Triglycerides

Properties of Triglycerides

Triglycerides

or

Triacylglycerols (TAGs)

are

Complexes of non-polar lipids.

- Lipid is a general term that describes substances that are relatively hydrophobic: water-insoluble and extractable by non-polar solvents.
- **TAGs are esters of the alcohol glycerol reacting with fatty acids.**

Properties of Triglycerides…..cont.

Triglycerides

- TAG are the major form of

Neutral Fat

found in nature.

Properties of Triglycerides ….cont.

- **TAG** is the

Principal Fat

found in *foods*, *body tissues* and *blood*.

- Mammalian tissues also contain some ***diglycerides*** and ***monoglycerides***, these occur in trace levels when compared with ***Triglycerides***.

Properties of Triglycerides …..cont.

- Fat in food becomes fat (triglycerides) in our blood.

  - *It is the main constituent of vegetable oils and animal fats.*

  - ***A diet high in carbohydrates (sugar and starches) may also raise blood triglyceride levels.***

Properties of Triglycerides .....cont.

- TAG is the main fuel store in the body.

Structure of Triglycerides

Triglycerides (Triacylglycerol,TAG) are

Esters of

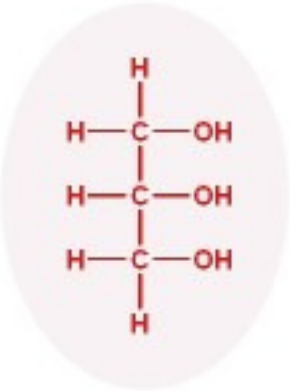
=

Glycerol

Reacting with

Three Fatty Acids

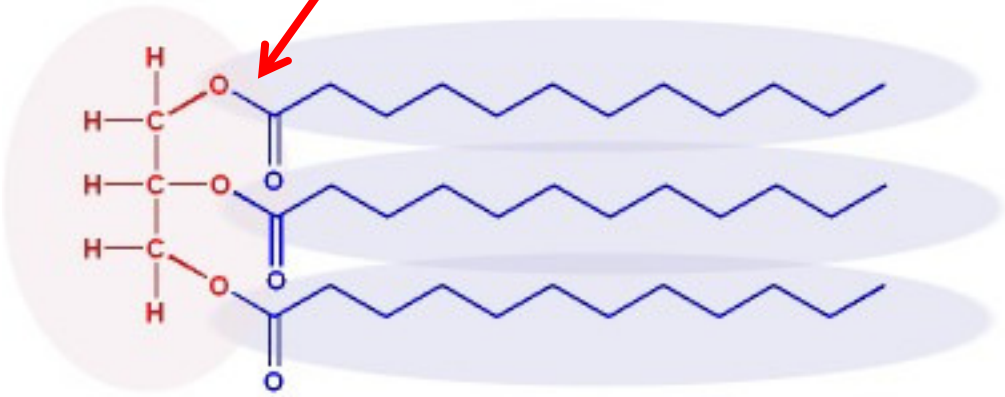
Glycerol



A "free" Fatty Acid

Ester Linkage

Triglyceride



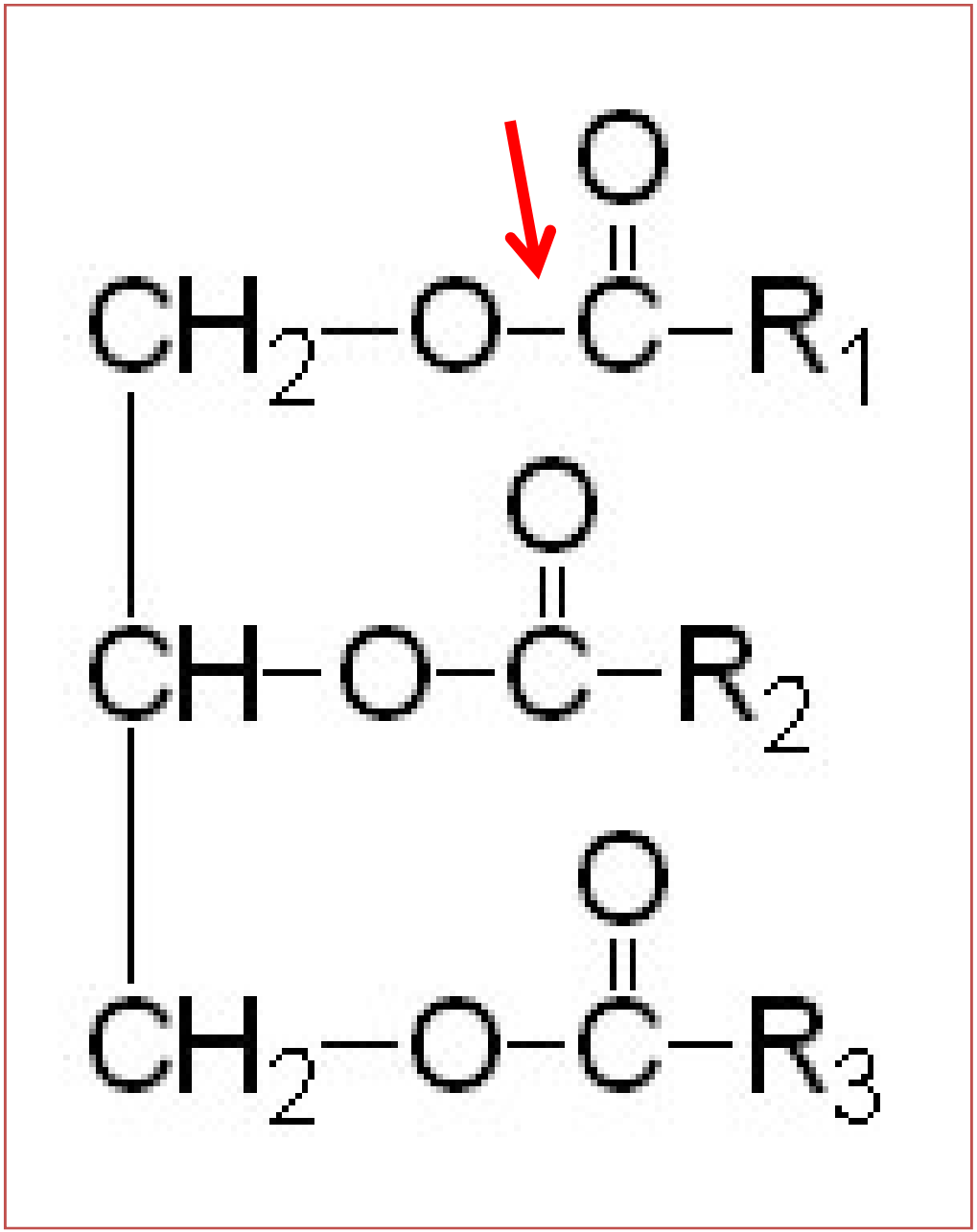
- **Glycerol, Free Fatty Acids and TAG Structures**

Structure of Triglyceride ……cont.

❑ Fatty Acids of TAG

•The three fatty acids can be all different, all the same, or only two the same. They can be saturated or unsaturated fatty acids.

• Chain lengths of the fatty acids in naturally occurring triglycerides can be of varying lengths but 16, 18 and 20 carbons are the most common.

