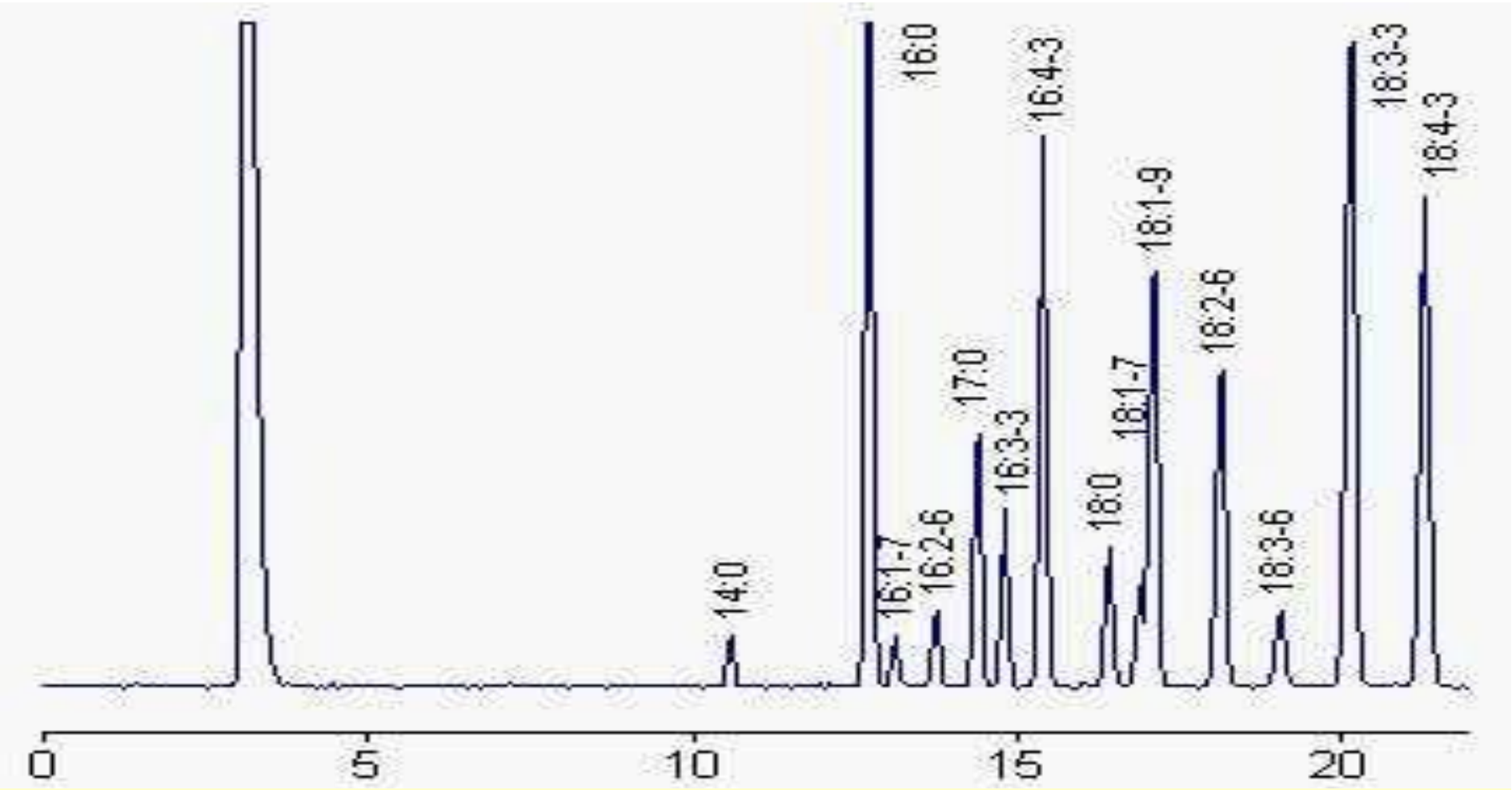
The discovery in the mid-1950's of gas-liquid chromatography (GLC or in short GC) has revolutionized the analysis of fatty acids and this technique is the most frequently used especially when combined with techniques which can be used to identify the chemical structure of the peaks, e.g., mass spectrometry or NMR. A chromatographic analysis involves passing a mixture of the molecules to be separated through a column which contains a matrix capable of retarding the flow of the molecules. Molecules in the mixture are separated because of their differing affinities for the matrix in the column. The stronger the affinity between a specific molecule and the matrix, the more its movement is retarded, and the slower it passes through the column.

Thus different molecules can be separated on the basis of the strength of their interaction with the matrix. After being separated by the column, the concentration of each of the molecules is determined as they pass by a suitable detector (e.g., UV-visible, fluorescence, or flame ionization). Chromatography can be used to determine the complete profile of molecules present in a lipid. This information can be used to: calculate the amounts of saturated, unsaturated, polyunsaturated fat and cholesterol; the degree of lipid oxidation; the extent of heat or radiation damage; detect adulteration; determine the presence of antioxidants. Various forms of chromatography are available to analyze the lipids in foods, e.g. gas chromatography (GC), high pressure liquid chromatography (HPLC), thin layer chromatography (TLC).



Scheme of a gas chromatograph

http://www.aquaculture.ugent.be/Education/coursematerial/online%20courses/ATA/content.htm



Chromatogram of the fatty acid profile from a green algae Notice the presence of 17:0 as an internal standard, and the three fatty acids: 16:2n-6, 16:3n-3 and 16:4n-3