Lipogenesis

Specific Learning Objectives
Lipogenesis

- What is Lipogenesis ?
- Which forms of Lipids biosynthesized?
- When Lipogenesis Occur?
- Where Lipogenesis takes place ?
- Why Lipogenesis Occurs ?
- How Lipogenesis is made possible?
- Associated Disorders to Lipogenesis ?

What Is Lipogenesis?

- Lipogenesis is **biosynthesis of various forms of Lipids in human body.**

Which Forms Of Lipid Biosynthesized In Human Body Tissues?

FORMS Of LIPID BIOSYNTHESIZED

1. Fatty acid Biosynthesis
2. Triacylglycerol Biosynthesis
3. Phospholipids Biosynthesis
4. Glycolipids Biosynthesis
5. Cholesterol Biosynthesis
6. Eicosanoids Biosynthesis

When Lipogenesis Occurs?

- **Lipogenesis occurs in well fed condition.**

Conditions Favoring Lipogenesis

- **Excess of Free Glucose** after heavy Carbohydrate meals.
- **Insulin** promotes Lipogenesis

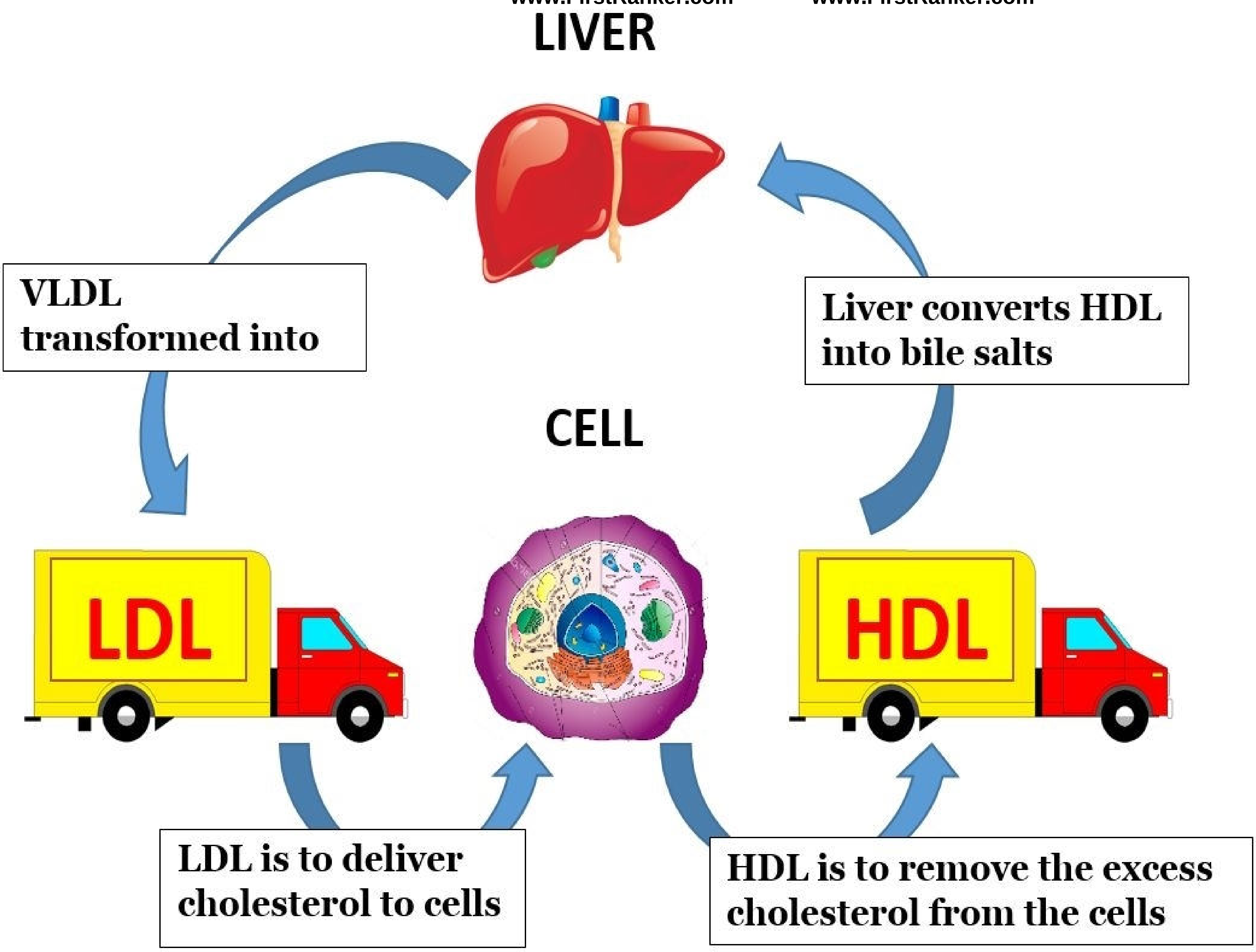
Where Does Lipogenesis Occur?

Site Of Lipogenesis

- **Predominant site** for Lipogenesis
- **Liver Cytoplasm**

- **Other tissues for Lipogenesis**
  - **Intestine**
  - **Mammary glands**

Lipoprotein VLDL
Mobilizes Out and Transport
Endogenously Biosynthesized Lipids
From Liver To Extra hepatocytes



- **Endogenously biosynthesized Lipids at Liver** are
- **Gathered and mobilized out** in a form of **Lipoprotein VLDL to extrahepatic tissues.**

- **VLDL** carries **endogenous Lipids rich in TAG** from **Liver to extra Hepatocytes.**

- **TAG is stored** as reserve food material in **Adipose tissue in an unlimited amount.**

Why Lipogenesis Takes Place?

Excess Carbohydrates Are Transformed To Triacylglycerol (Fat)

Reasons For Lipogenesis

- **Free excess Glucose cant be stored as it is** in body cells and tissues.

- Free excess Glucose is **first converted and stored in the form of Glycogen**

- **Storage of Glycogen is limited**

- In a well fed condition **after limited storage of Glycogen**
- **When still there remains Free excess Glucose**
- This free excess Glucose is Oxidized to Pyruvate via Glycolysis
- Further Pyruvate to **Acetyl-CoA via PDH complex reaction**
- **This Acetyl-CoA when excess is then diverted for Lipogenesis.**

- Thus **Lipogenesis occur** in a **well fed condition**
- **To transform free excess Glucose to Acetyl-CoA further into Fatty acids.**
- **Fatty acids are stored as TAG**
- Storageable form of Lipid (TAG).
- **TAG in Adiposecytes** can be stored in **unlimited amounts**.

Hormonal Influences On Lipogenesis

- **In a well fed condition**
- Hormone **Insulin stimulates Lipogenesis**.
- Hormone **Glucagon inhibits Lipogenesis.**

Alterations Of Lipogenesis In Clinical Conditions

- **Inhibition of lipogenesis** occurs in **Type 1 (insulin-dependent) Diabetes mellitus**
- **Variations in Lipogenesis** affect nature and extent of **obesity**

How Lipogenesis Occur?

- Complex Mechanism
- Tissue Specific
- Compartmentalized
- Regulated

Precursors For Lipogenesis

Precursors For Lipogenesis

- **Acetyl-CoA** serve as a **precursor for Fatty acids and Cholesterol biosynthesis.**
- This Acetyl-CoA comes **from excess and free Glucose Oxidation in a well fed condition.**

- Phospholipid biosynthesis **needs Lipotropic factors.**

De Novo Biosynthesis Of Fatty Acids

- Fatty acid biosynthesis is a **reductive biosynthetic mechanism**.
- To form **reduced molecules** of **Fatty acid (Palmitate).**

- De novo biosynthesis of Fatty acids is a **new biosynthesis of Fatty acids.**

- Using simple carbon units **Acetyl-CoA** and **reducing equivalents** as **NADPH+H$^+$** to a long chain fatty acids.

- Palmitic acid (16:0) can be **further modified to higher Fatty acids .**

Site For Fatty Acid Biosynthesis

Organs Involved For Fatty Acid Synthesis

- In humans, Fatty acids are biosynthesized in **Cytosol** of:
  - **Liver (Predominantly)**
  - Adipose tissue
  - Intestine
  - Lungs
  - Brain
  - Renal Cortex
  - Mammary glands during lactation

Reductive Biosynthesis Of Fatty acids

Extra Mitochondrial/Cytosolic Biosynthesis of Fatty acids

- Biosynthetic pathway of Fatty acids involves
- Use of **reducing equivalents NADPH+H$^+$** in **reduction steps.**

- To form **reduced molecule** of **fatty acids,**
- **Hence** it is **termed as reductive Synthesis of Fatty acids.**

- Fatty acids biosynthesized are **later used up for biosynthesis of :**
  - **Triacylglycerol**
  - **Phospholipid**
  - **Glycolipid**
  - **Cholesterol Ester**

- **Fatty acids are stored as Triacylglycerol,** especially **in Adipose tissue.**

Biosynthesis Of Palmitic Acid/Palmitate (C16)

Requirements Of De Novo Biosynthesis Of Palmitate

Prerequisites for Fatty acid Biosynthesis

- Immediate Substrate/Hydrocarbon Units
- Enzyme Systems
- Coenzymes and Cofactors

- Precursor for Palmitic acid biosynthesis are 8 molecules of Acetyl-CoA

Source
Of Acetyl-CoA for Fatty acid Biosynthesis ?

- **Free and Excess Glucose** in a **well fed condition**

- Is **major source of carbon/Acetyl-CoA for De novo fatty acid biosynthesis**.

- **Free and excess Glucose** remained **after limited Glycogen storage**

- Is used for **Acetyl-CoA production** and **diverted for Fatty acid biosynthesis.**

- **Glucose** is oxidized **to Pyruvate** via **Glycolysis.**

- **Pyruvate**(3C) is then **oxidatively decarboxylated**

- To a high energy compound **Acetyl-CoA** (2C)in **Mitochondria** by **PDH Complex.**

- **Excess of Acetyl CoA** formed and **present in Mitochondrial matrix**

- Is **diverted for Denovo Biosynthesis of Fatty acids**.

- **8 molecules of Acetyl-CoA** (C2) are **required**

- **For biosynthesis** of **1 molecule** of **even carbon Palmitate (C16).**

- **Enzymes Involved:**
  - **Acetyl-CoA Carboxylase**
  - **Fatty Acyl Synthase (FAS) Multi Enzyme Complex**

Coenzymes and Cofactors for Fatty acid Biosynthesis

- Bicarbonate ions
- Biotin
- NADPH+ H^+
- ATP
- Mn $^+$

- **Requirement of $HCO3^-$** (Bicarbonate Ions) : Provides CO2 **for Acetyl-CoA Carboxylation Reaction.**

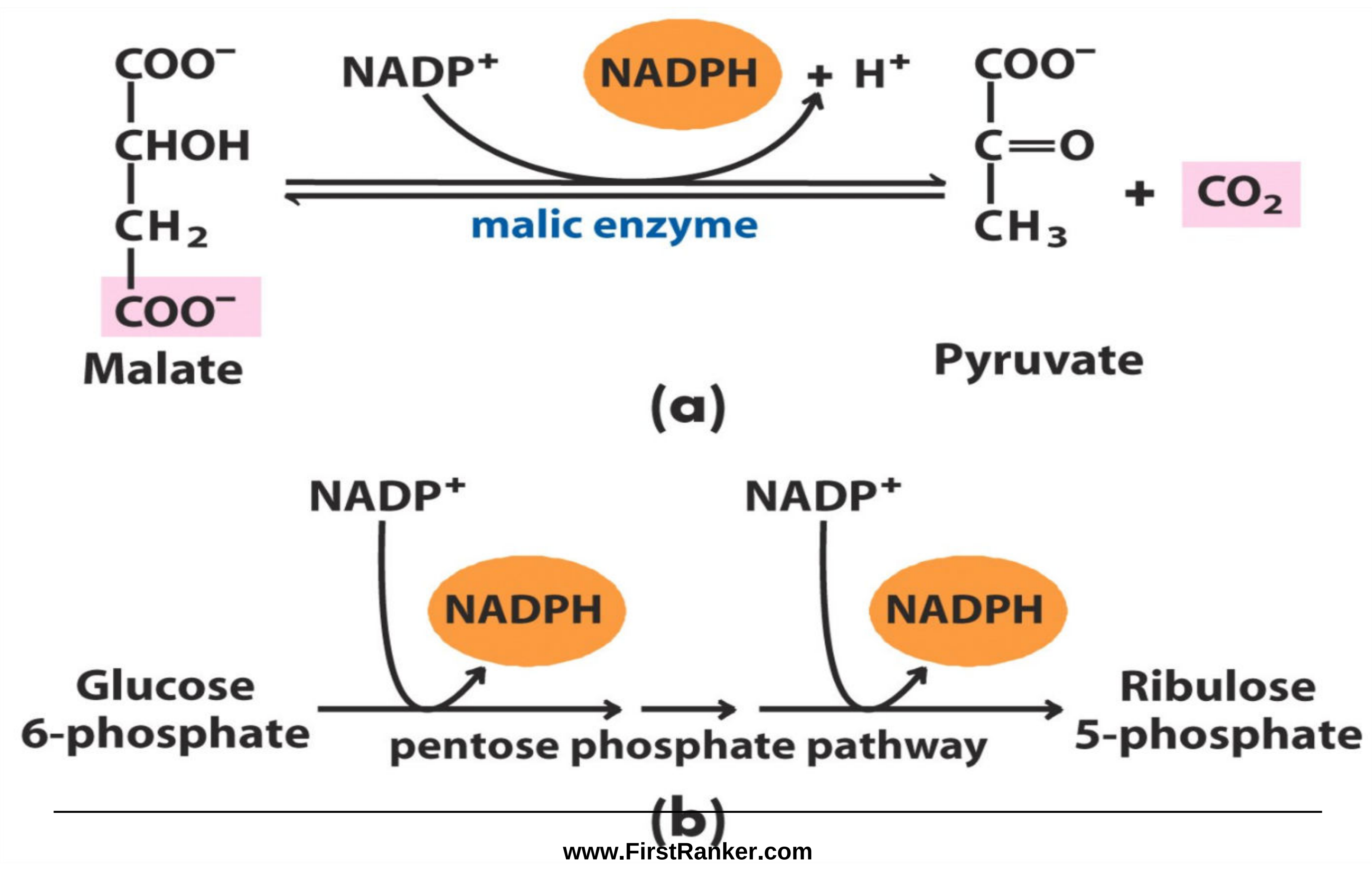
Sources of Coenzyme Required

- **Reducing Equivalent :**
  - **$NADPH+H^+$**

❑ **Main source** of **$NADPH+H^+$** is mainly by **Pentose Phosphate Pathway.**

❑ **Another source of $NADPH+H^+$ Malic enzyme activity** converts Malate to Pyruvate which is

Production of $NADPH+H^+$



- **NADPH+H⁺** serves as an electron donor in two reactions
- **Involving substrate reduction in De Novo Fatty acid biosynthesis.**