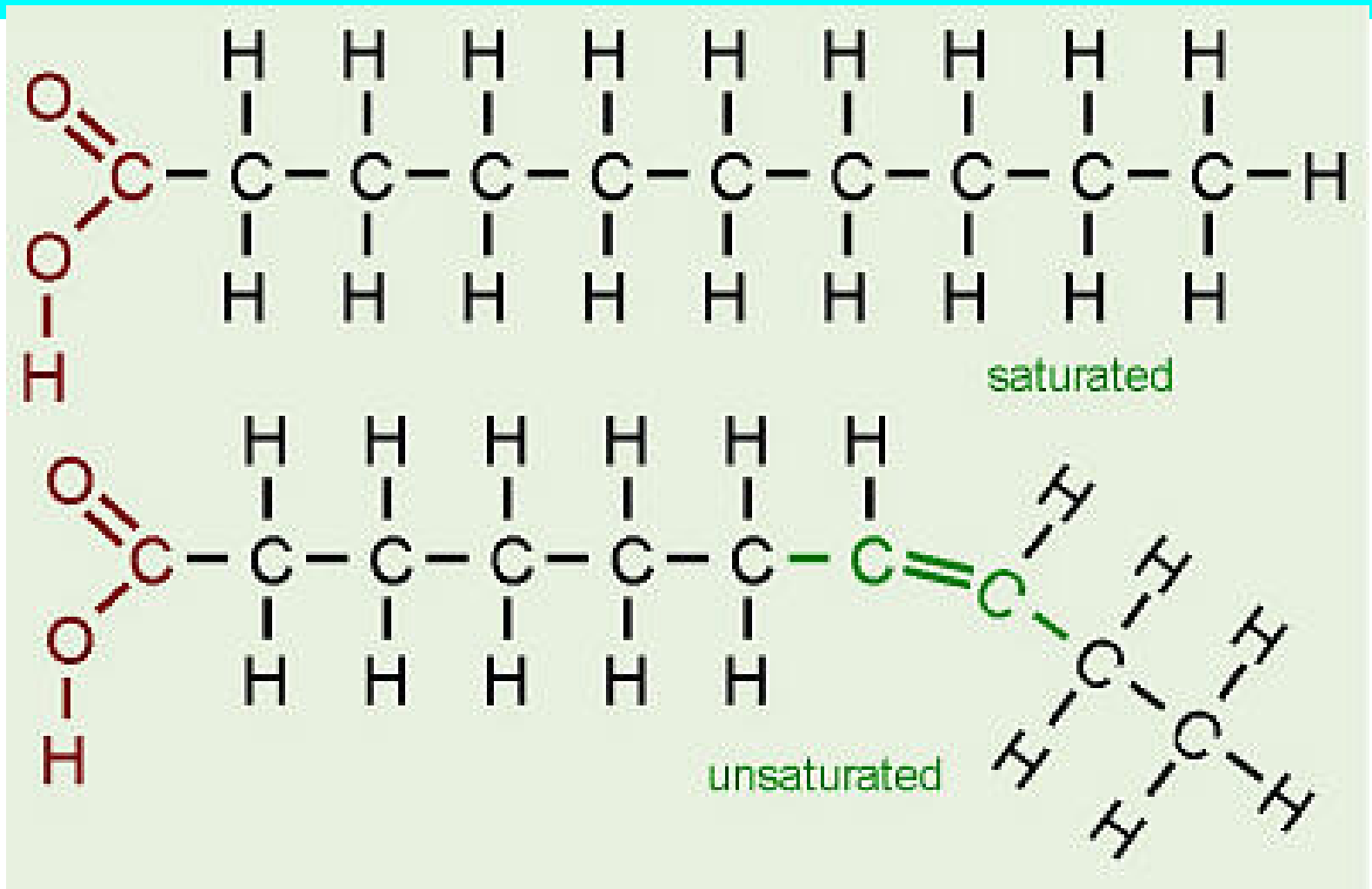


Structure of TAG

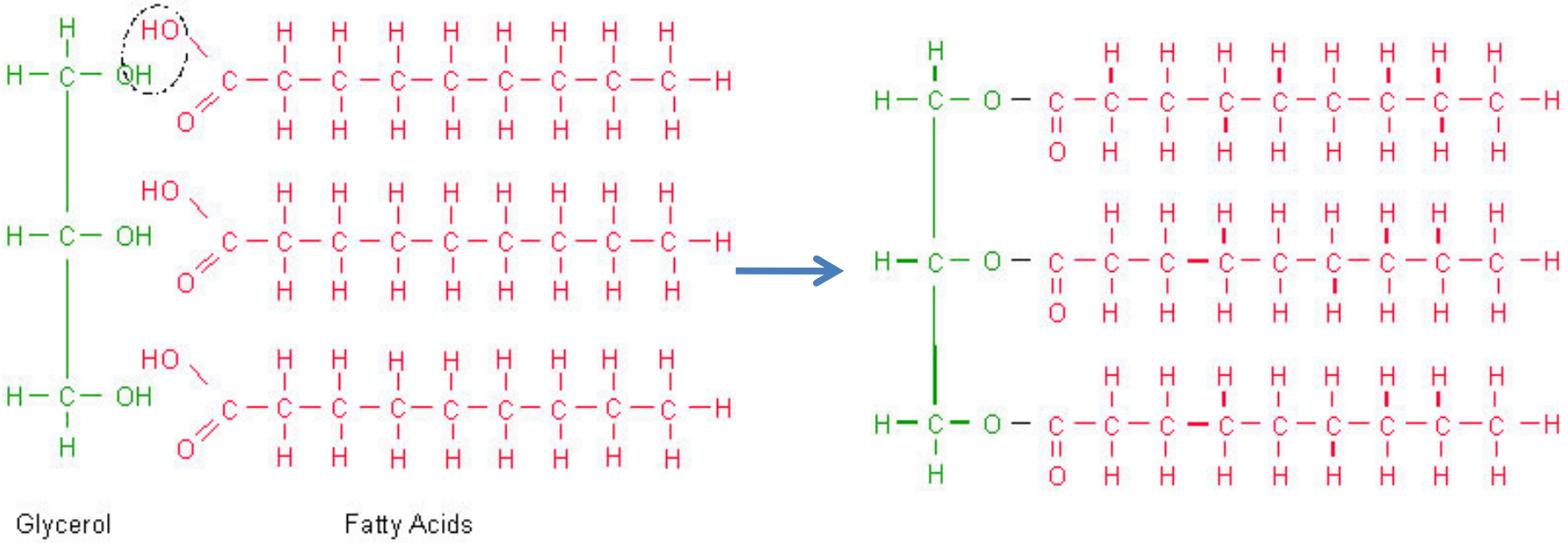
↓ = Ester Bond

Structure of Triglyceride …..cont.

Saturated and Non-Saturated FFAs



Formation of TAG from Glycerol and FFAs



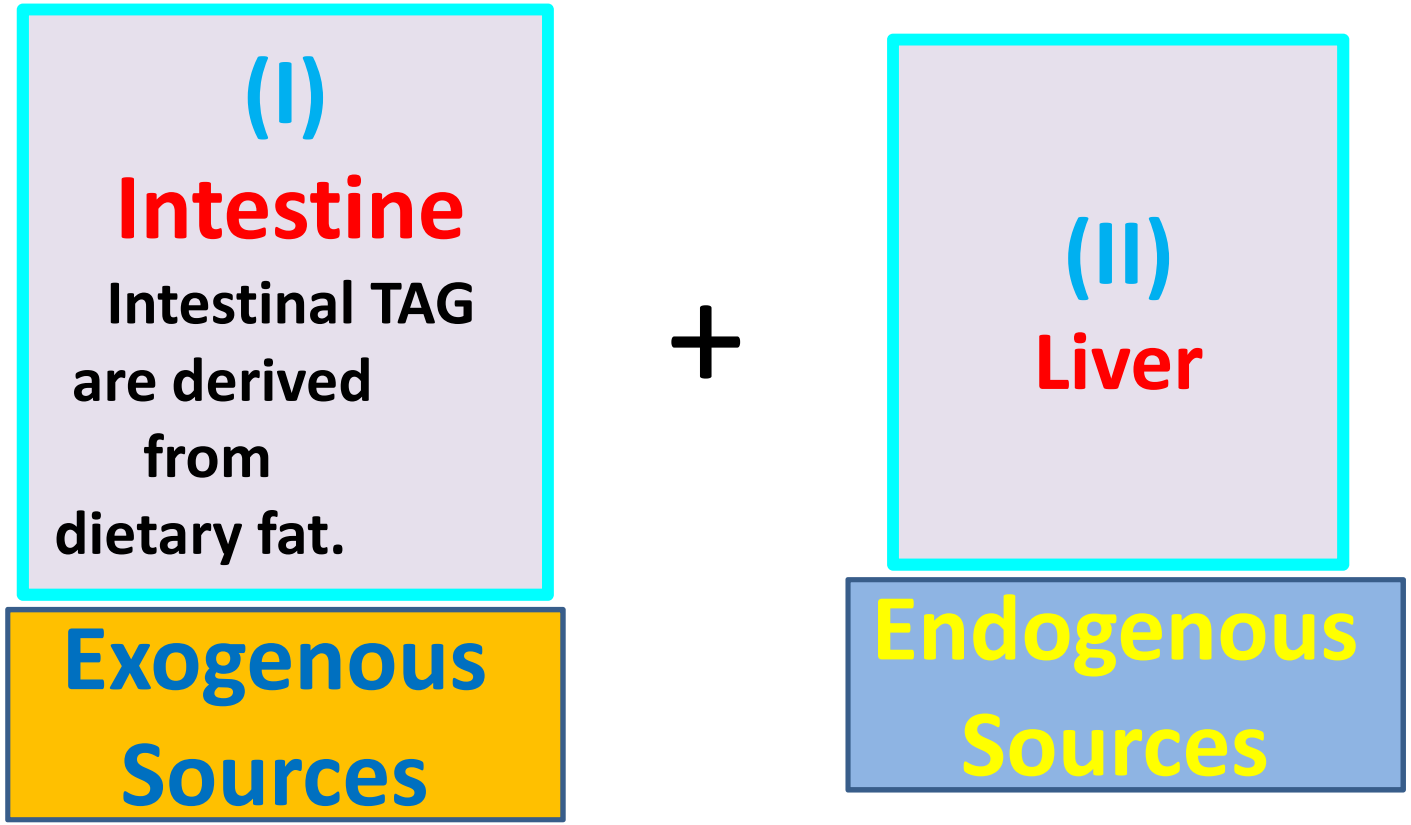
Glycerol Fatty Acids

+

$3H_2O$

Sources of Plasma Triglycerides

Plasma TAG are derived from 2 sources:



