



Medical Laboratory Techniques Department

Practical Biochemistry

SECOND STAGE \ SECOND COURSE

Lab 1-2

Lipids tests

Assist Lecturer

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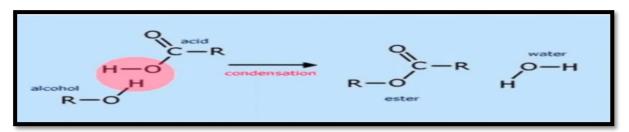
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Lipids

- ➤ Lipids are a class of biological molecules that are insoluble in water and soluble in nonpolar solvents.
- > Lipids are esters of long chain fatty acids and alcohols.
- > Fatty acids are lipids' building blocks
- ➤ It can be defined as nonpolar organic compound insoluble in polar solvent, but soluble in organic solvents such as benzene, ether, chloroform and boiling alcohol.

> Lipids are

- 1) Simple lipids are esters of fatty acids with various alcohols:
 - a. Fats & oil (esters of fatty acids with glycerol) and
 - b. Waxes (esters of fatty acids with higher molecular weight of monohydric alcohols).
 - 2) Complex lipids are esters of fatty acids containing groups in addition to an alcohol and a fatty acid, e.g., phospholipids or glycolipids etc.
 - **3) Derived lipids** include fatty acids, glycerol, steroids, other alcohols, fatty aldehydes, and ketone bodies, lipid soluble vitamins, and hormones.



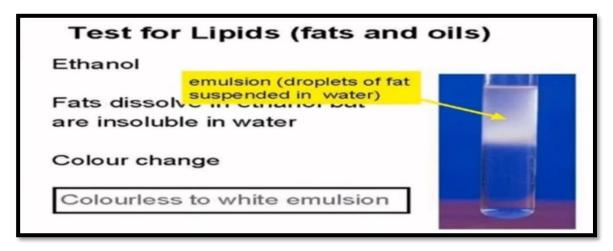
Note: Structures linked by ester bonds (R-COOR') and water is released

1- Ethanol emulsion Test

(A general group test for detection of lipids)

Principle:

- -The presence of lipids is observed by the appearance of cloudy white layer on top of the reaction mixture.
- -This test is based on the fact that lipids are dissolve in ethanol (due to hydrophobic interactions) but on addition of water, lipids spontaneously disperse to form micelles (small droplets), which form the top layer (being less dense than water and ethanol) and appear cloudy white.
- -The lipids come out of the solution because the overall strength of hydrogen bonding interaction between ethanol and water is much greater than hydrophobic interaction between lipid and ethanol.



Procedure:

	Tube A	Tube B	
Oil	5 drops		
DW		5 drops	
Ethanol	2 ml	2 ml	
	Shake wel	1	
DW	2 ml	2ml	
	Shake wel	1	
	Record your obse	ervation	

Note: Samples with high lipid content will form a thicker cloudy suspension.

Result sheet

Experiment	Observation	Inference
Tube A		
Tube B		

Homework

- 1. What the purpose of the test?
- 2. When cloudy white layer will appearance in the test?
- 3. Why the lipids come out of the sample?

2- Acrolein Test

Objective:

To detect presence of fats or glycerin.

Principle:-

When glycerol is heated with potassium bisulphate or concentrated H2SO4, dehydration occurs and aldehyde Acrolein formed which has characteristic odour.

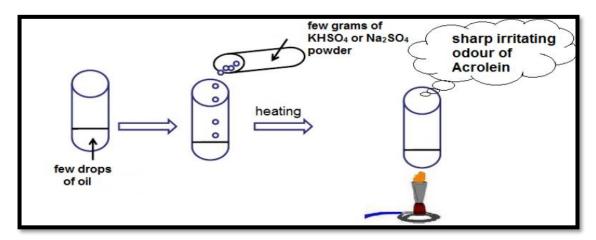
$$\begin{array}{c|cccc} CH_2-OH & Heat & CH_2 \\ \hline CH-OH & KHSO_4 \text{ or } \\ \hline CH_2-OH & COnc.H_2SO_4 & CHO \\ \hline \\ Glycerol & Acrolein \\ \end{array}$$

Material:-

- 4. Olive oil or fat.
- 5. Potassium bisulphate or sodium sulfate powder

Procedure:-

- 1. Place 5 drops of test compound in a clean and dry test tube
- 2. Add 1 ml of conc. H2SO4 carefully. Or 1.0 g of KHSO4
- 3. Heat the test tube directly.
- 4. Note the characteristic pungent odour of Acrolein.



3-Sudan IV Test

Objective:

To detect presence of lipids.

Principle:

Sudan IV is a non-polar stain that readily stains lipids red-orange

but does not stain **polar compounds** or aqueous solutions