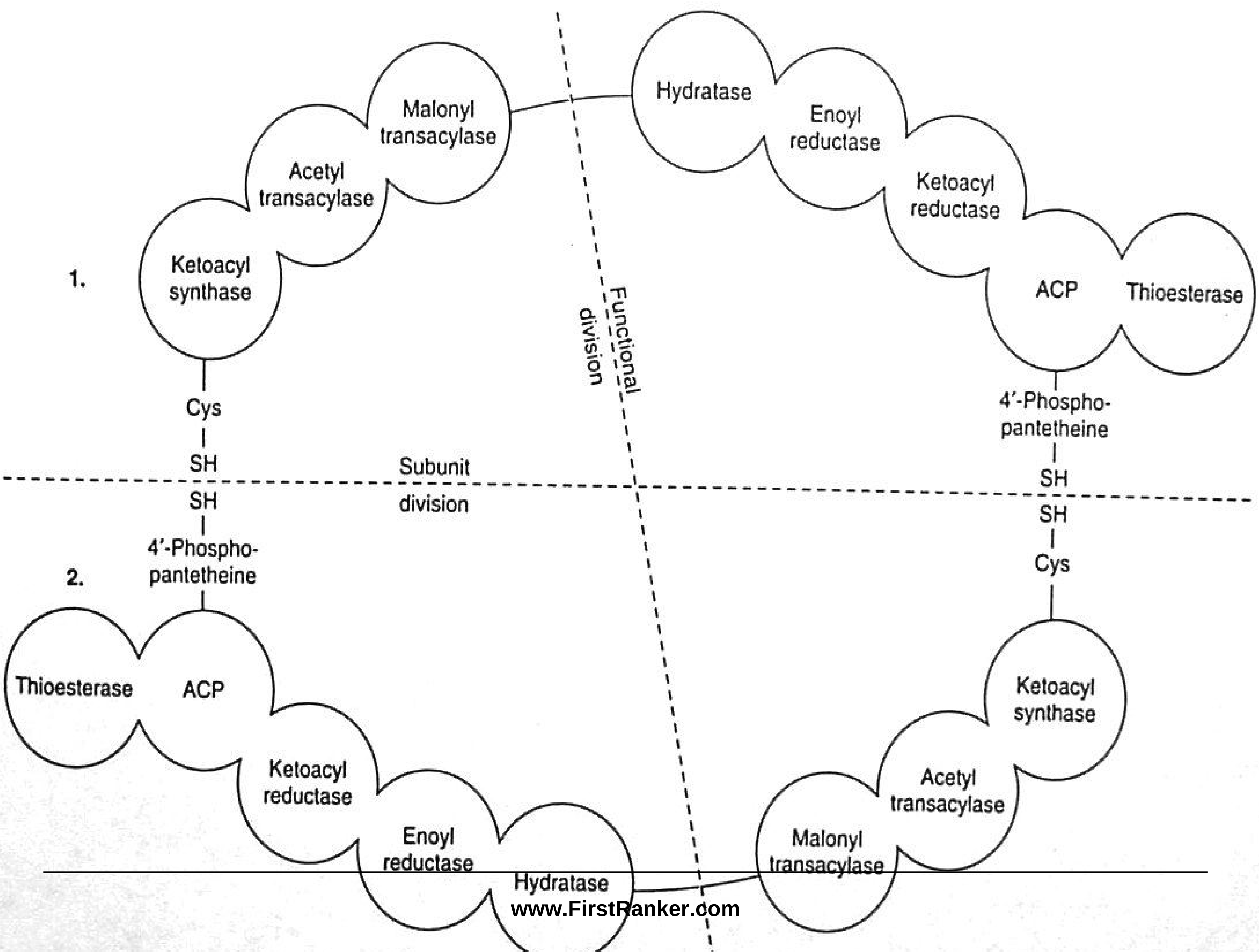
Fatty Acyl Synthase (FAS) Multi Enzyme Complex

For De Novo Biosynthesis Of Fatty Acids

Acyl Carrier Protein

Carrier of Intermediates in Fatty acid biosynthesis

- Discovered by **P. Roy Vagelos.**



Fatty Acyl Synthase (FAS) Complex

- **FAS** is a **Multi Enzyme Complex Used in De Novo Biosynthesis of Fatty acids.**

- Structurally **FAS is a Homodimer**

- **Two alike monomeric subunits**

- **Linked together** in **head to tail fashion (Anti Parallel)**

Structural Aspects Of FAS

- FAS is Composed of 8 Components in one subunit.
  - 7 Enzymes and 1 Protein

Three Subunits/Domains Of FAS Complex

1.Condensation Unit

Has 3 Enzymes

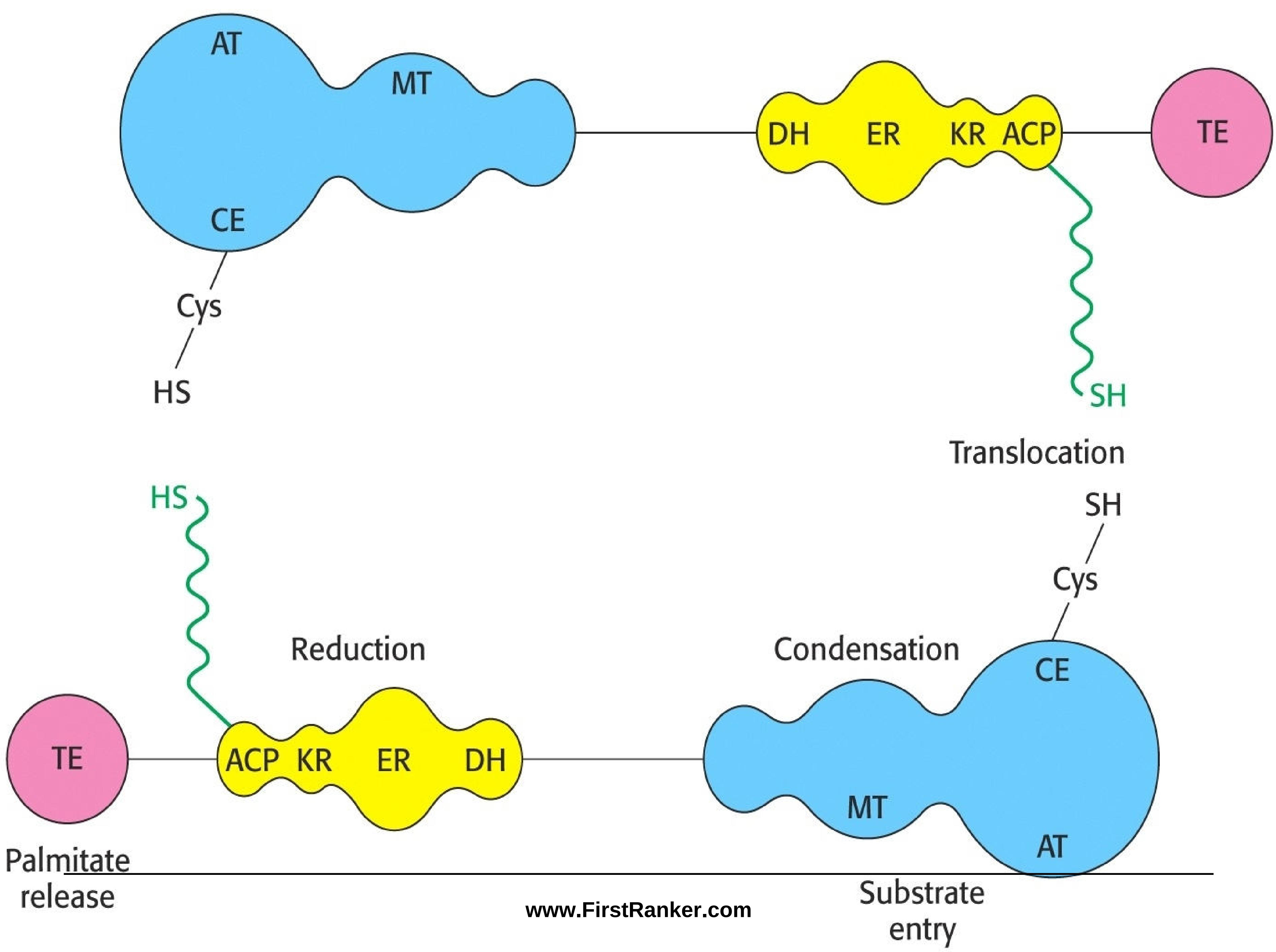
- Acetyl Transacylase
- Malonyl Transacylase
- Beta Keto Acyl Synthase

2. Reduction Unit

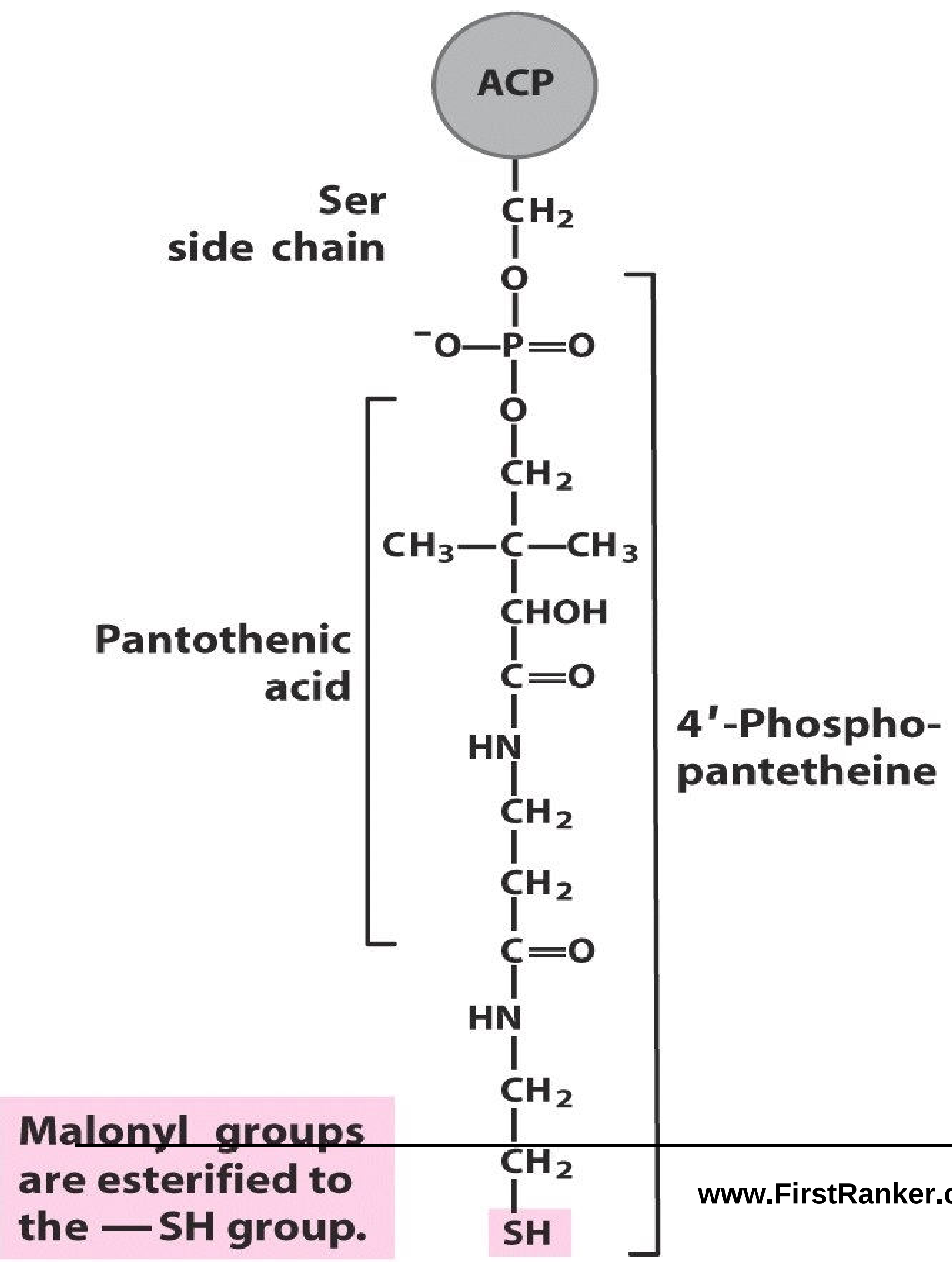
- ACP-(Acyl Carrier Protein)
- Beta Keto Acyl Reductase
- Dehydratase
- Enoyl Reductase

3. Cleavage /Releasing Unit

- **Thioesterase (Deacylase)**



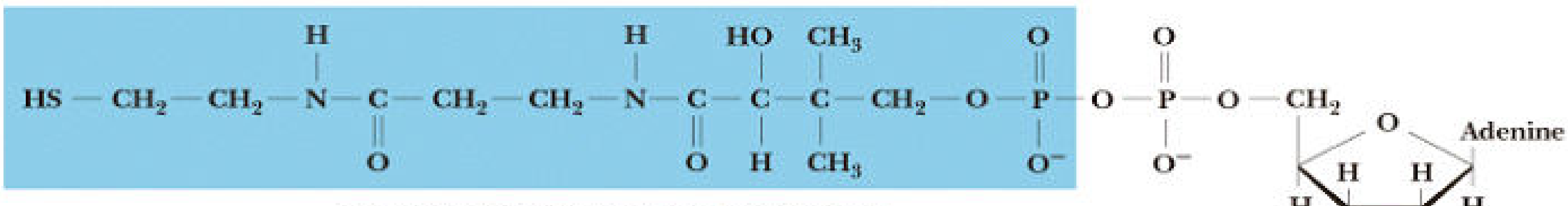
- In terms of function, **ACP is a large CoA.**



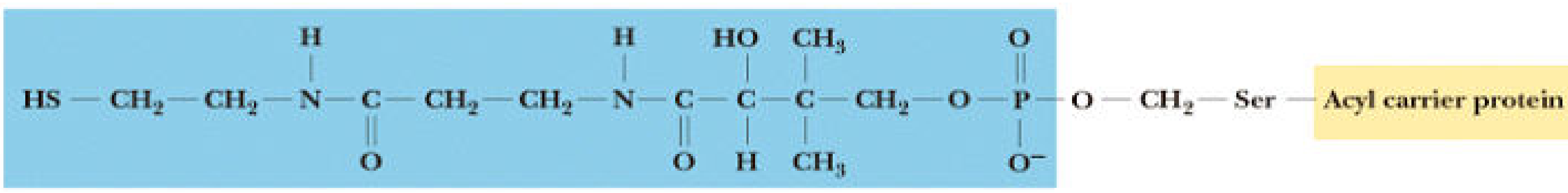
Key Player:
Acyl Carrier Protein(ACP)

- "Macro" CoA, carries growing fatty acid chain via Thioester

ACP Vs. Coenzyme A



Phosphopantetheine group of coenzyme A

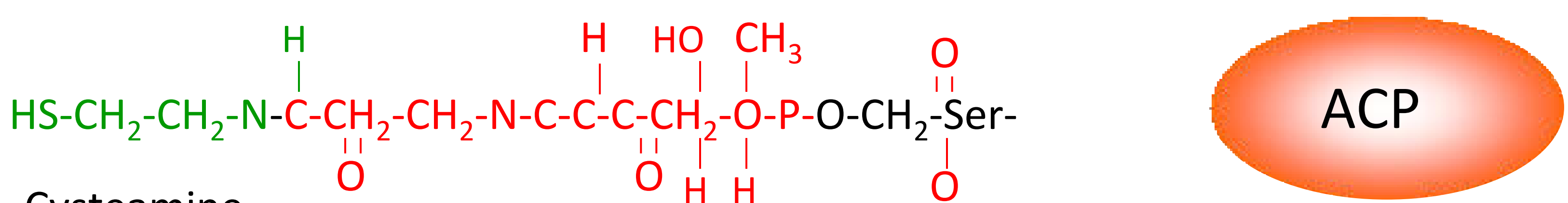


Phosphopantetheine prosthetic group of ACP

•Intermediates in synthesis are linked to -SH groups of Acyl Carrier Proteins (as compared to -SH groups of CoA)

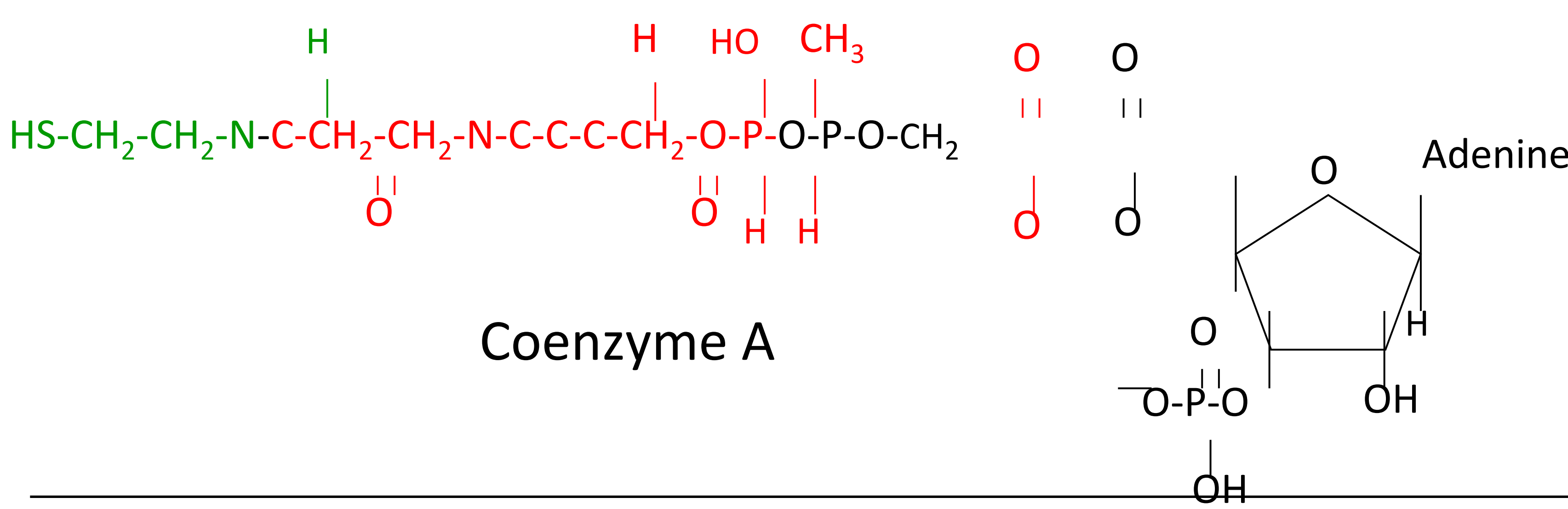
Acyl Carrier Protein

Phosphopantetheine



Cysteamine

Acyl carrier protein
10 kDa



Coenzyme A

- **ACP** is a **Conjugated Protein component of FAS complex.**

- ACP is a part of **Reduction unit** of **FAS complex.**

- **4- Phospho Pantethene** serves as a **prosthetic group of ACP.**

- **4-Phospho Pantethene** is a **derivative of Vitamin B 5- Pantothenic acid.**

- **4 Phosphopantetheine** (Pant) is covalently linked to **Serine hydroxyl** of Protein domain of ACP via a **phosphate ester linkage .**