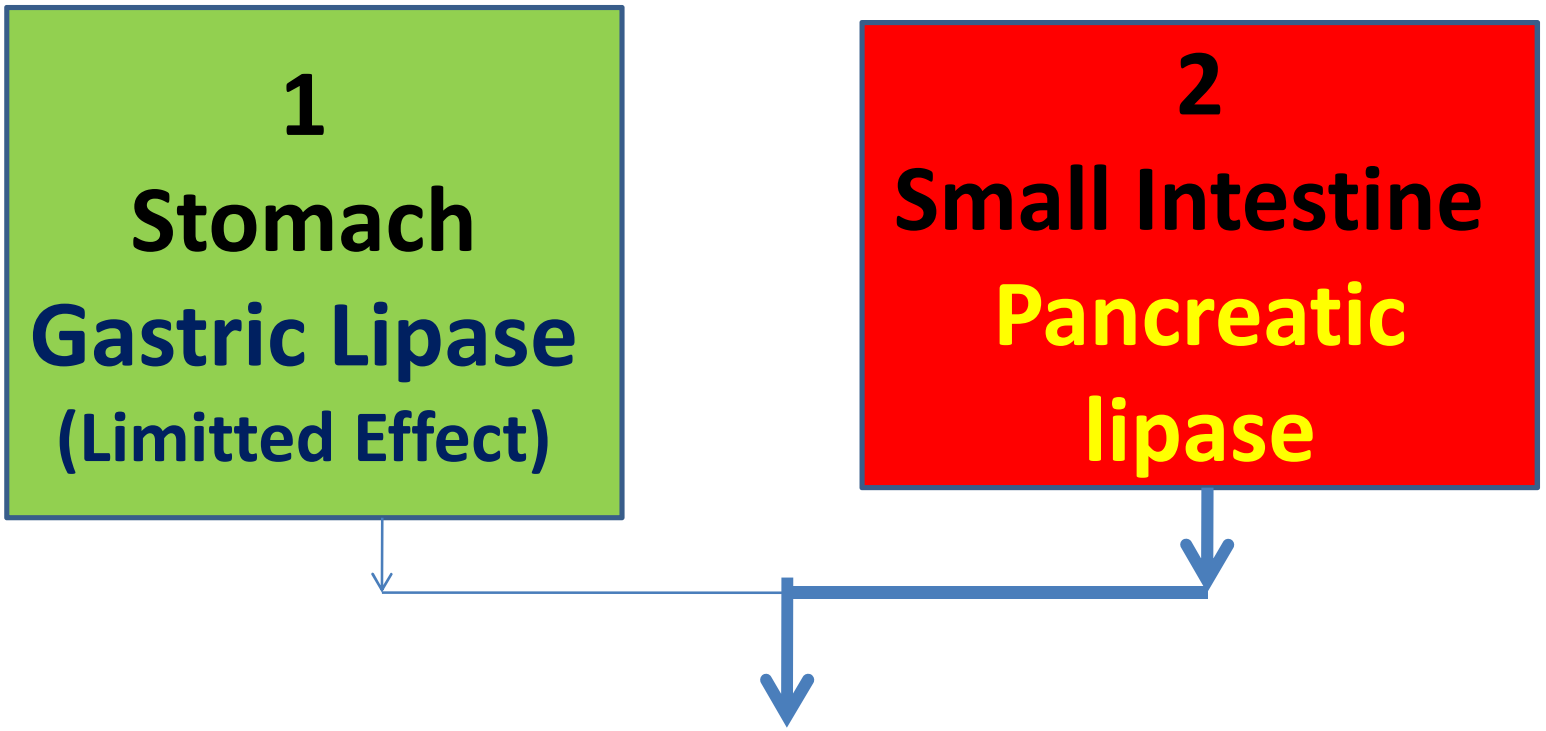
Sources of Triglycerides

1) Exogenous Sources

- An adult human ingest about 60- 150 g of lipid per day. Triglycerides consistitute more than 90% of this intake.
- Excess dietary fat ***can only be stored as TAG*** in adipose tissue.

Digestion of Exogenous TAG by Stomach and Small Intestine

TAG in the diet are digested in:



2-Monoacylglycerols + 2 Free fatty acids.

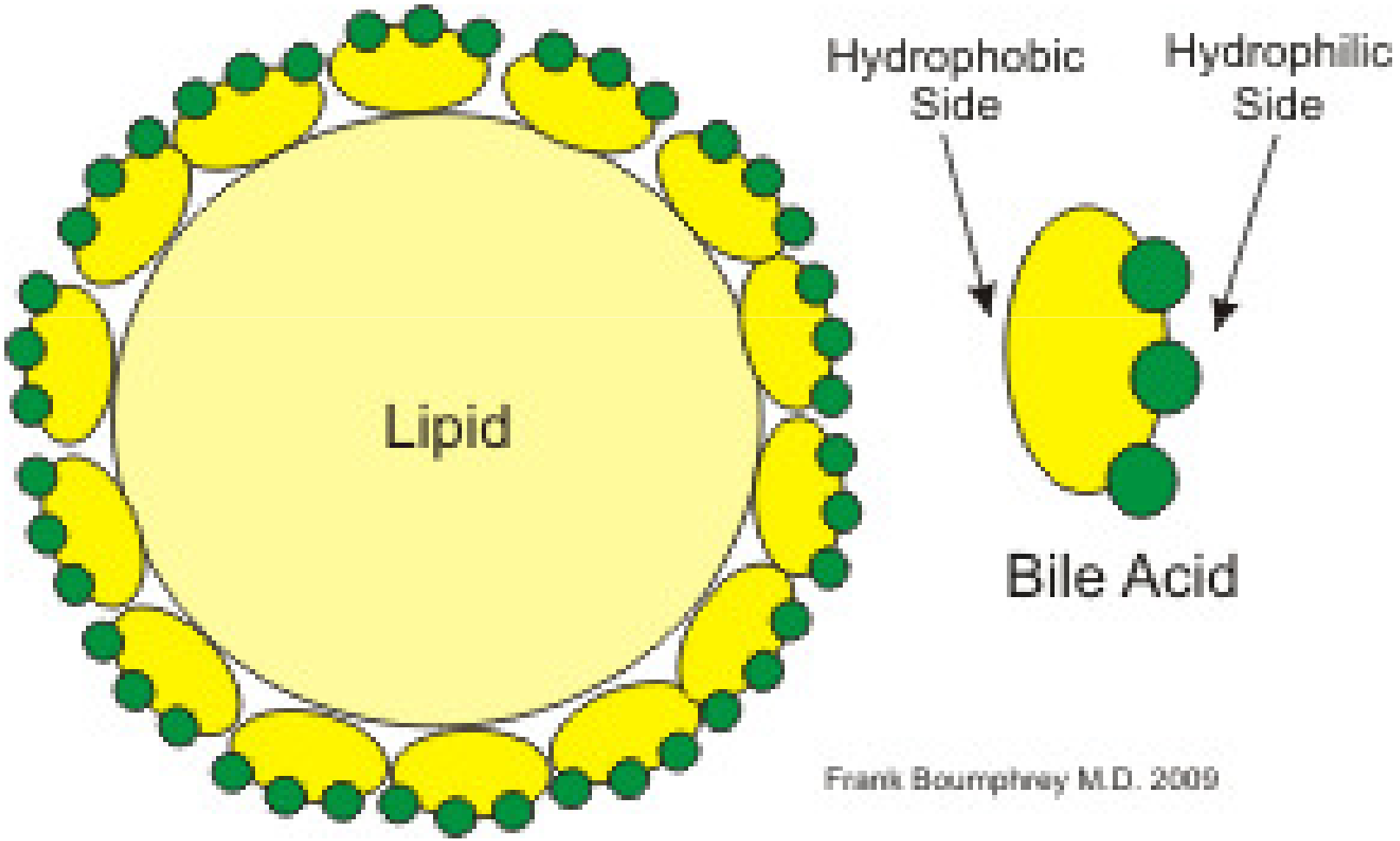
Digestion of Exogenous TAG in Stomach and Small Intestine ….cont.