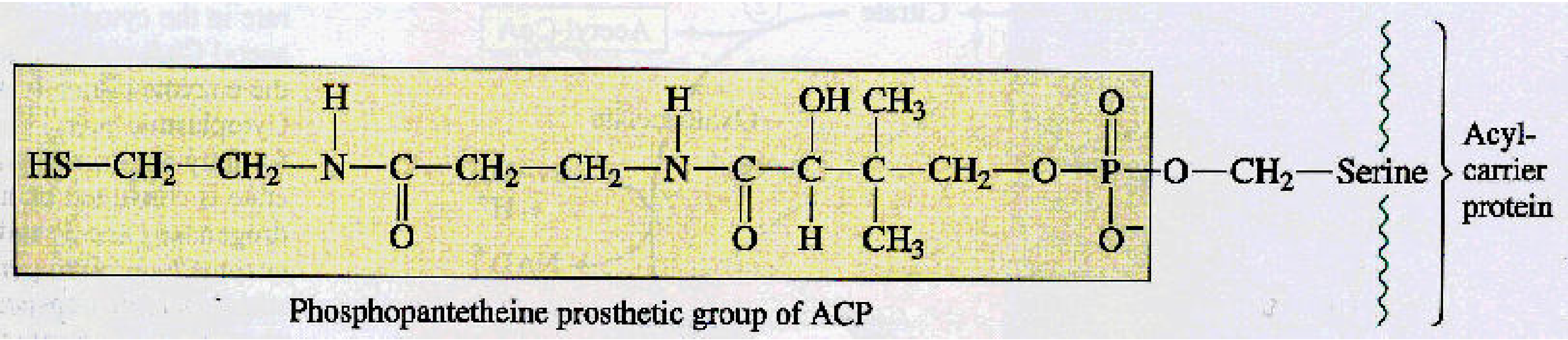
- ACP has **–SH group (Thiol)** as **functional group**.

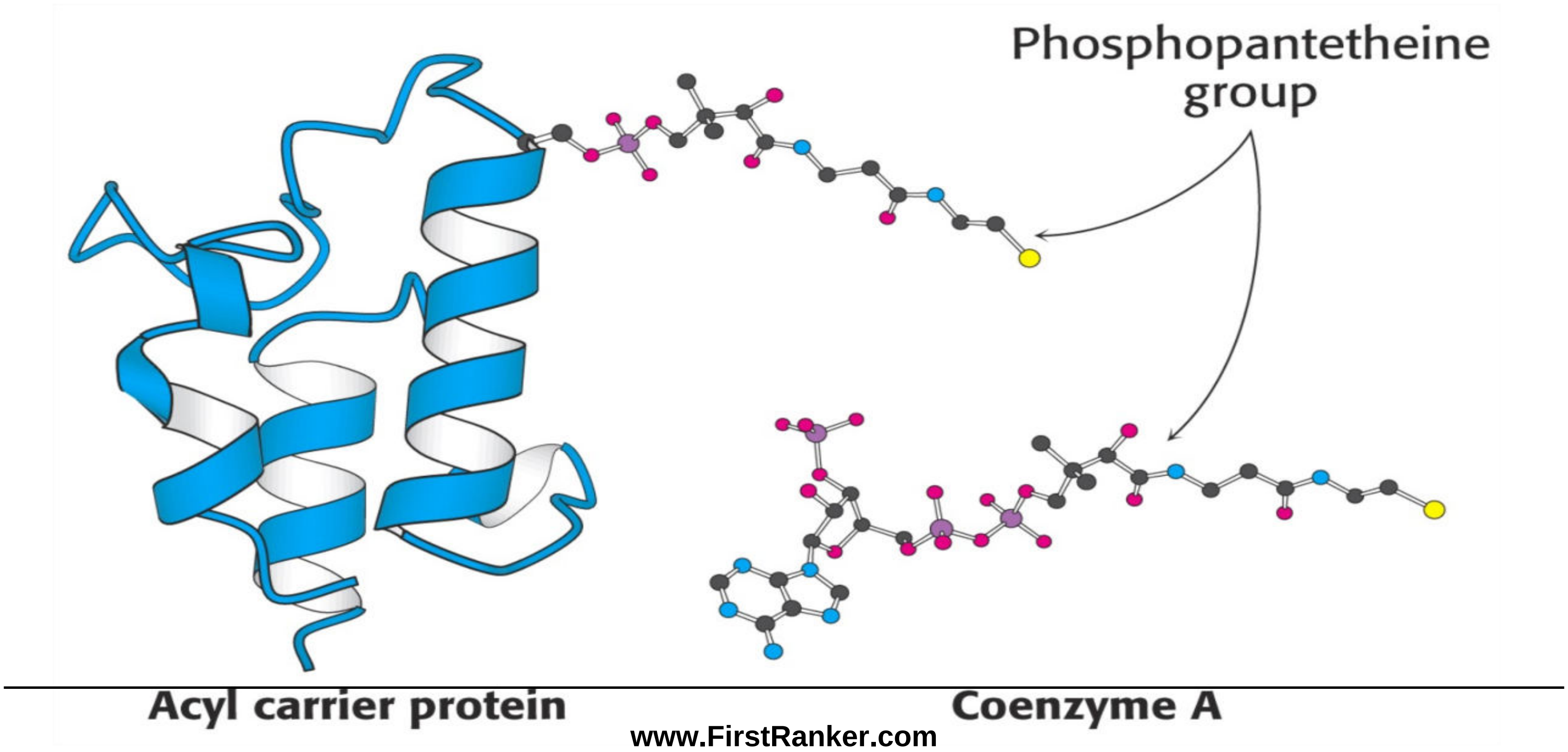
- **-SH group of ACP** is **an acceptor of Acetyl-CoA and Malonyl-CoA** during De novo biosynthesis of a Fatty acids.

Role of ACP In FAS Complex

- During **De novo biosynthesis of fatty acids.**
- **Acyl Carrier Protein** (ACP) of FAS complex is a **carrier of growing Acyl chain**
- At **end of Denovo Fatty acid biosynthesis**
- **Complete chain** of **Fatty acid** is **linked to ACP** of FAS complex.

- **Long flexible arm** of Phosphopantetheine **helps its Thiol**
- **To move from one active site to another within FAS complex.**





FAS Complex Is Coded By Single Gene

Advantage Of Multi Enzyme Subunits To Achieve

- An effect of compartmentalization of process
- Good coordination and Communication
- Speed of reactions
- Quality product

Location Of FAS Complex

- Cytosol
- Extra mitochondrial

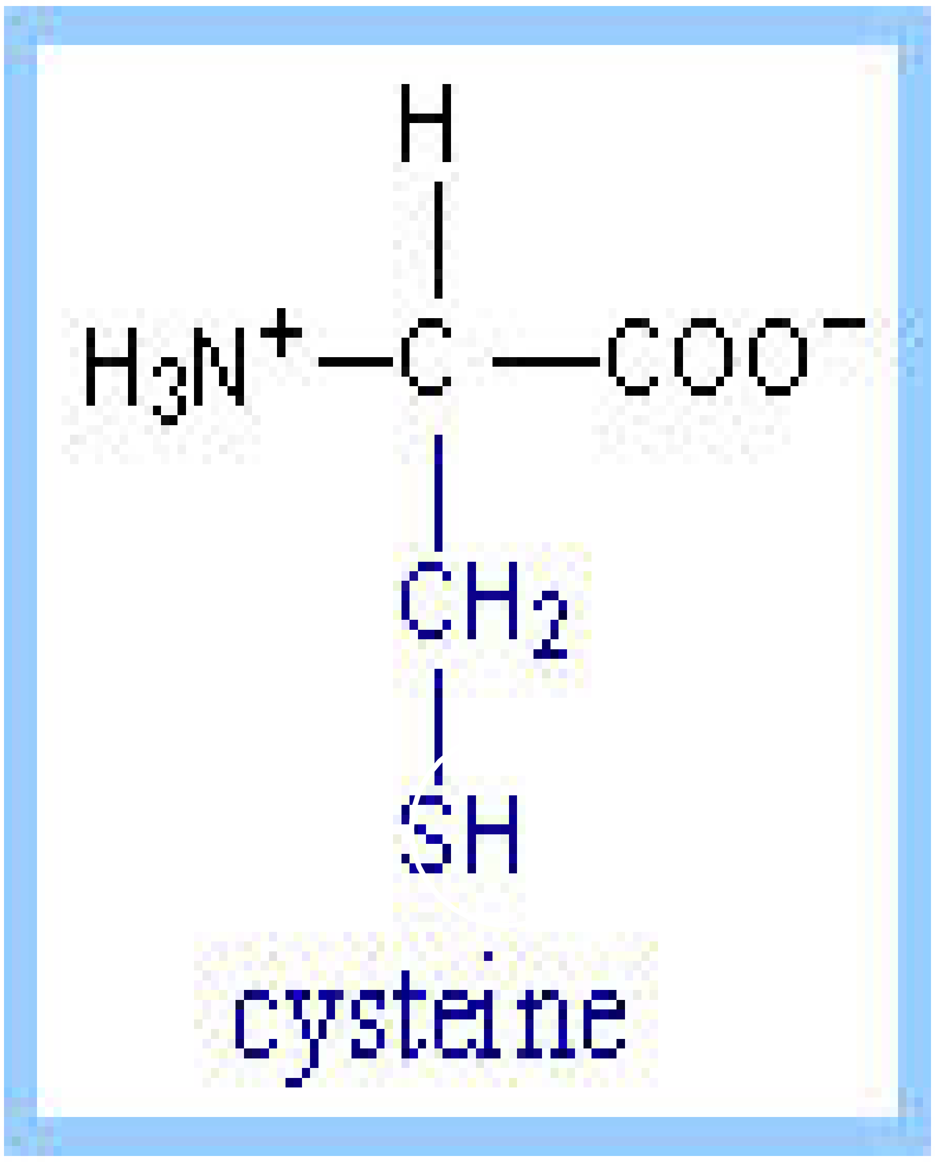
Hormones Regulating FAS Complex

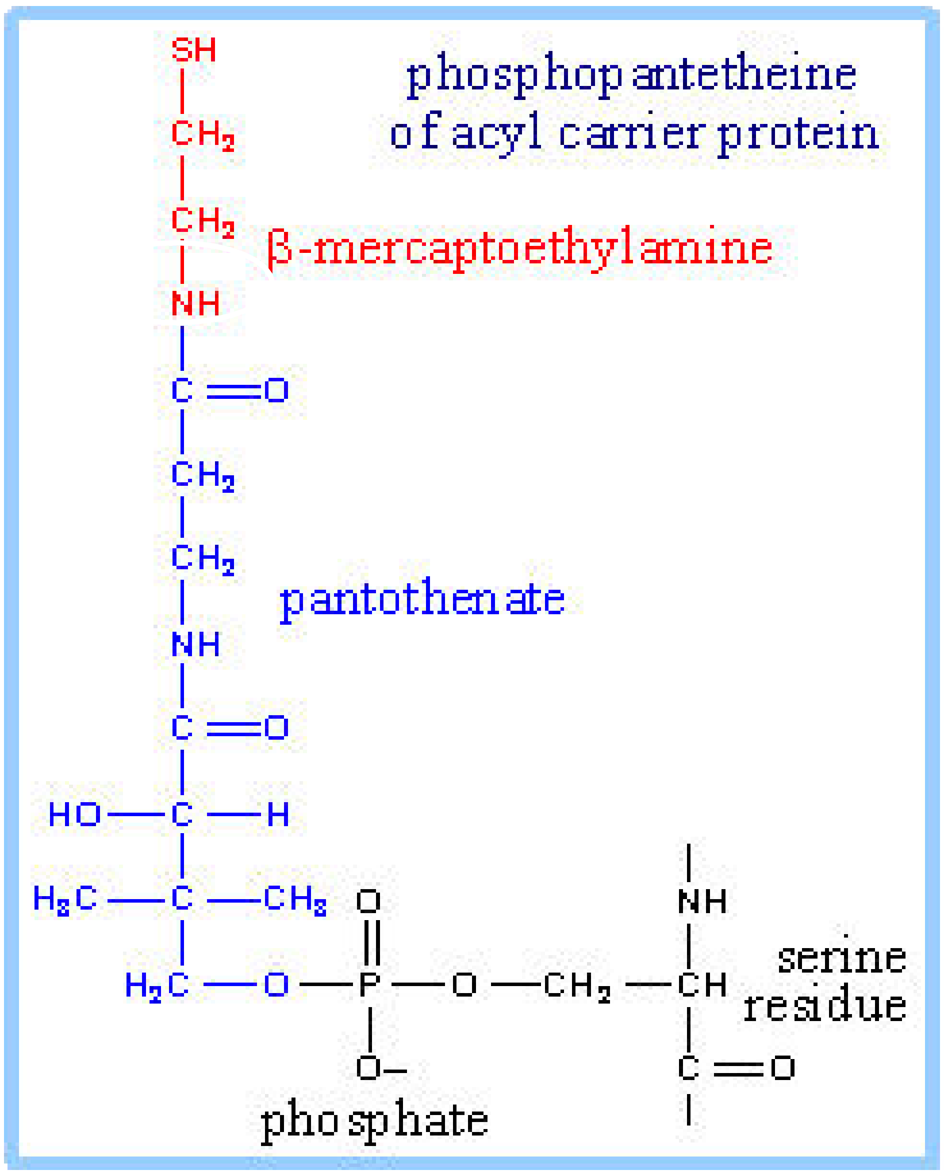
- **Insulin**- Stimulates FAS Complex
- **Glucagon**-Inhibits FAS Complex

Functional Parts Of FAS Complex

- **FAS complex** being **dimer has two functional Units.**
  - **-SH (Thiol) group of Cysteine** of condensation Enzyme **β Keto Acyl Synthase.**
  - **-SH (Thiol) group of 4 Phospho Pantethene of ACP**.

Thiol Cysteine residue **Thiol of Phosphopantetheine**

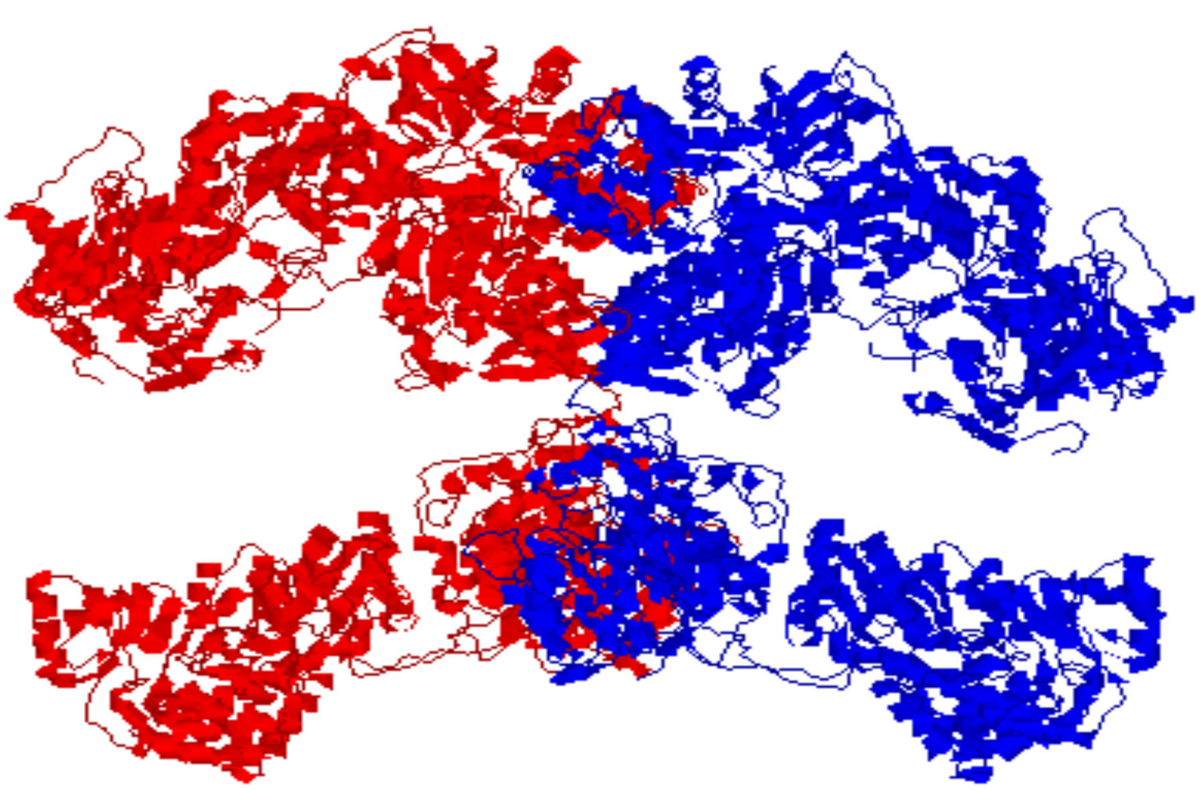




- As there are **two functional units**
- When **FAS complex operates at a time**
- There is **biosynthesis of two Fatty acids (Palmitate) molecule.**

- **Rate of Fatty acid biosynthesis is high in well-fed state.**

X-Ray crystallographic analysis at 3.2 Å resolution shows the Dimeric Fatty Acid Synthase to have an **X-shape**.