1. Gastric lipase. The effect of gastric lipase on TAG to break it down is a limited effect because fat is not yet emulsified in the stomach. TAG reaches the duodenum largely unaltered.

In the intestine, TAG is solubilized by bile acids, which are secreted from liver / gall bladder. High CMC (critical micellar concentration) of bile acids ensures rapid action.

2. Pancreatic Lipase. After emulsification by bile acids, solubilized TAG is degraded by pancreatic lipase (that is secreted into duodenum).

How Bile Acids Emulsify Lipids ?



Intestinal Lipolysis and Absorption of Triglycerides

- After TAG emulsification (micelles formation) by the bile :

Pancreatic lipases breakdown TAG

↓

Lipolysis

TAG molecules are split into

Glycerol + Free fatty acids

↓

Which are then moved into the cells lining the intestine (absorptive enterocytes).

How Non-polar TAG is Transported from Intestine to the Liver and other Tissues by the Blood ?

The TAG are rebuilt in the enterocytes from their fragments:

FFA + glycerol

How Non-polar TAG is Transported from Intestine to the Liver and other Tissues by the Blood? .....cont.

- **Since TAG are insoluble in water, the problem of how to transport TAG in the aqueous blood is solved by the ability of enterocytes to associate:**

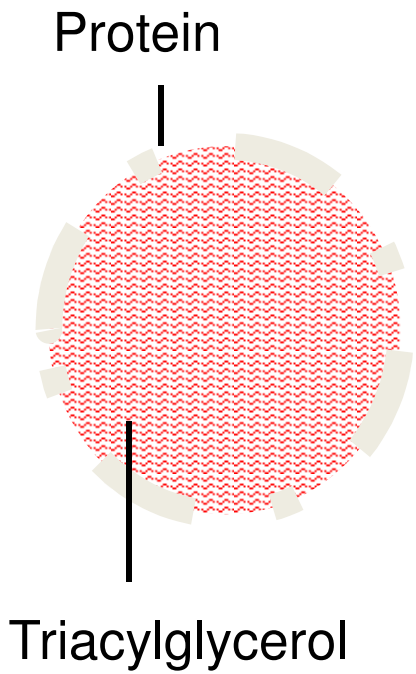
  1) **Nonpolar lipids** (**TAG** and cholestryl esters)

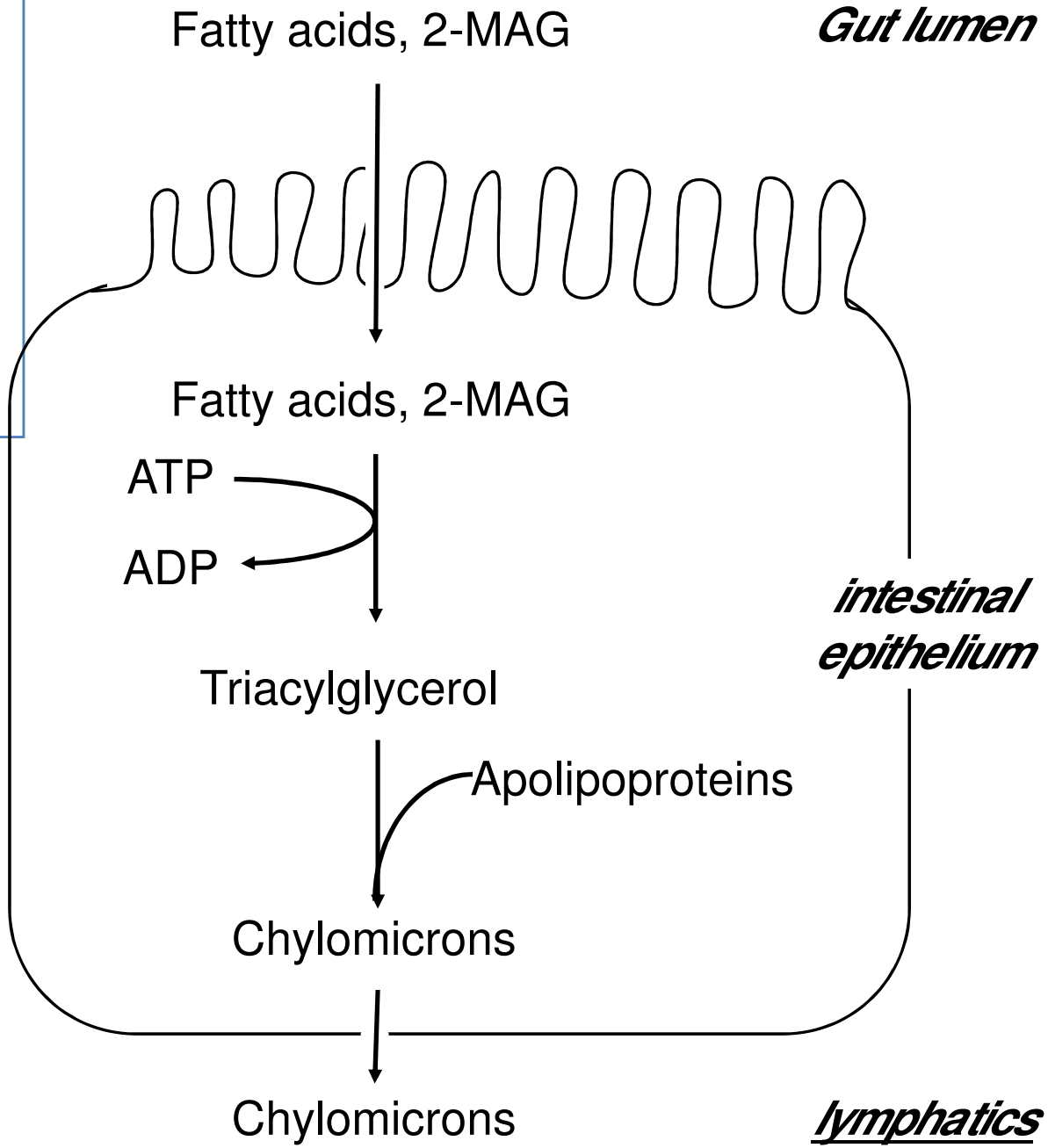
  2) **Amphipathic lipids** (phospholipids and cholesterol)