Fatty Acid Synthase PDB 2VZ8

Fatty Acid Synthase Complex

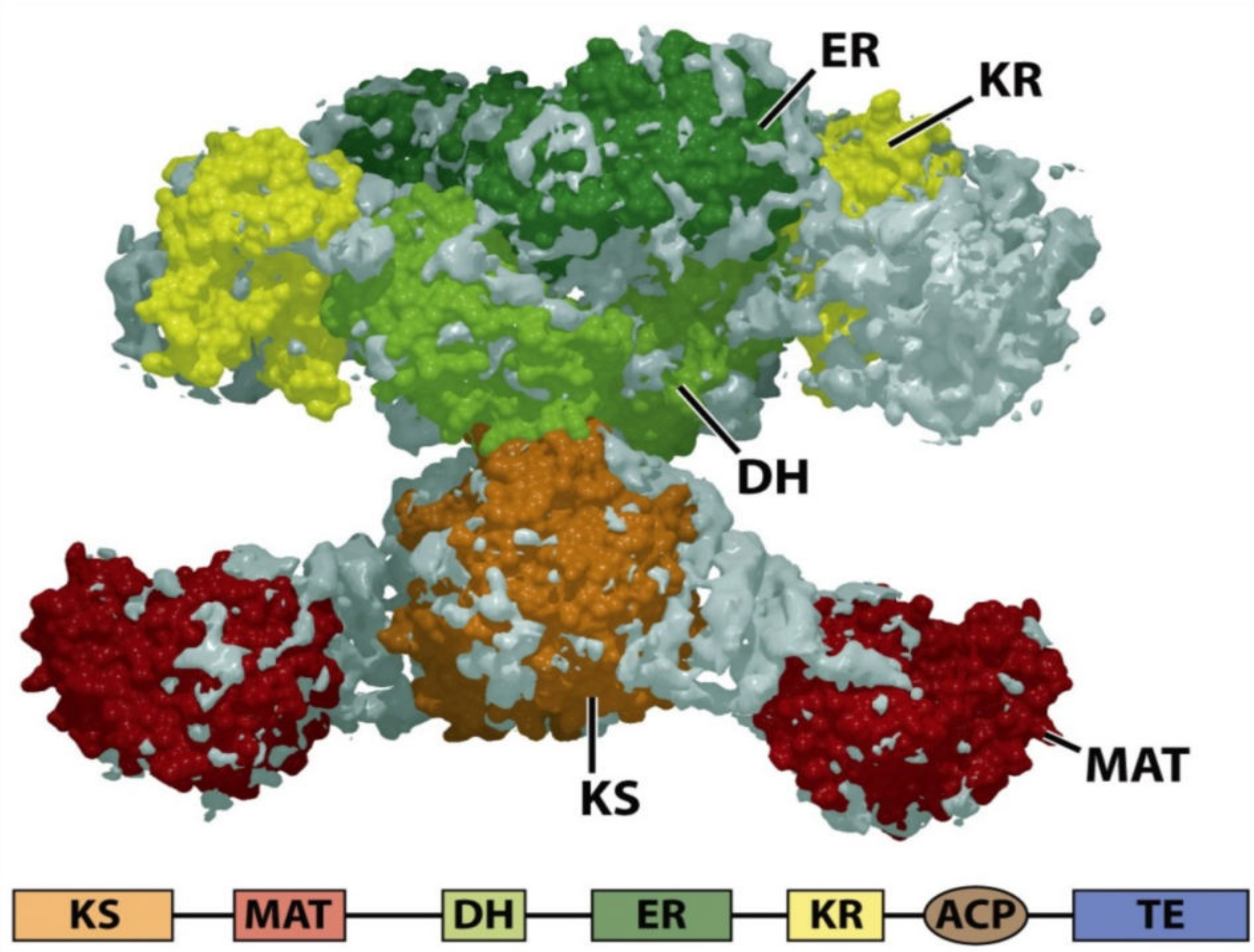
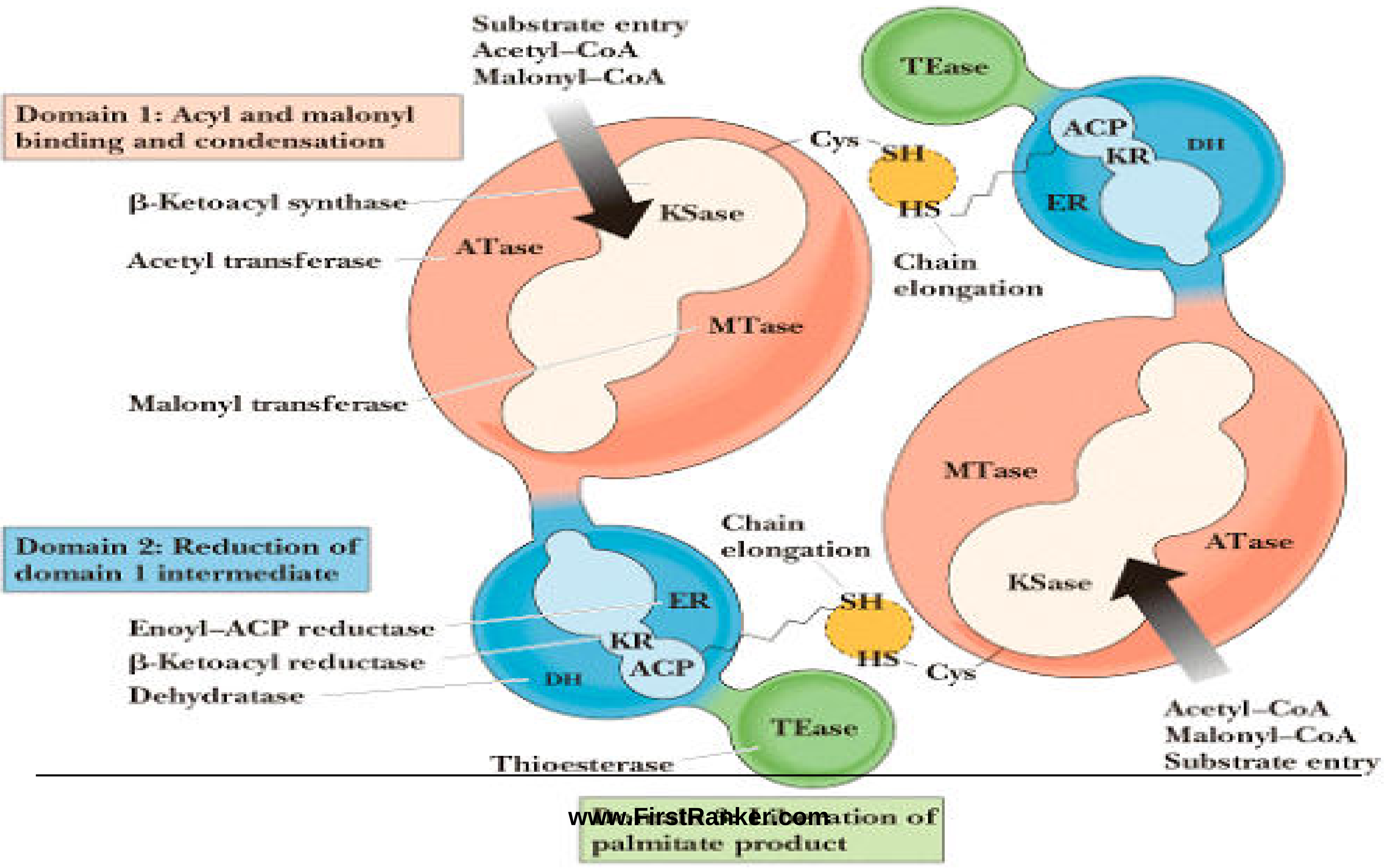


Figure 21-3a
Lehninger Principles of Biochemistry, Fifth Edition

Fatty Acid Synthase Complex



Stages And Steps
Of De Novo Biosynthesis
Of Fatty Acids

Three Stages
Of
De novo Biosynthesis
Of Fatty acid

I. Translocation of Acetyl-CoA from Mitochondria to Cytosol.

II. Carboxylation of Acetyl-CoA to Malonyl-CoA

III. Reactions of FAS Complex

Stage I

Translocation of Acetyl-CoA from Mitochondria to Cytosol

Transport Of Mitochondrial Acetyl-CoA To Cytosol

Since

Fatty Acid Synthesis

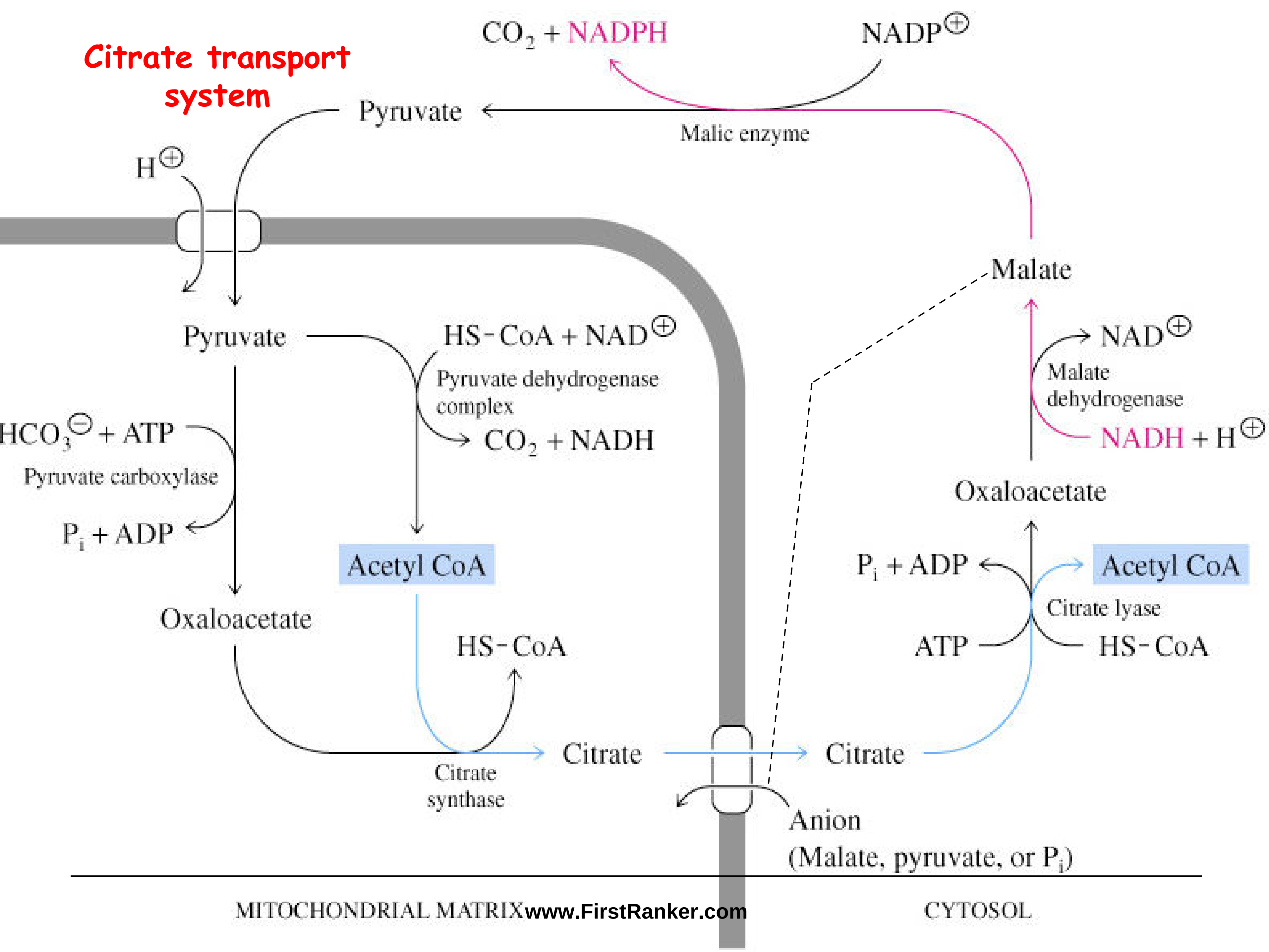
Occurs in the Cytosol

Mitochondrial Acetyl-CoA

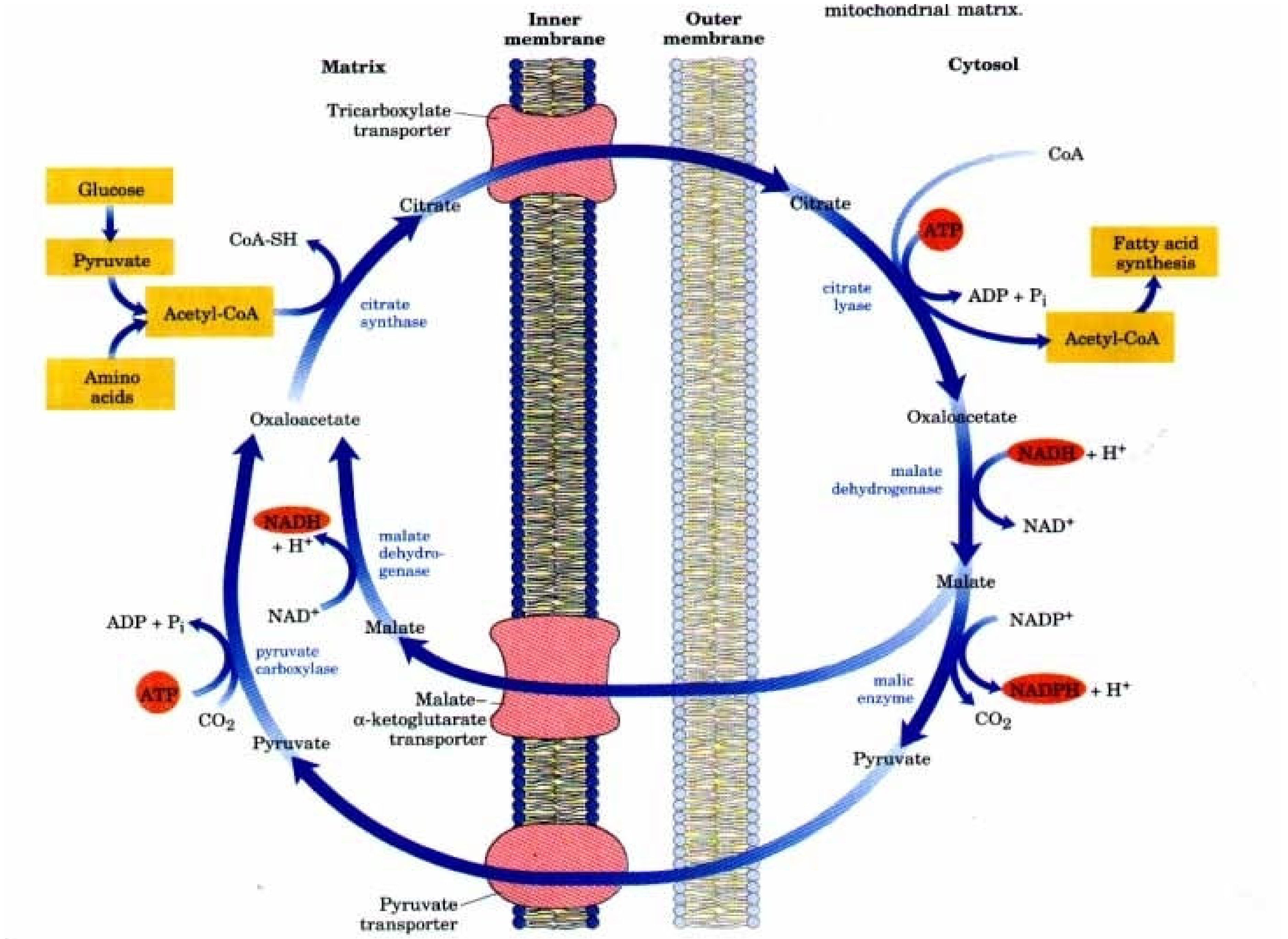
Is to be translocated In Cytosol

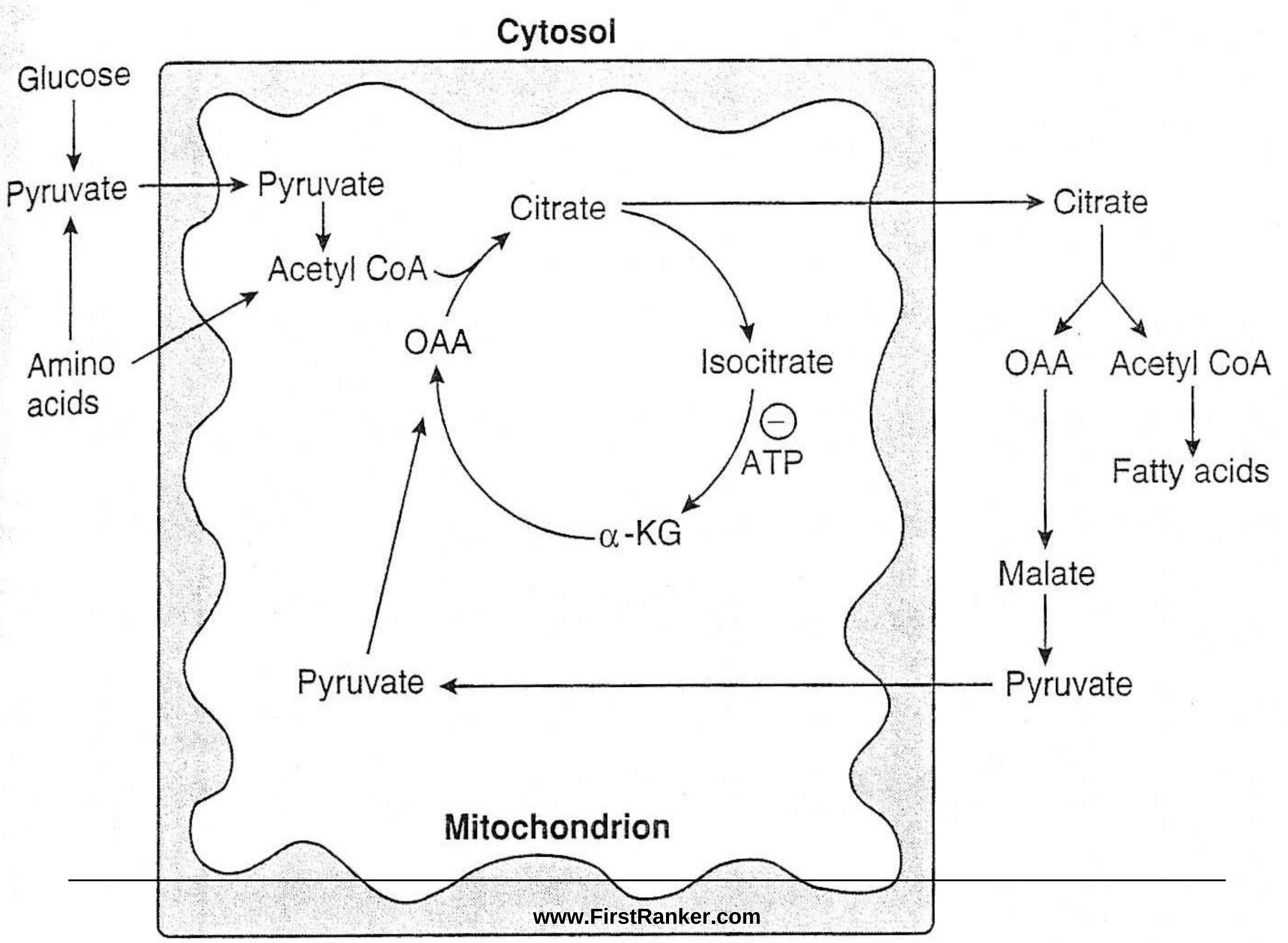
Translocation Of Acetyl-CoA Through Citrate Shuttle

Citrate Malate Pyruvate Transport System



mitochondrial matrix.





- Mitochondrial **Acetyl CoA** is **impermeable** due to the **complex CoA** .
- Impermeable Acetyl CoA is transformed to **permeable** **Citrate** by **Citrate Synthase.**

- Citrate is translocated out in cytosol.
- Citrate in cytosol is cleaved by **Citrate Lyase** to **liberate Acetyl-CoA in cytosol.**

Significance Of Citrate Malate Pyruvate Shuttle

- **Citrate-Malate-Pyruvate shuttle during De novo Fatty acid biosynthesis :**
  - **Translocate Acetyl CoA to cytosol**
  - **Provides reducing equivalents NADPH+H+**

- **Acetyl CoA** from **catabolism of Carbohydrates and Amino acids** is **exported from Mitochondria** via the **Citrate transport system**

- **2 ATPs are required during work of this system.**

- Impermeable **Acetyl-CoA** is translocated out

- **From Mitochondrial Matrix into Cytosol** in the form of **permeable Citrate**.

– **Acetyl-CoA**(impermeable) produced in the **Mitochondria** is condensed with **Oxaloacetate** to form **Citrate**(permeable) by **Citrate Synthase**.

- **Permeable Citrate** is then transported out into **Cytosol**

- **Citrate Lyase in Cytosol act upon Citrate to regenerate Acetyl-CoA** and **Oxaloacetate** with consumption of **ATP**

- **Most Acetyl-CoA used for FA synthesis comes from Mitochondria.**

Stage 2

Carboxylation of Acetyl-CoA to Malonyl-CoA In Cytosol

Fatty Acid Biosynthesis Initial Controlling Step

Carboxylation of Acetyl-CoA (2C) to Malonyl-CoA (3C) By Acetyl CoA Carboxylase (ACC)

Acetyl-CoA(2C) Units Are Activated To Malonyl-CoA(3C)

For Transfer To Growing Fatty Acid Chain

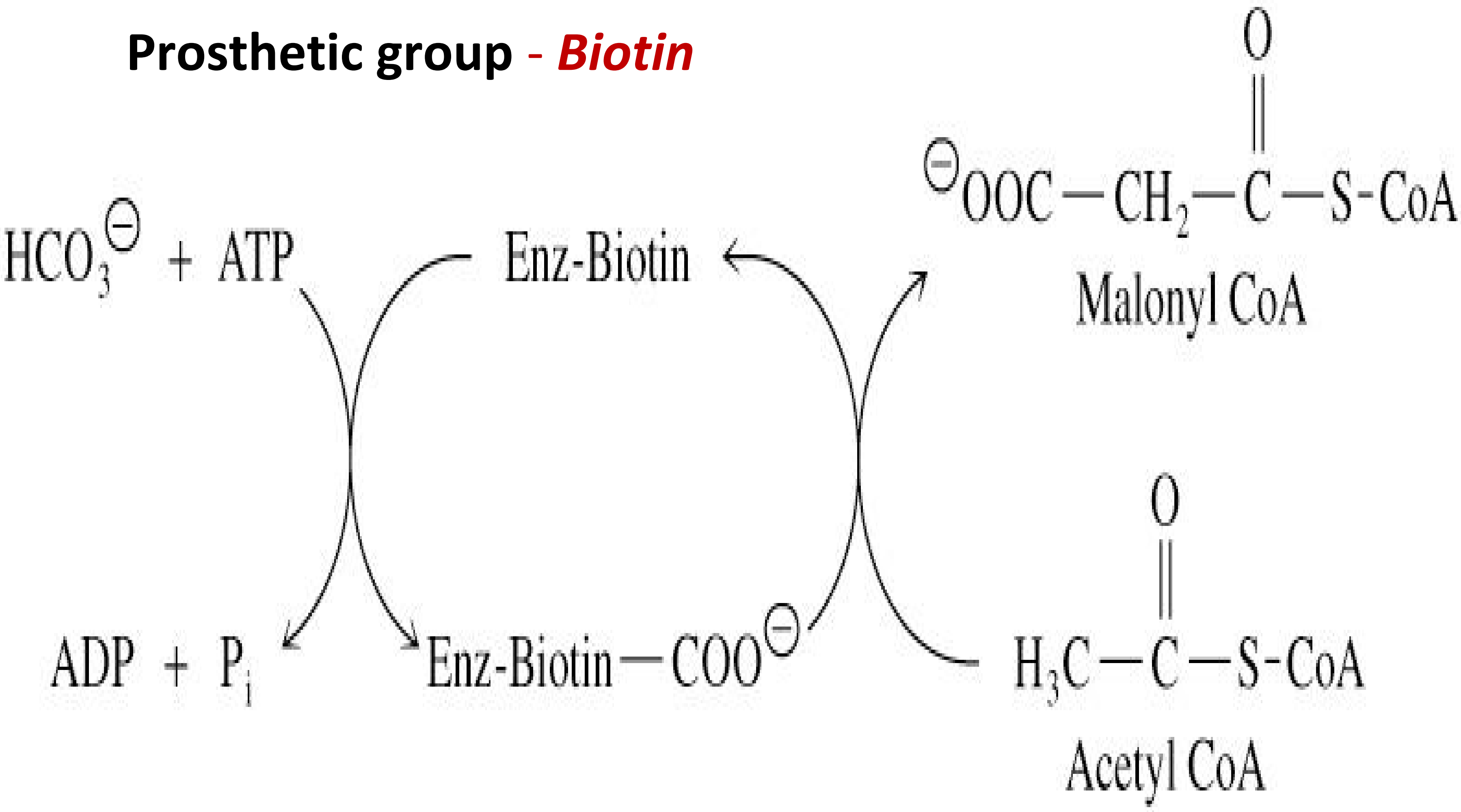
Malonyl-CoA (3C) Is a High Energy Compound

With a High Energy Bond In Its Structure

B. Carboxylation of Acetyl CoA

Enzyme: ***Acetyl CoA Carboxylase***

Prosthetic group - ***Biotin***



- During **biosynthesis** of 16 C saturated **Palmitic acid**
- There **requires total 8 molecules of Acetyl-CoA**

- **During FAS complex** Fatty acid **synthetic steps**

- **Only one molecule of Acetyl-CoA** (C2) **enters as it is in first step of Third Stage of Fatty acid biosynthesis.**

- **Remaining 7 molecules of Acetyl-CoA are entered in form of Malonyl-CoA (C3).**

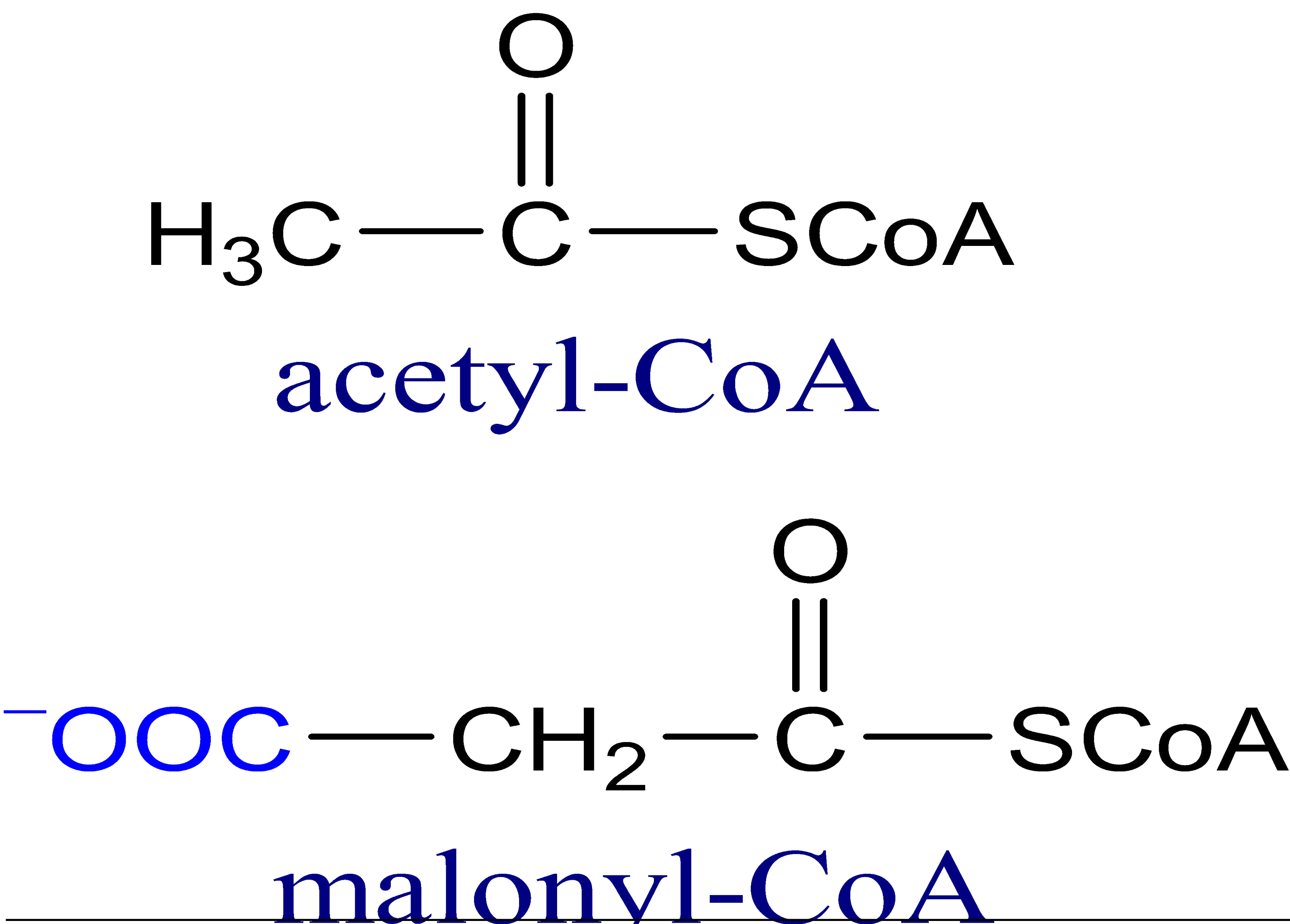
- Thus **Seven Molecules** of **Acetyl-CoA** are
- **Transformed to Seven molecules of Malonyl-CoA.**

- **Malonyl-CoA** is obtained from **carboxylation reaction** of Acetyl-CoA
- In presence of, enzyme **Acetyl Carboxylase** and **coenzyme Biotin and ATP.**

- Conversion of Acetyl-CoA to Malonyl CoA , is by catalytic activity of **Acetyl CoA Carboxylase , Biotin and ATP.**
- This is an Carboxylation reaction which **provides energy input.**
- To form still more high energy compound **Malonyl-CoA(C3).**

- This carboxylation reaction after **use of high energy ATP**
- Builds a high energy bond in a **high energy compound Malonyl-CoA.**