  3) **Proteins**

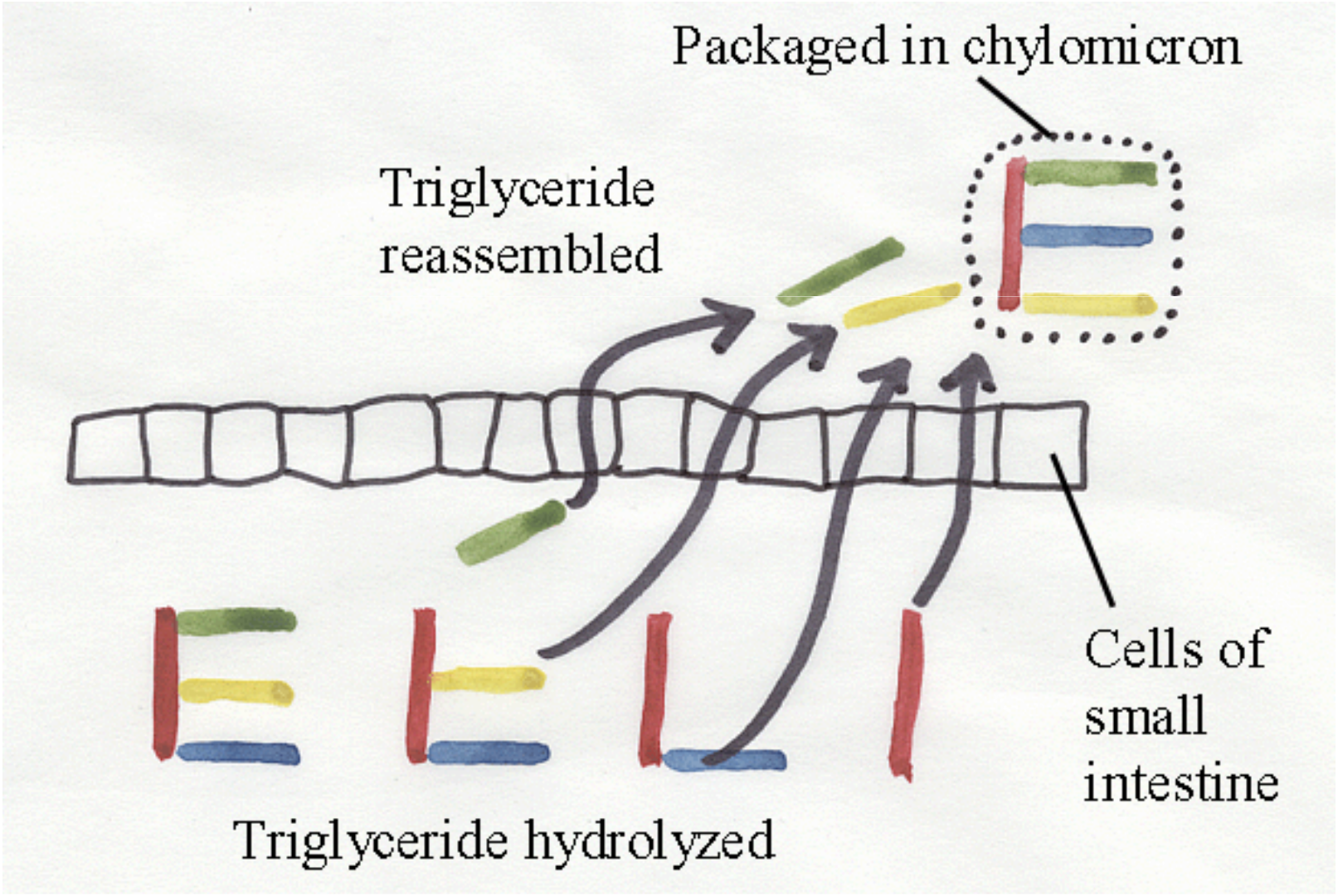
  in large particals; **chylomicrons**.

After digestion and uptake by intestinal cells, triacylglycerol is resynthesized and packaged into **chylomicrons**





The Role of Chylomicrons in Transporting TAG



Transport of TAG from Intestine to Liver and other Tissues

- **Chylomicrons**; water-miscible lipoproteins ,are excreted from the intestinal cells and collected by the lymph system.
- Chylomicrons are responsible for the transport of all dietary lipids from intestine to the circulation, however, **TAG is the predominant lipid in chylomicrons**.

Transport of Triglycerides from Intestine to Liver and other tissues

- Various tissues can capture chylomicrons, which will release TAG into the cells to be used as a source of energy.
- **Adipose tissues** are (after the **liver**) the main tissue that clear chylomicrons from the circulation and taking TAG, however, this is mainly for storage and not for energy production.

Sources of Triglyecrides

2) Endogenous Sources

❑ Biosynthesis of TAG

- Most mammalian tissues convert fatty acids to triacylglycerols by a common sequence of reactions :

(1) Liver **(2) Adipose tissue**

These 2 tissues carry out this process to the greatest extent than others.

Endogenous Sources of TAG .....cont.

- **(I) Biosynthesis of TAG by the Liver (from Exogenous FFAs)**
- Chylomicron remenants remaining after digestion by **lipoprotein lipase** are cleared from the blood by the liver. TAG present in the remenants are hydrolzed by lysosomal lipase.

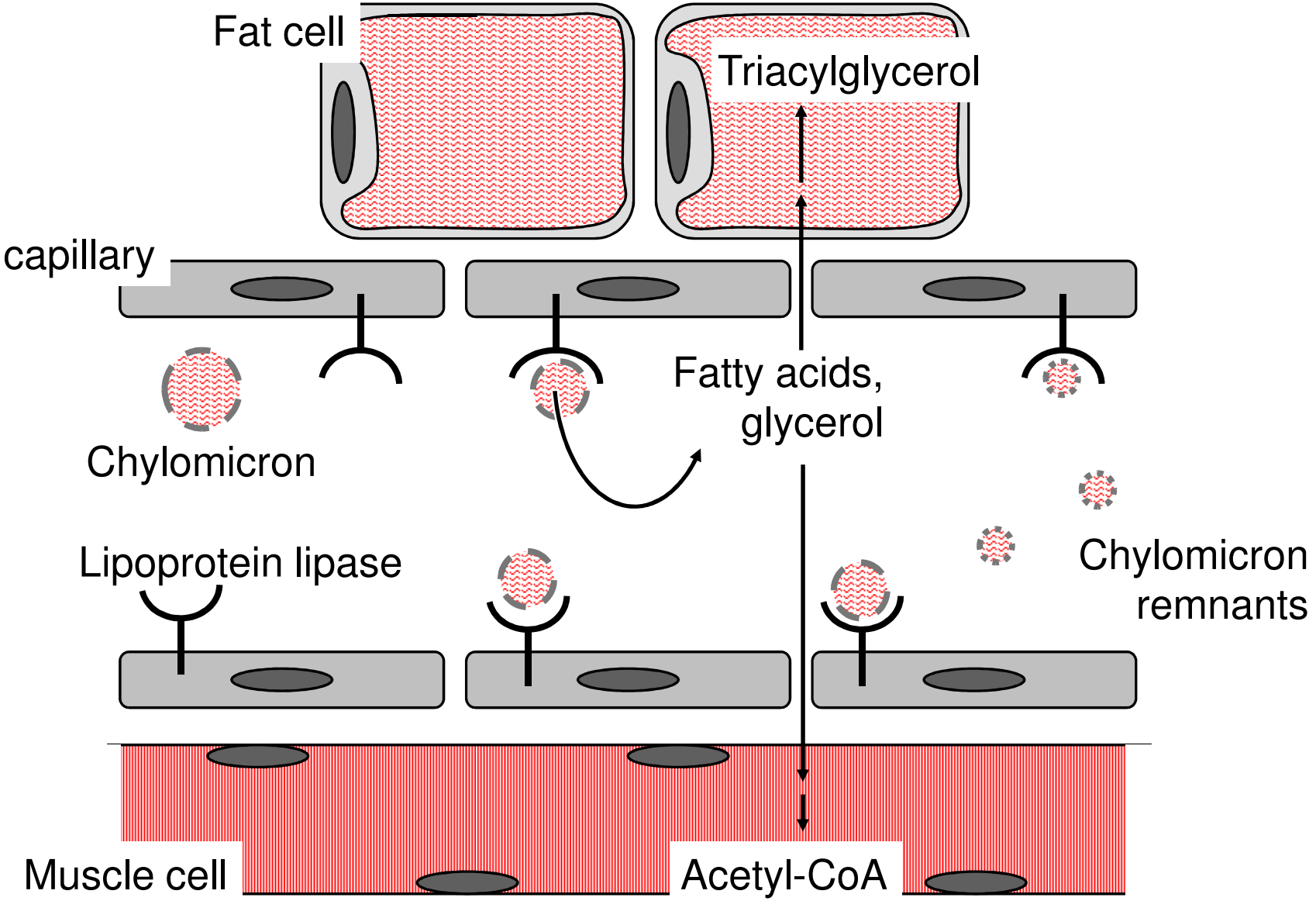
The role of Lipoprotein Lipase in Utilizing TAG in Chylomicrons and VLDL by Different Tissues