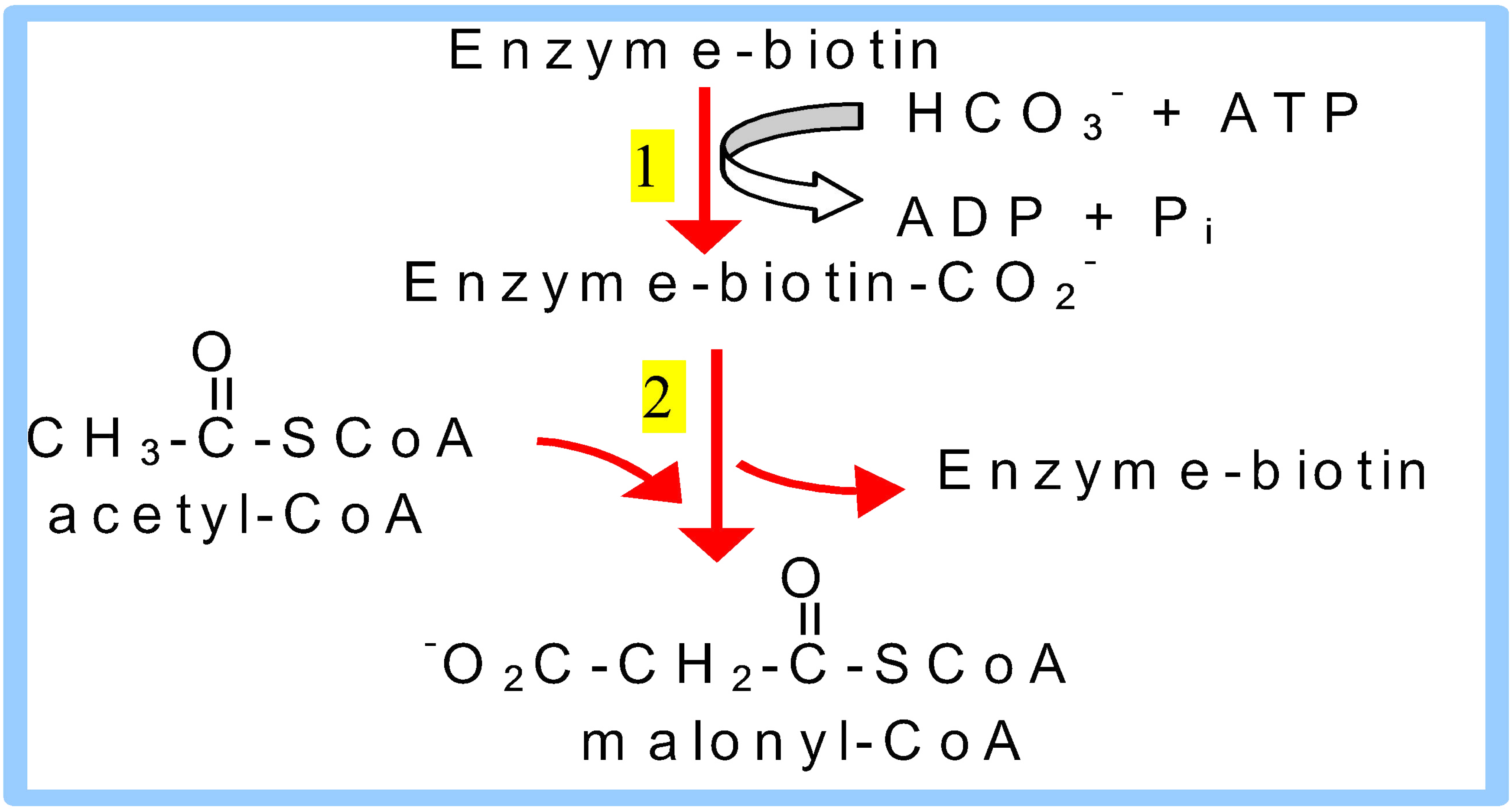
- Input of **Acetyl-CoA**, into Fatty acid biosynthesis is by its Carboxylation to **Malonyl-CoA**.



❖Later this **Malonyl CoA cleaves its high energy bond and looses** CO_2 and energy

❖This **released energy is used for condensation reaction during third stage of Fatty acid biosynthesis** for initiation and growing of Fatty acid.

- Thus **spontaneous Decarboxylation** of **Malonyl-CoA**
- **Drives condensation reaction of FAS complex.**

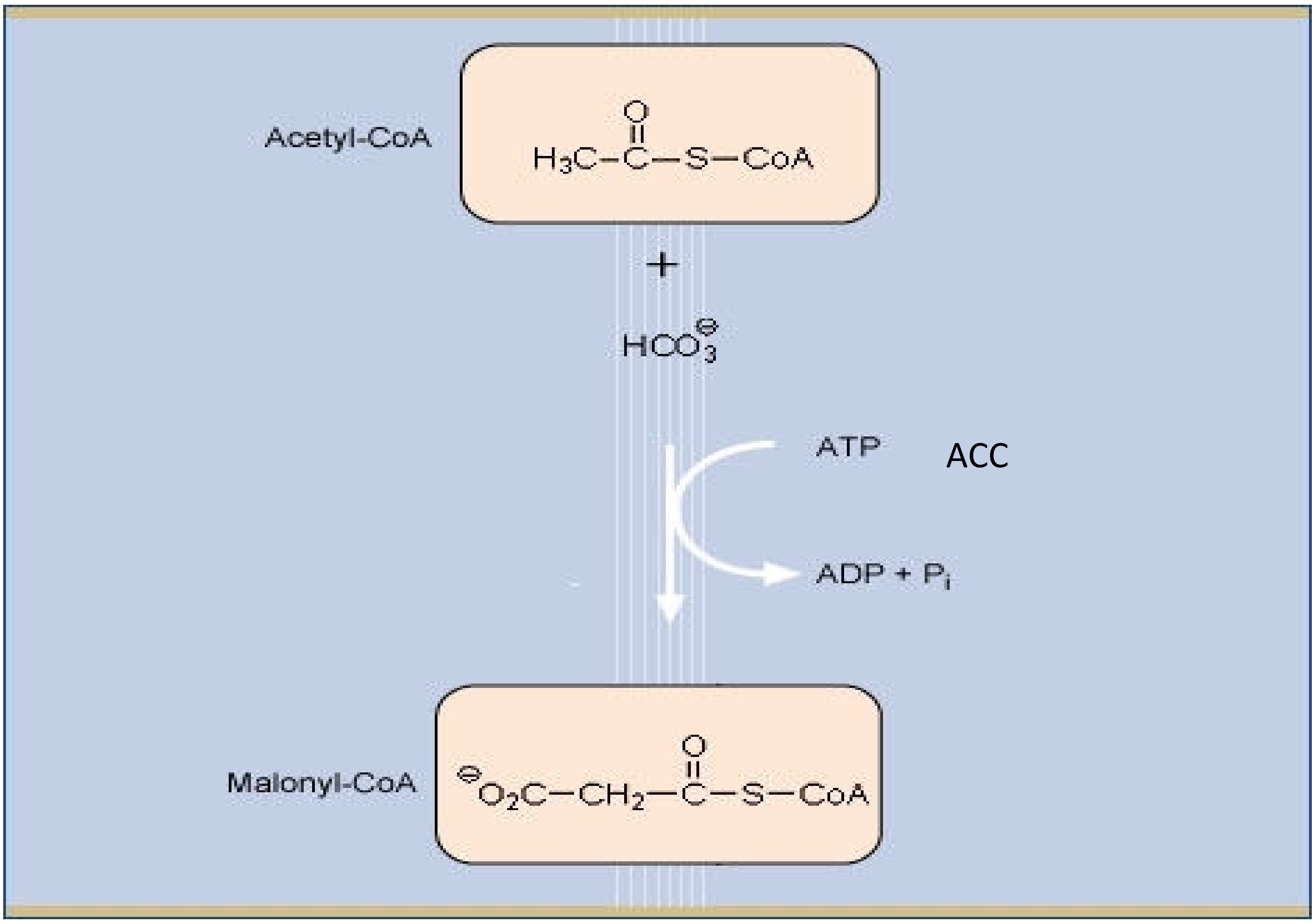


HCO_3^- + ATP + Acetyl-CoA → ADP + P_i + Malonyl-CoA

Acetyl-CoA + HCO_3^- + ATP → Malonyl-CoA +ADP + P_i

ACC-Biotin

Acetyl CoA Carboxylase (ACC)



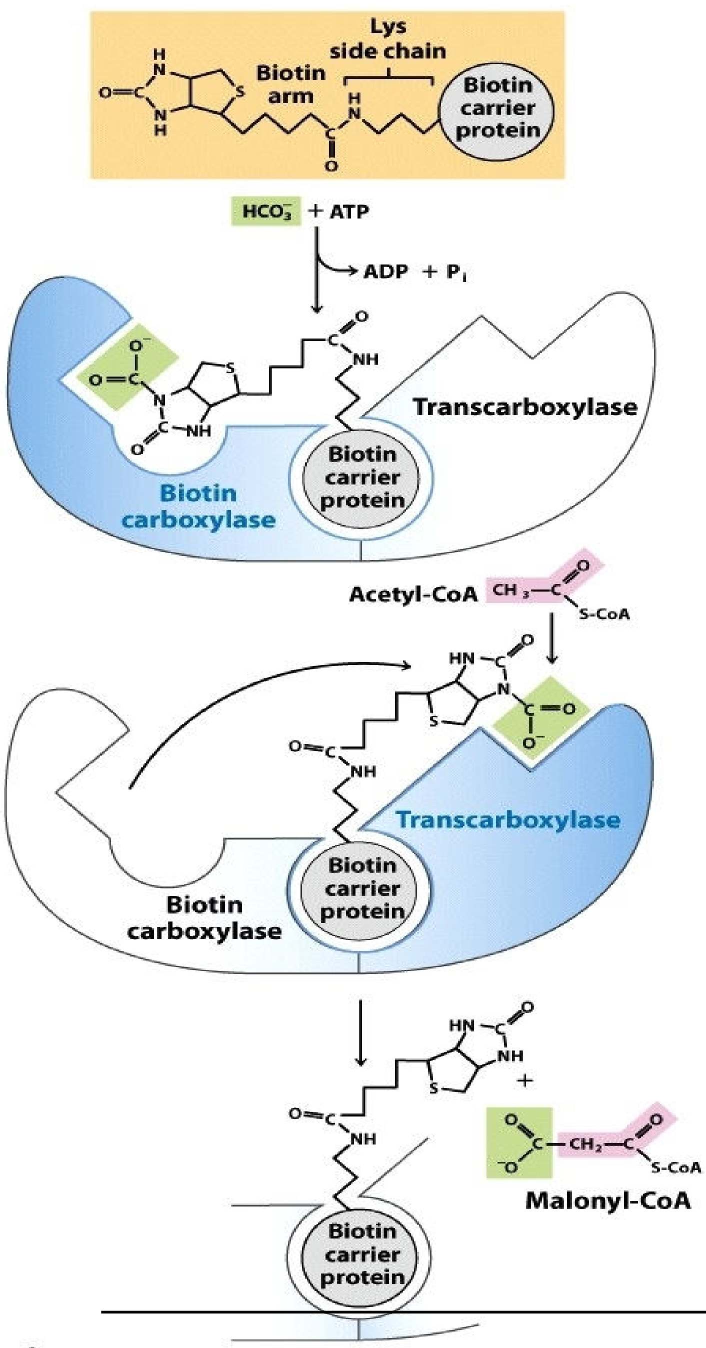


Figure 21-1
Lehninger Principles of Biochemistry, Fifth Edition

Formation of Malonyl-CoA

Acetyl-CoA Carboxylase has three activities:

- Biotin carrier Protein
- Biotin Carboxylase
- Trans Carboxylase

Bicarbonate is Phosphorylated, then picked up by Biotin

Biotin swinging arm transfers CO_2 to acetyl-CoA

Significance Of Formation of Malonyl-SCoA

Significance Of Formation of Malonyl-SCoA

- This Carboxylation reaction is **considered as activation step.**
- As the breaking of the CO_2 bond of Malonyl-SCoA releases lot of energy
- That **"drives" the reaction forward for condensation reaction of FAS complex.**

- High energy bond of Malonyl-CoA is hydrolyzed later
- To liberate energy which is **used up** for **Condensation reaction** of FAS complex.

- **Malonyl-CoA serves as activated donor of Acetyl groups** in FA synthesis.

- **Fatty acid synthesis**, from **Acetyl-CoA and Malonyl-CoA**,
- Occurs by a **series of reactions catalyzed by FAS complex.**

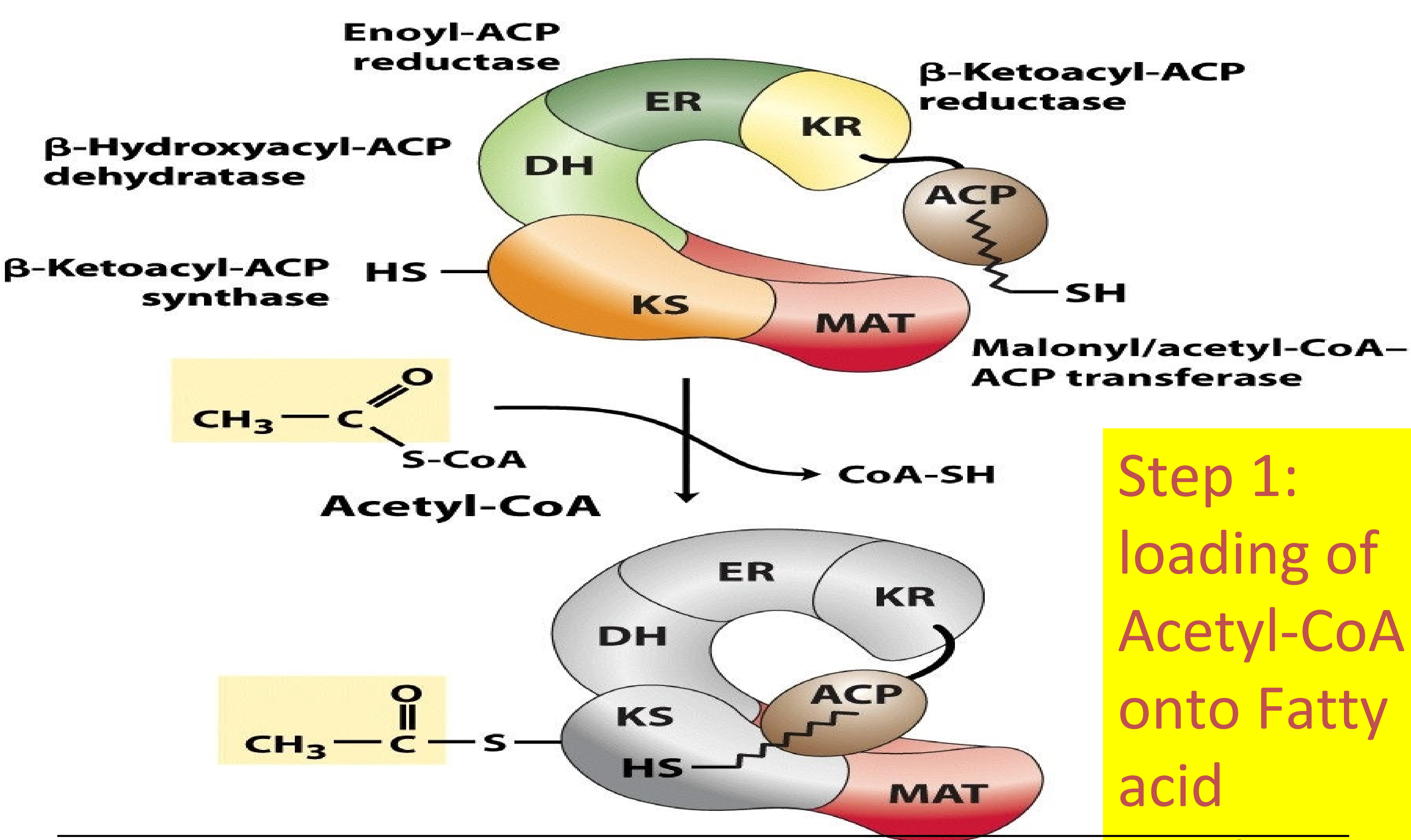
Stage 3

Reactions Of FAS Complex During De Novo Biosynthesis Of a Fatty Acid-Palmitic Acid

Step I-Step III

Loading Of Precursors
Acetyl CoA and Malonyl-CoA
On FAS Complex
By
Acetyl and Malonyl Transacylases

Loading Of Precursor Acetyl CoA



Step 1: loading of Acetyl-CoA onto Fatty acid Synthase

Figure 21-6 part 1
Lehninger Principles of Biochemistry, Fifth Edition

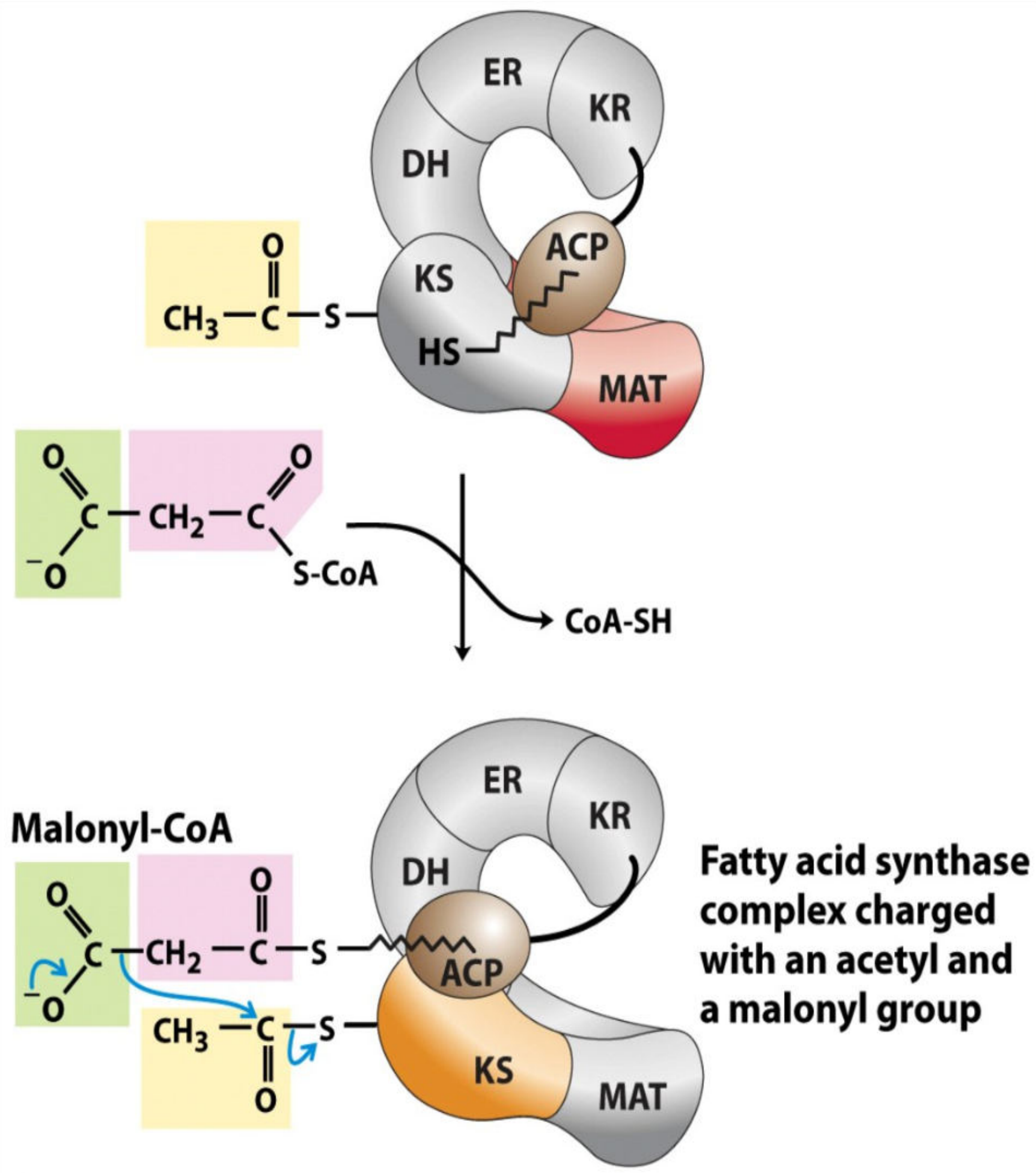


Figure 21-6 part 2
Lehninger Principles of Biochemistry, Fifth Edition

Entry Of Malonyl-CoA

Step 2: loading of Malonyl-CoA onto Fatty acid Synthase

- The **Acetyl-CoA** (2C) primer molecule is **first taken** up by –**SH group of ACP of FAS complex**
- To form **Acetyl-S-ACP** catalyzed by **Acetyl Transacylase.**

- **Acetyl group from ACP** is shifted to Cysteine-SH of enzyme β Keto Acyl Synthase of FAS complex.

- To form **Acetyl-S-Enzyme β Keto Acyl Synthase** in presence of **Acetyl Transacylase.**

- Malonyl-CoA (3Carbon unit) enters and is **taken up by -SH of ACP of FAS complex**

- To form **Malonyl-S ACP** catalyzed by **Malonyl Transacylase.**

Step IV
Condensation Reaction
Catalyzed By
Beta Keto Acyl Synthase
To Generate
Keto group
At Beta Carbon Atom

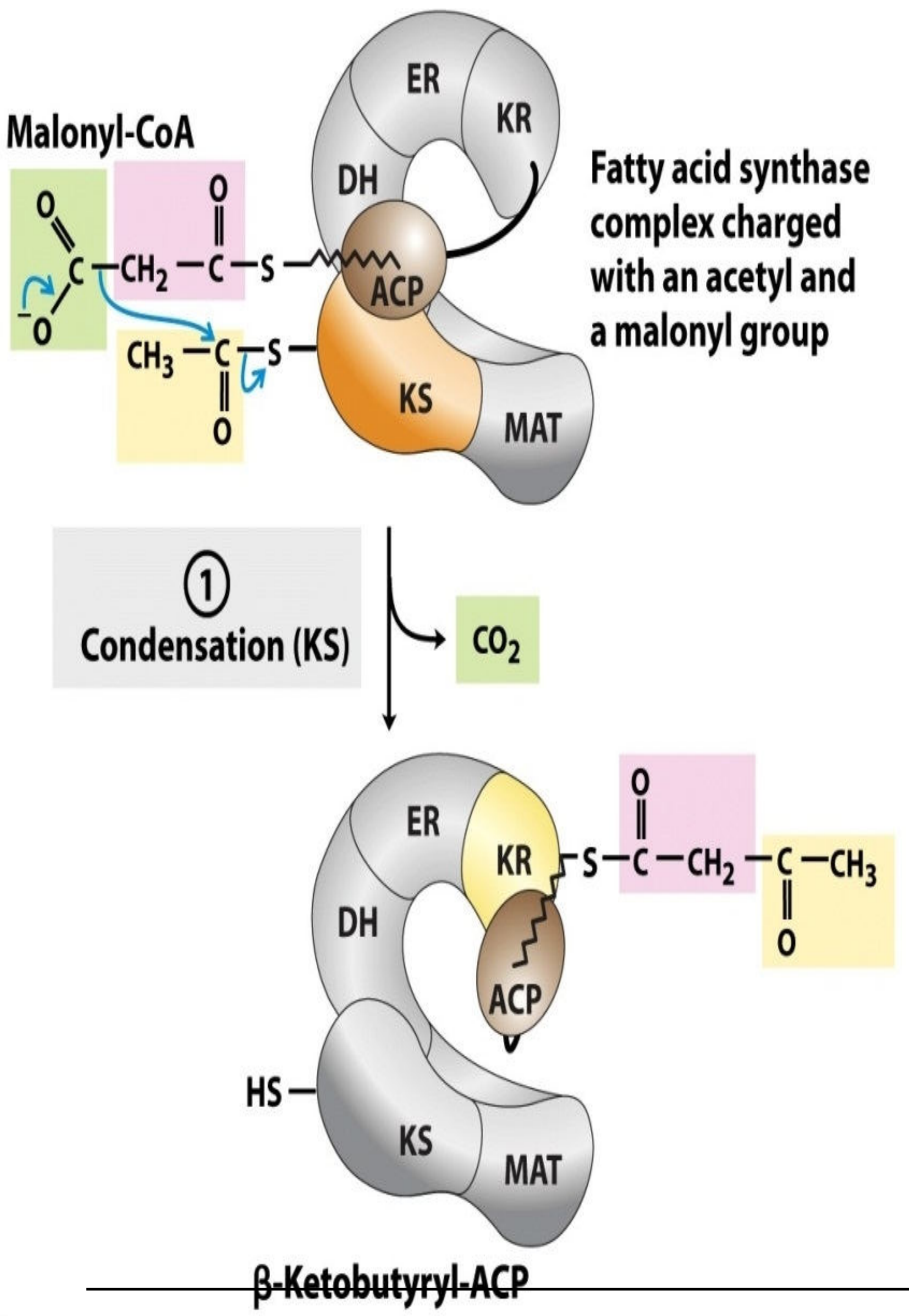


Figure 21-6 part 3
Lehninger Principles of Biochemistry, Fifth Edition

Step 2: Condensation

- Reaction of Malonyl group with Acetyl group to form Acetoacetyl- ACP
- Loss of CO_2 and energy from decarboxylation of Malonyl-CoA.

- Malonyl Group is **decarboxylated releasing** CO_2 and **high energy**

- **W**hich is **used for bond building and condensation reaction**.

- During condensation reaction there is linking of 2C units of Acetyl and 2C units of decarboxylated Malonyl carbon units

- To form a 4 C **Beta Keto Butyryl ACP/β Keto Acyl ACP**.

Step V
Reduction Reaction
By
Keto Acyl Reductase
To
Generate Beta Hydroxyl group

Step 3: Reduction of beta Keto group to form beta Hydroxyl group

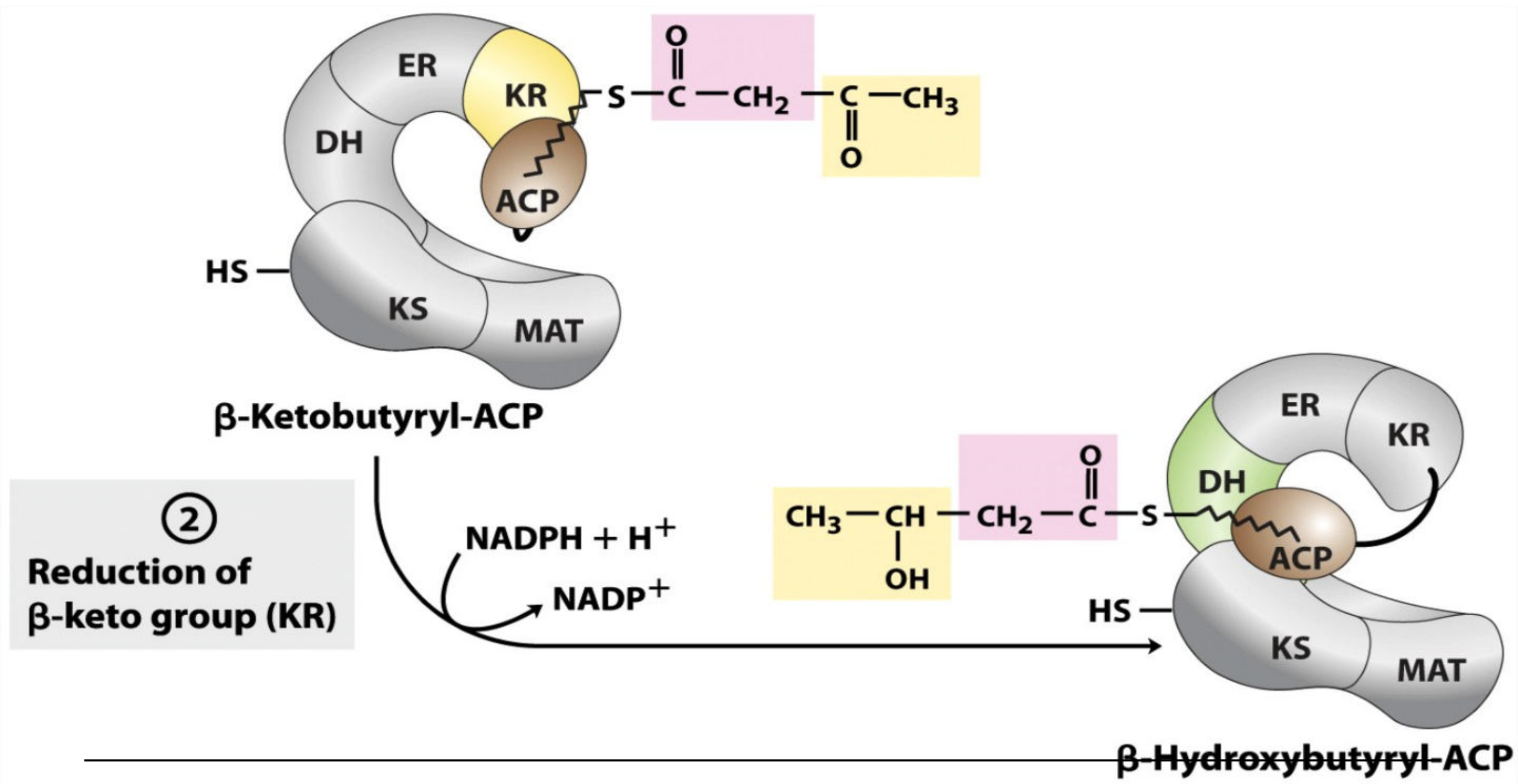


Figure 21-6 part 4
Lehninger Principles of Biochemistry, Fifth Edition

Reduction Of β Keto Acyl- ACP

- **β Keto Acyl- ACP** is reduced to **β Hydroxy Acyl- ACP**
- In presence of reducing equivalents **NADPH+H+ and Enzyme β Keto Acyl Reductase.**

Step VI
Dehydration Reaction
By
Dehydratase
To
Develop Double Bond

Step 4: Dehydration Reaction

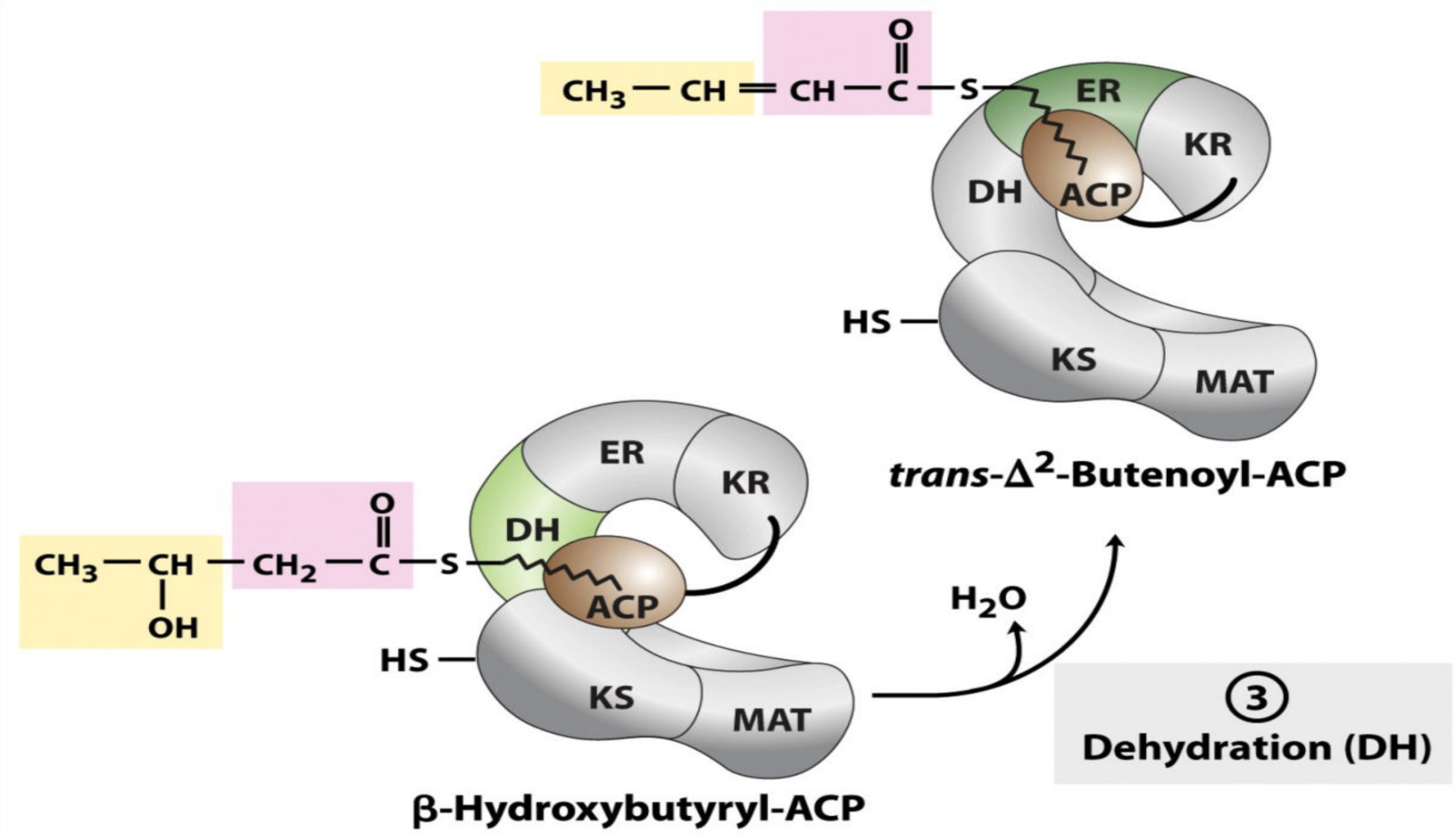


Figure 21-6 part 5
Lehninger Principles of Biochemistry, Fifth Edition

- **β Hydroxy Acyl- ACP** is dehydrated to **Enoyl CoA/α –β Unsaturated Acyl ACP** by the catalytic action of **Dehydratase.**