- **Lipoprotein lipase is expressed in**

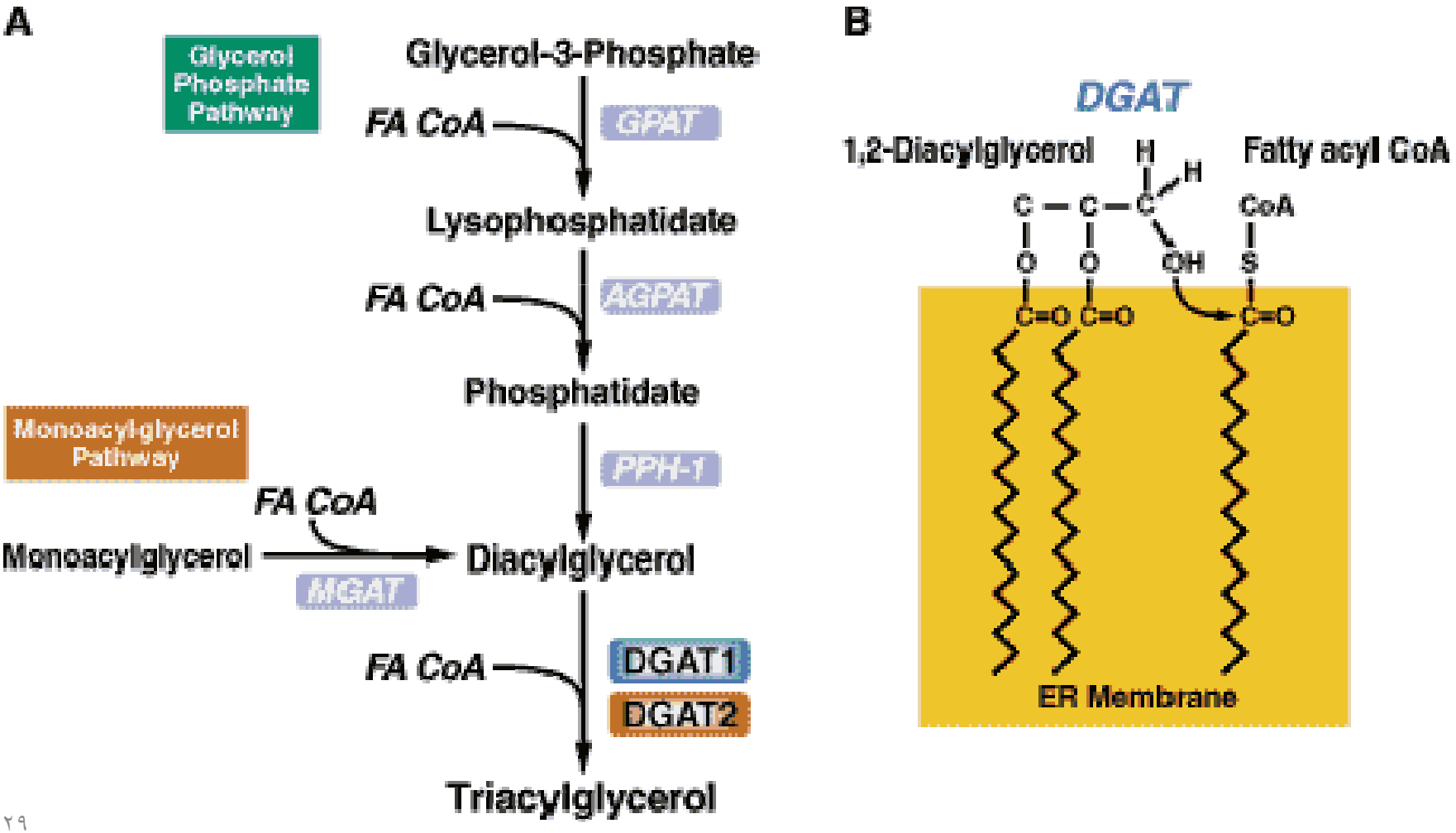
1. Adipose tissues
2. Cardiac muscle
3. Skeletal muscle

Allowing these tissues to utilize TAG from lipoproteins.

Utilization of Chylomicrons in Peripheral Tissues



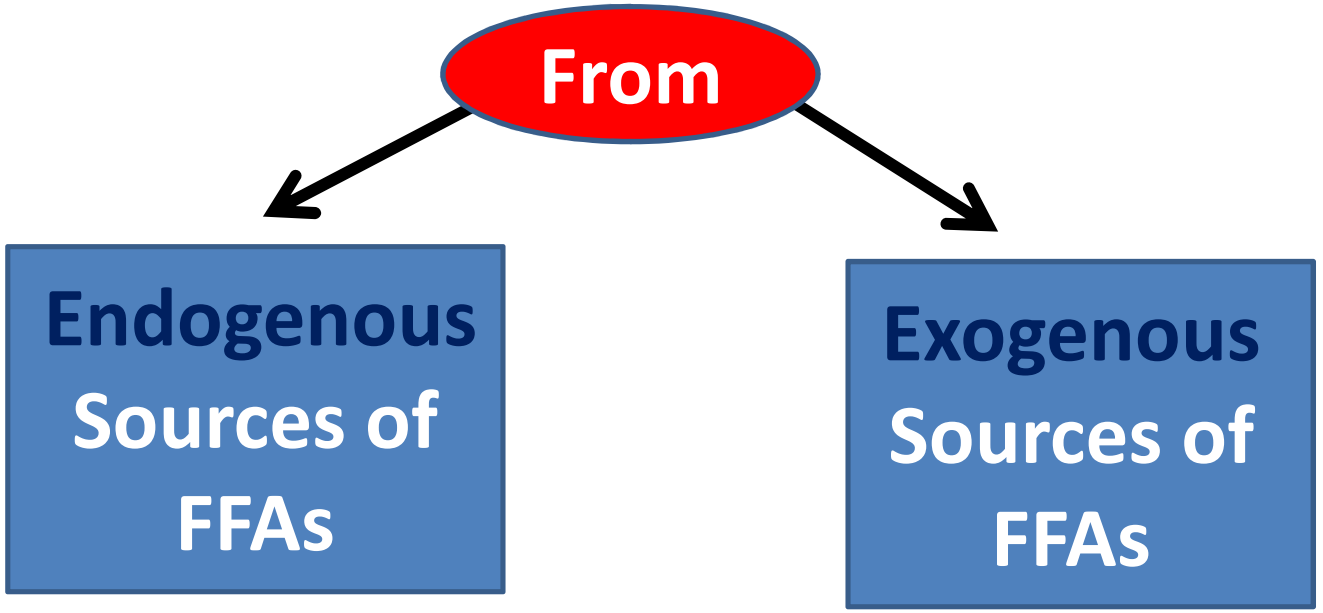
TAG Synthesis



Sources of Triglycerides ……cont.

2) Endogenous Sources

I) Biosynthesis of TAG by the Liver



Transport of TAG from Liver to Extrahepatic Tissues

1. **FFA**

comes from the chylomicrons
re-esterified with

2. **Glycerol -3- phosphate**

(derived from free glycerol and glucose)

↓

TAG

which is then packaged into

VLDL

which is secreted into the bloodstream to go to extrahepatic tissues.

Transport of Triglycerides from Liver to Extrahepatic Tissues....cont.

VLDL

are vehicles of transport of

TAG

from
the liver to the extrahepatic tissues.

Transport of lipids between organs

1. **Lipoproteins. These basically are lipid droplets with a hydrophilic protein coat. Important examples are:**

- Chylomicrons. Distribute triacylglycerol (TAG) from intestine to peripheral organs (***bypassing*** the liver)

- VLDL = very low density lipoprotein. Moves TAG and some other lipids from liver to periphery

- LDL = low density lipoprotein. Moves lipids (particularly ***cholesterol***) from liver to periphery

HDL = high density lipoprotein. Moves excess cholesterol from peripheral organs to liver

٢. Free fatty acids. These are bound to albumin. This is the major transport mechanism for release of fat from fat tissue

Where are Triglycerides Stored in the Body?

- TAG is confined largely to storge sites in **adipose tissue**.
- TAG synthesis in **liver** is primarly for the production of plasma lipoproteins, rather than for energy storage.
- Some storage of TAG also occurs in **skeletal** and **cardiac muscle**, but only for local consumption.

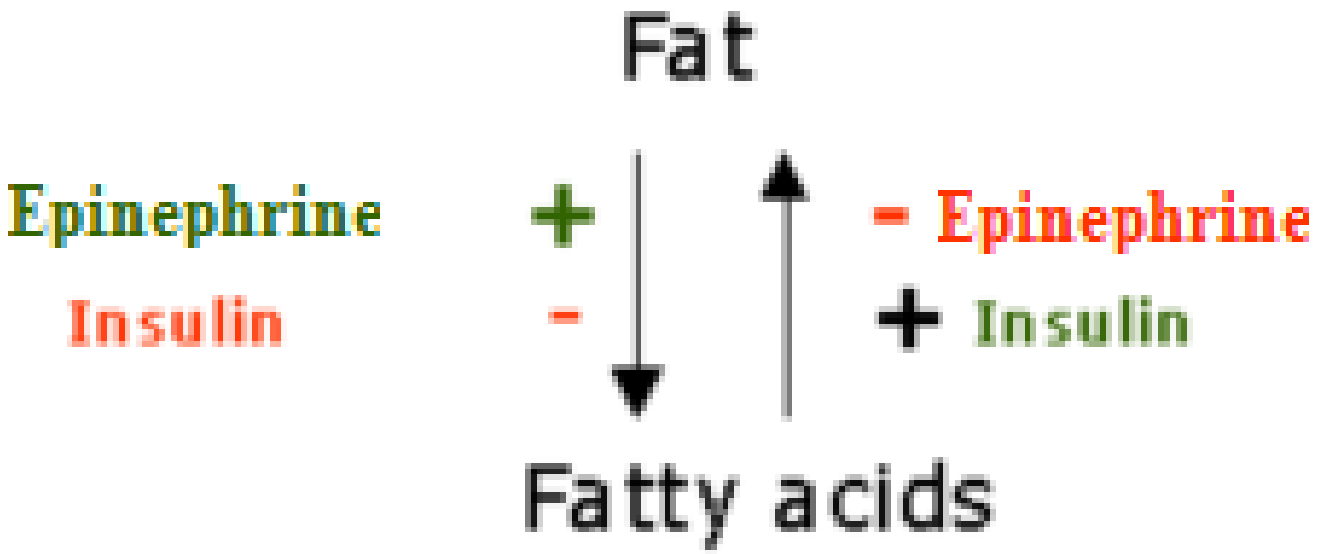
Where are Triglycerides Stored in the Body?

	Synthesis	Storage Site for Local Consumption to Produce Energy	Storage Site to Export to Different Tissues	Storage Site for Lipoproteins Production
Adipose Tissues	TAG (mainly in this tissue)	TAG	TAG	
Liver	TAG (mainly in this tissue)	TAG		TAG (mainly in this tissue)
Skeletal Muscle and Cardiac Muscle	TAG	TAG (mainly in this tissue)		

TAG Transport in the Fasted State

- When the body needs FFAs, TAG stored in *adipose tissue* are mobilized for use as fuel in the fasted state. This process is initiated by the *hormone-sensitive lipase*, which is located within adipocytes.
  - Insulin inhibits the activity of this enzyme.
- During fasting, glucagon, epinephrene and norepinephrene signals the breakdown of TAG by increasing the activity of this lipase to release free fatty acids.

Effect of Hormones on Hormone-Sensitive Lipase Activity



Function of Triglycerides

1) The primary function of TAG is to provide energy to the cell. TAG are directly utilized by many tissues as an energy source. They contain more than twice as much energy (9kcal/g) as carbohydrates and proteins.

2) TAG, as major components of very low density lipoproteins (VLDL) and chylomicrons, play an important role in the body metabolism as energy sources.

3) FFAs produced from TAG hydrolysis, can be converted by many tissues to phospholipids which are important consistituents of membranes.

Levels of TAG in Plasma

- **The Concentration of TAG**

in the plasma at any given time is a balance between:

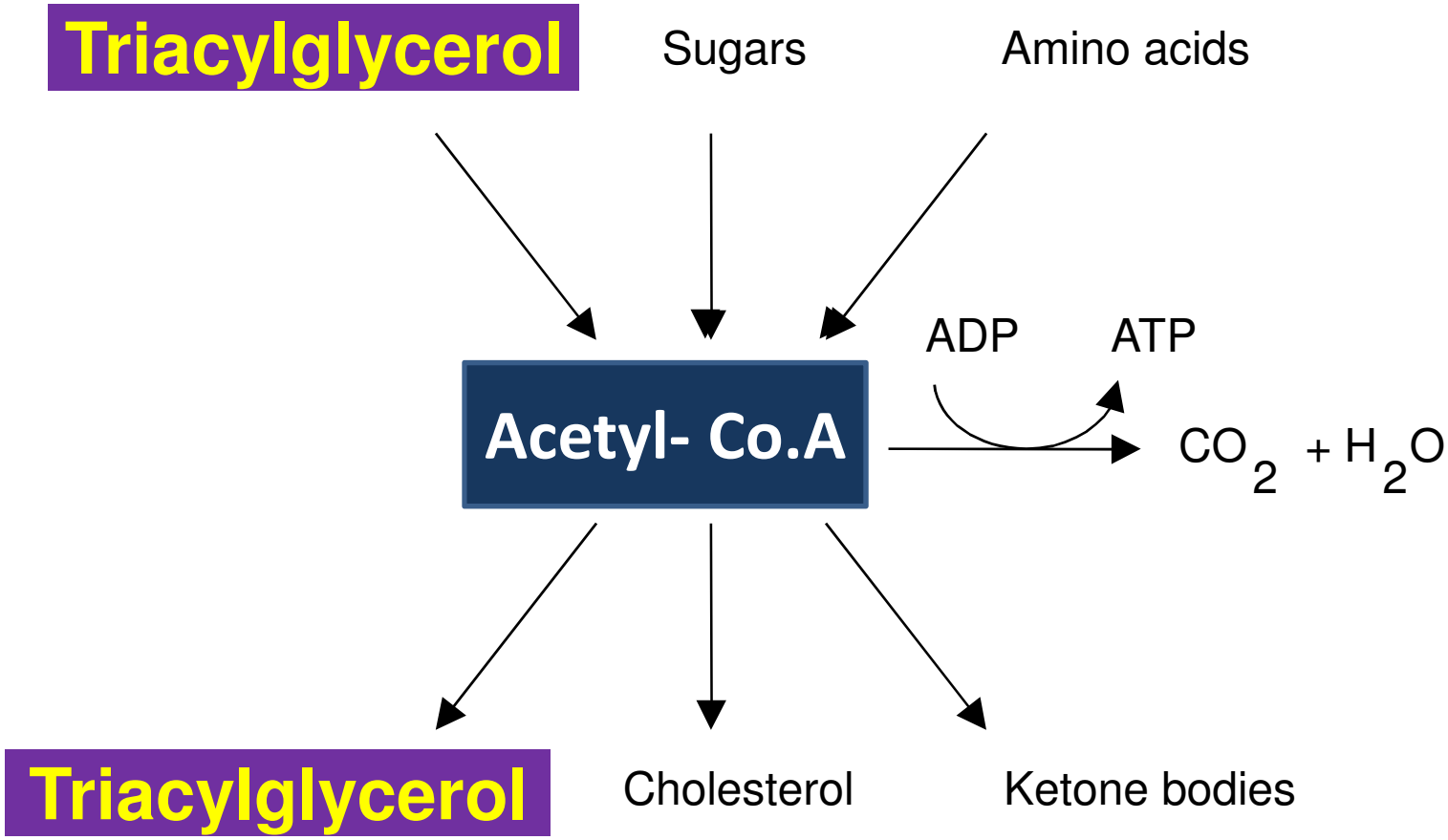
(I) **The Rate of Entry into the Plasma**

and

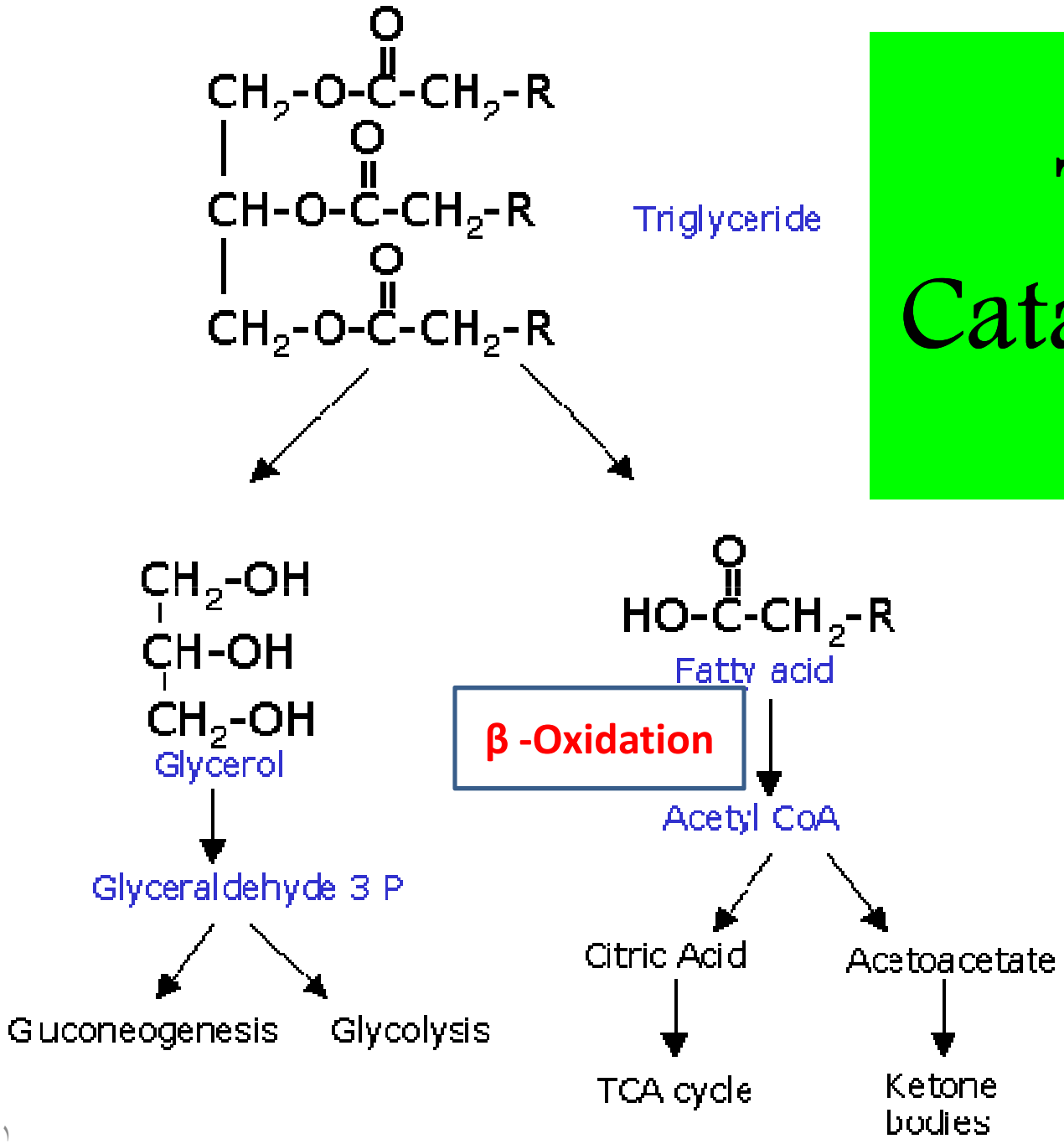
(II) **The Rate of Removal**.

- A change in the concentration may therefore be a result of a change in either or both of these factors.

Metabolism of Triacylglycerol Overview



TAG Catabolism



Why Hypertriglyceridemia is Dangerous?

- Elevated Levels of Triglycerides in Plasma

have been identified as
risk factor related to

Atherosclerotic Disease.

Hypertriglyceridemia

FACTORS THAT CONTRIBUTE TO ELEVATED SERUM TRIGLYCERIDES

- Excess weight or obesity
- Physical inactivity
- Stress
- Excessively high carbohydrate diets (<60% of the caloric intake)
- Type II diabetes
- Chronic renal failure
- Drugs (such as corticosteroids, estrogens, retinoids, high does of beta adrenergic blocking agents)
- Certain genetic metabolic disorders (including familial combined hyperlipidemia, and familial hypertriglyceridemia.

HIGH TRIGLYCERIDES LEVELS

can be controlled in most cases, but not cured.

To do so you must make permanent, beneficial changes in your lifestyle.

How can Blood Triglycerides be Lowered?

High triglyceride levels can be controlled in most cases, but not cured. To do so you must make permanent, beneficial changes in your life style.

How can Blood Triglycerides be Lowered?

1. Achieve and maintain your ideal body weight.

To do that, examine your eating habits. Are you overeating, eating only one large, late meal a day, having a bedtime snack?

- There are many reasons for overeating, not just hunger (stress, boredom).

High triglyceride levels can be controlled in most cases, but not cured. To do so you must make permanent, beneficial changes in your life style…….cont.

How Can Blood Triglycerides be Lowered?

2. Increase your Activity

If you are overweight, you have taken in more calories than you have used up. "Burn up" calories by exercising - moderate brisk walking (1/2 hour 3 - 4 times per week, or as directed by your physician).

High triglyceride levels can be controlled in most cases, but not cured. To do so you must make permanent, beneficial changes in your life style…..cont.

How can Blood Triglycerides be Lowered?

3. Decrease your Calorie Intake

- Take smaller portion sizes at each meal. Use low calorie foods and snacks. Have three meals a day rather than one large, late meal.
- Choose whole grain, higher in fiber breads, cereals, crackers, whole grain pastas, and rice.
- Try homemade, high fiber, low sugar baked goods.
- Fruits contain natural sugars. Limit fruit juice and use only 100 % fruit juice, no sugar added brands.
- Choose whole fruits more often.
- Use artificial sweeteners.

Hypotriglyceridemia

Hypotriglyceridemia

is a state of

Low Triglyceride Levels

❑ **Differential diagnosis of underlying causes**

- Hyperthyroidism
- Malabsorption syndrome
- Hereditary abetalipoproteinemia
- Hypobetalipoproteinemia

(I) OBJECTIVE OF THE EXP.

- **To determine the level of TAG in a serum or plasma sample.**

(II) METHOD PRINCIPLE

The standard method used for the measurements of triglycerides concentration ,which is used in this experiment , is an enzymatic one. This formulation makes use of the enzymatic hydrolysis of TAG and quantification since it is specific and not subject to interference by phospholipids.

(I) METHOD PRINCIPLE…..cont.

• The present procedure involves hydrolysis of triglyerides by lipase. The glycerol concentration is then determined by enzymatic assay coupled with **Trinder reaction,** which measure the H_2O_2 activity,that terminates in the formation of a Quinoneimine dye. The amount of the dye formed, determined by its absorption as 505 ± nm is directly proportional to the concentration of triglycerides present in the sample.

Principle of the Test

The enzymatic reaction sequence employed in the assay of triglycerides is as follows:

Lipase

Triglycerides $+H_2O$ ------> Glycerol + 3 Free Fatty Acids

Glycerol Kinase

Glycerol + ATP --- -→ Glycerol – 3 Phosphate + ADP

Glycerol phosphate oxidase (GPO)

Glycerol -3 Phosphate + O_2 --- -→
Dihydroxyacetone phosphate + H_2O_2

Peroxidase

H_2O_2 + 4- Aminoantipyrine + 4- Cholorophenol -----—→
Quinoneimine Dye (red dye) + $2H_2O$

RANGE OF EXPECTED TAG VALUES IN SERUM

Normal distributions vary with age , the following concentrations if exceeded, clearly indicate hyperlipidemia.

0-29	**years**	**10-140 mg/dl**
30-30	**years**	**10-150 mg/dl**
40-49	**years**	**10-160 mg/dl**
50-59	**years**	**10-190 mg/dl**

(III) MATERIALS REQUIRED

REAGENTS COMPOSTION

- **R1. TRIGLYCERIDES BUFFER REAGENT :**
- Pipes Buffer 40 mmol/L. pH 7.5

  4-Chlorophenol 5.0 mmol /L. Magnesium –ions 5.0 mmol /L.

- **R1a, TRIGLYCERIDES ENZYME REAGENT:**
- ATP 3.3 mM, 4- Aminoantipyrine 0.7 mM, Glycero-30phosphate Oxidase 7000 U/L Sodium Azide 0.01% Lipase 200, 000 U/L

  Glycerol Kinase 100 U/L and peroxidase 3,000 U/L

- **TRIGLYCERIDES STANDARAD (200 MG/ DL AS Triolein) :**
- 2.2584 mmol/L of Glycerol with Surfactant. Sodium azide

  0.01% Added as a preservative.

(III) MATERIALS REQUIRED

- **MATERIALS**

- Tg- Buffer Reagent (R1),
- Tg – Enzyme Reagent (R1a)
- Tg Standard (200 mg/dl)
- Spectrophotometer,
- Cuvettes
- Pipettes
- Constant temperature incubator set at 37 oC
- Timer and Distilled water.

(III) MATERIALS REQUIRED

- **MATERIALS**
- **SPECIMEN**
- **SERUM**
- Fresh , non- hemolyzed serum from fasting pastients is recommended.
- Triglycerides in serum appear stable for 3 days when stored at 2-8 o C
- Prolonged storage of the samples at room temperature is not recommended since other glycerol containing compounds may hydrolyze, releasing free glycerol with an apparent increase in total triglycerides content.
- Blood collection devices lubricated with glycerin
- (glycerol) should not be used.

(IV) CALCULATIONS

- **CALCULATIONS**

- CONCENTRATION IN TEST (mg/dl) =

A= Absorbance

A (TEST) x CONC. OF STD (mg/dl) =

A (STANDARD)

- EXAMPLE : $\frac{0.17}{0.22}$ x 200 mg / dl = 154. 5mg/dl