Step VII
Reduction Reaction
By
Enoyl-CoA Reductase
To Generate
Saturated Bond

Step 5: Reduction of double bond to Single bond

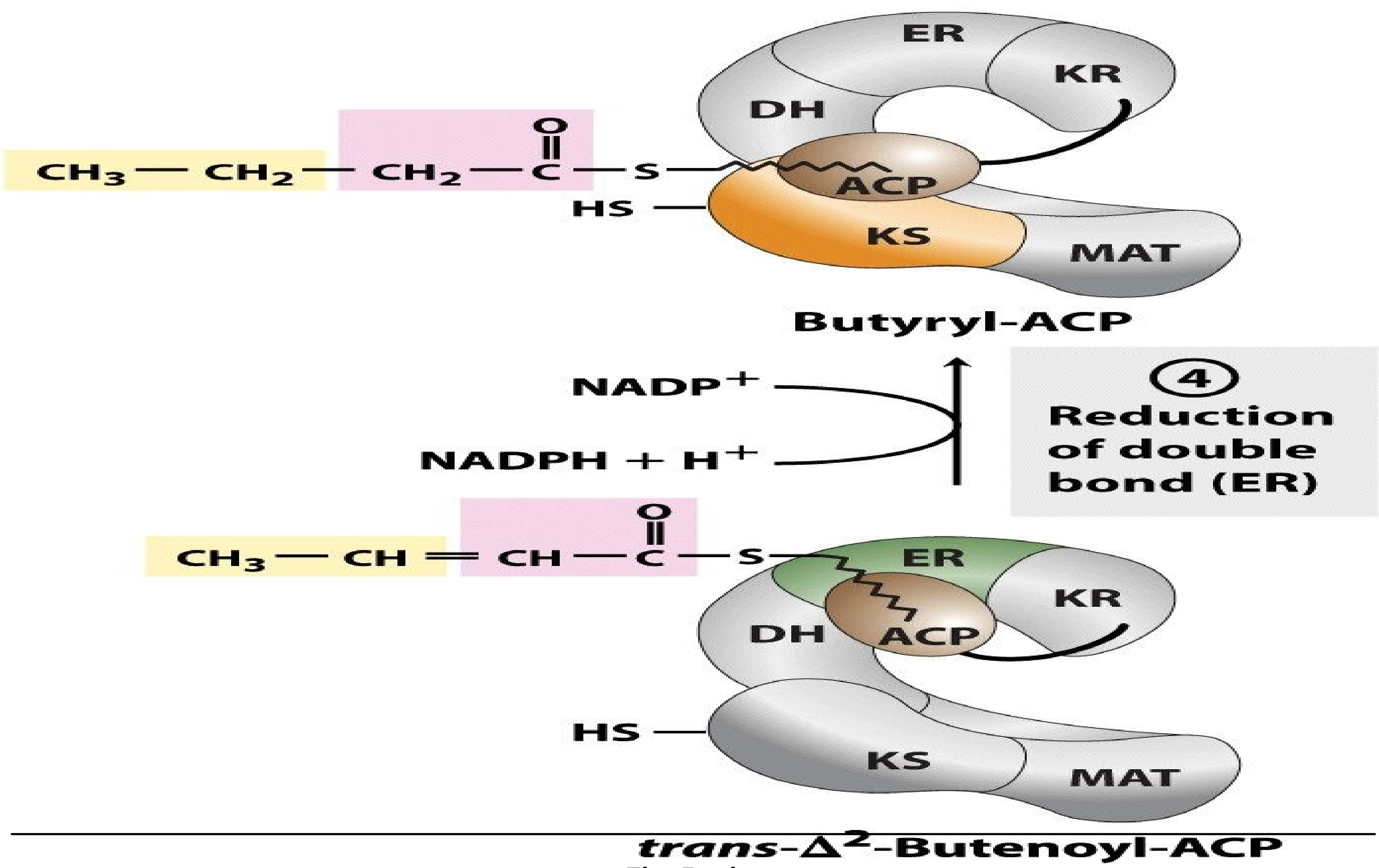
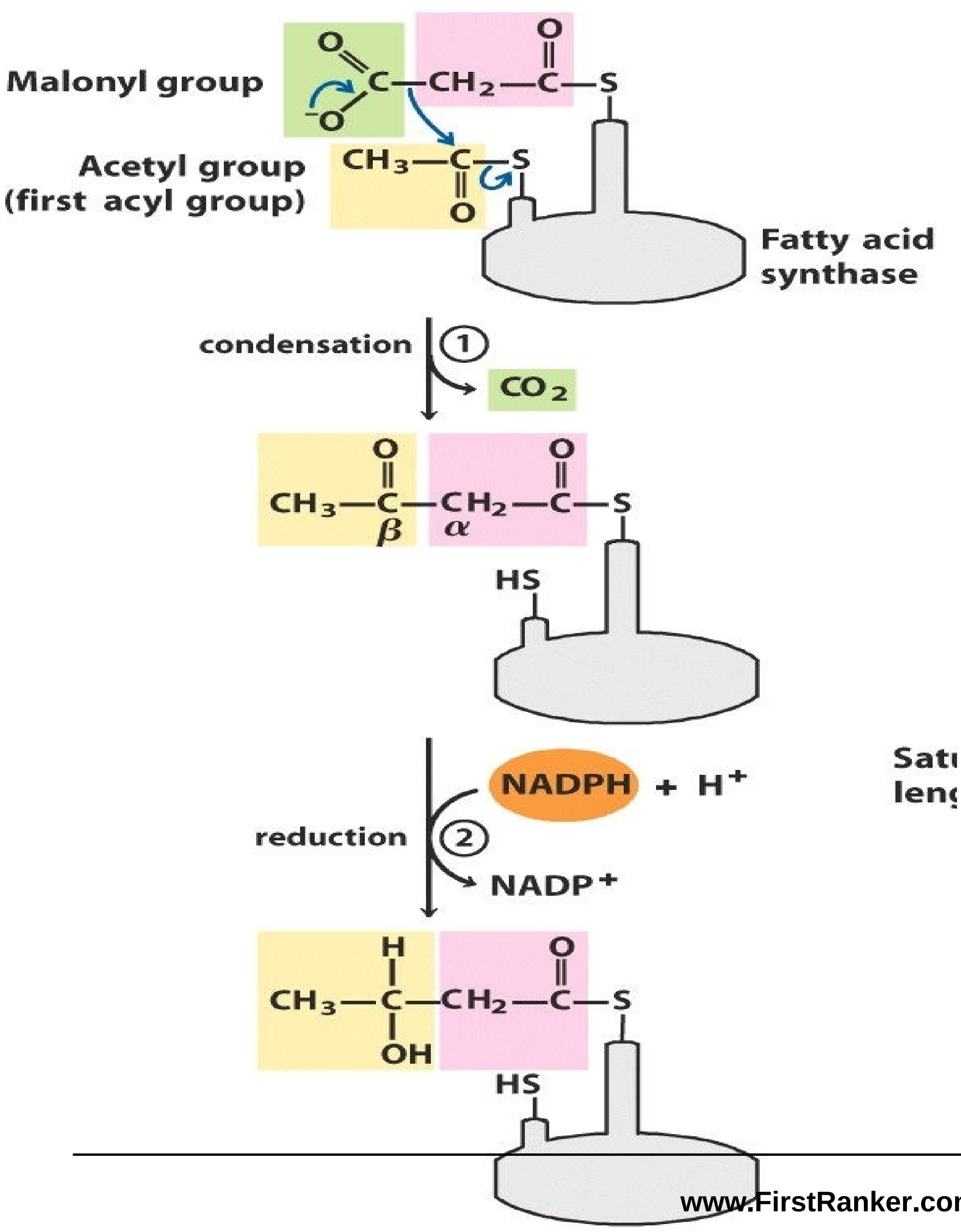


Figure 21-6 part 6
Lehninger Principles of Biochemistry, Fifth Edition

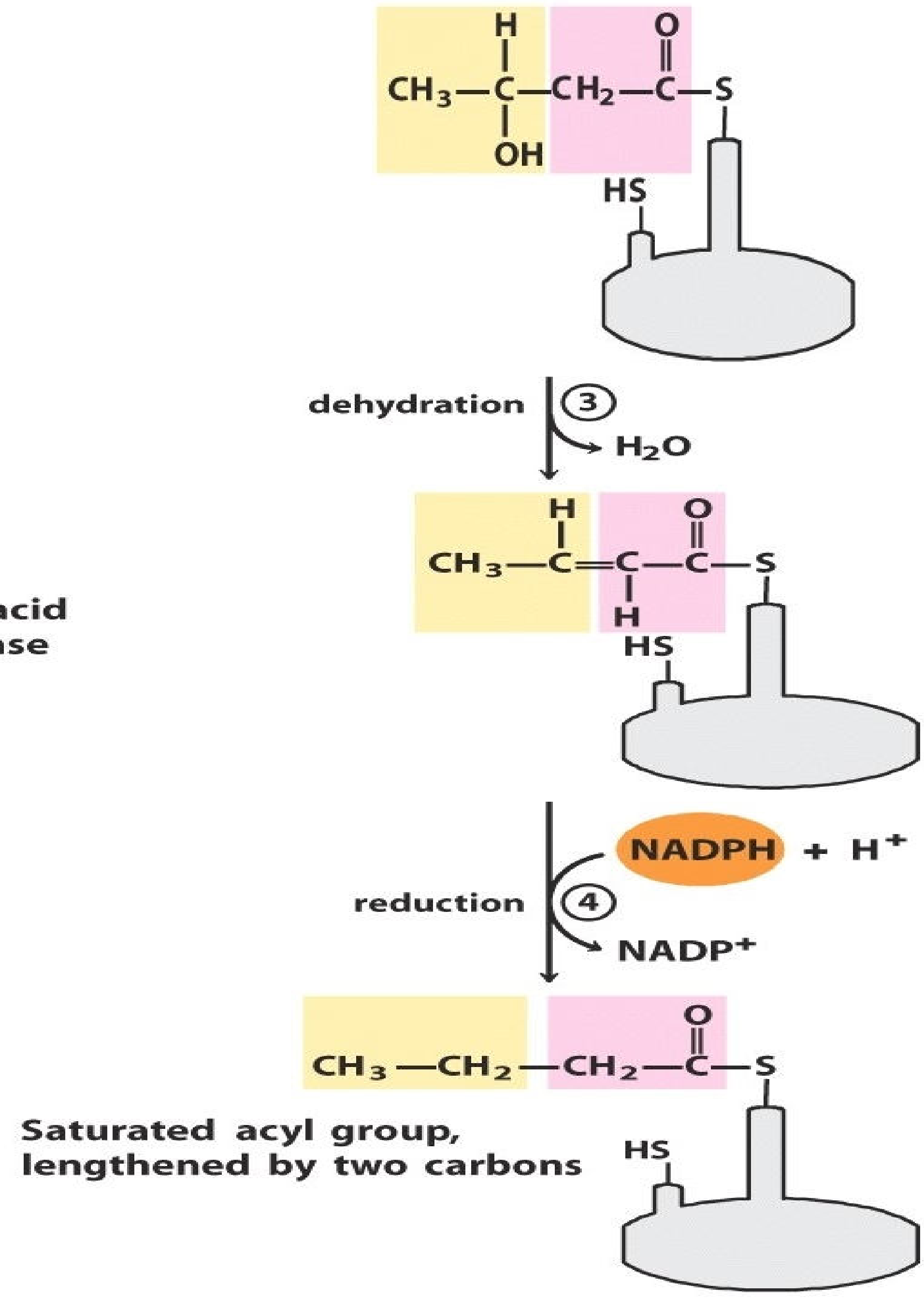
- α – β Unsaturated Acyl ACP is reduced to Butyryl –S-ACP
- By **NADPH+ H+ and enzyme Enoyl Reductase**.



Overview of
Assembly Stage

4 steps:

- Condensation
- Reduction



Overview of Assembly Stage

- Dehydration
- Reduction

Step VIII
Translocation Of Butyryl-S CoA to SH group of Condensing Enzyme Beta Keto Acyl Synthase

Transfer of Butyryl Chain to SH group of Beta Keto Acyl Synthase

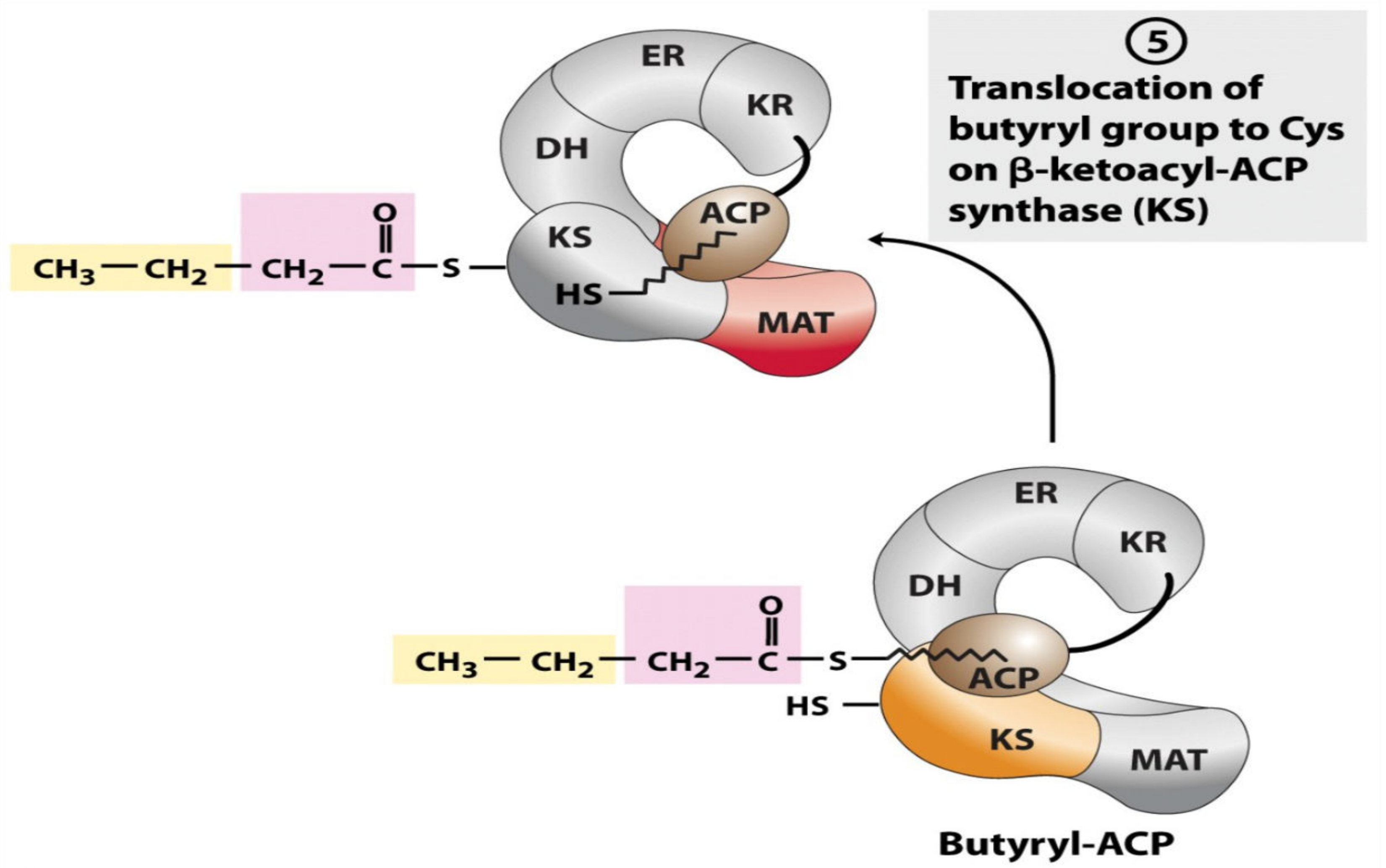


Figure 21-6 part 7
Lehninger Principles of Biochemistry, Fifth Edition
© 2008 W. H. Freeman and Company

Elongation and Growing Of Fatty Acid Chain

To Elongate the Fatty Acid Chain To 16 Carbon Palmitate

There Should Be Entry

Of

6 More Molecules of Malonyl CoA

By Six Time Repetitions of Steps III-VIII

1 Malonyl-CoA entry each Time

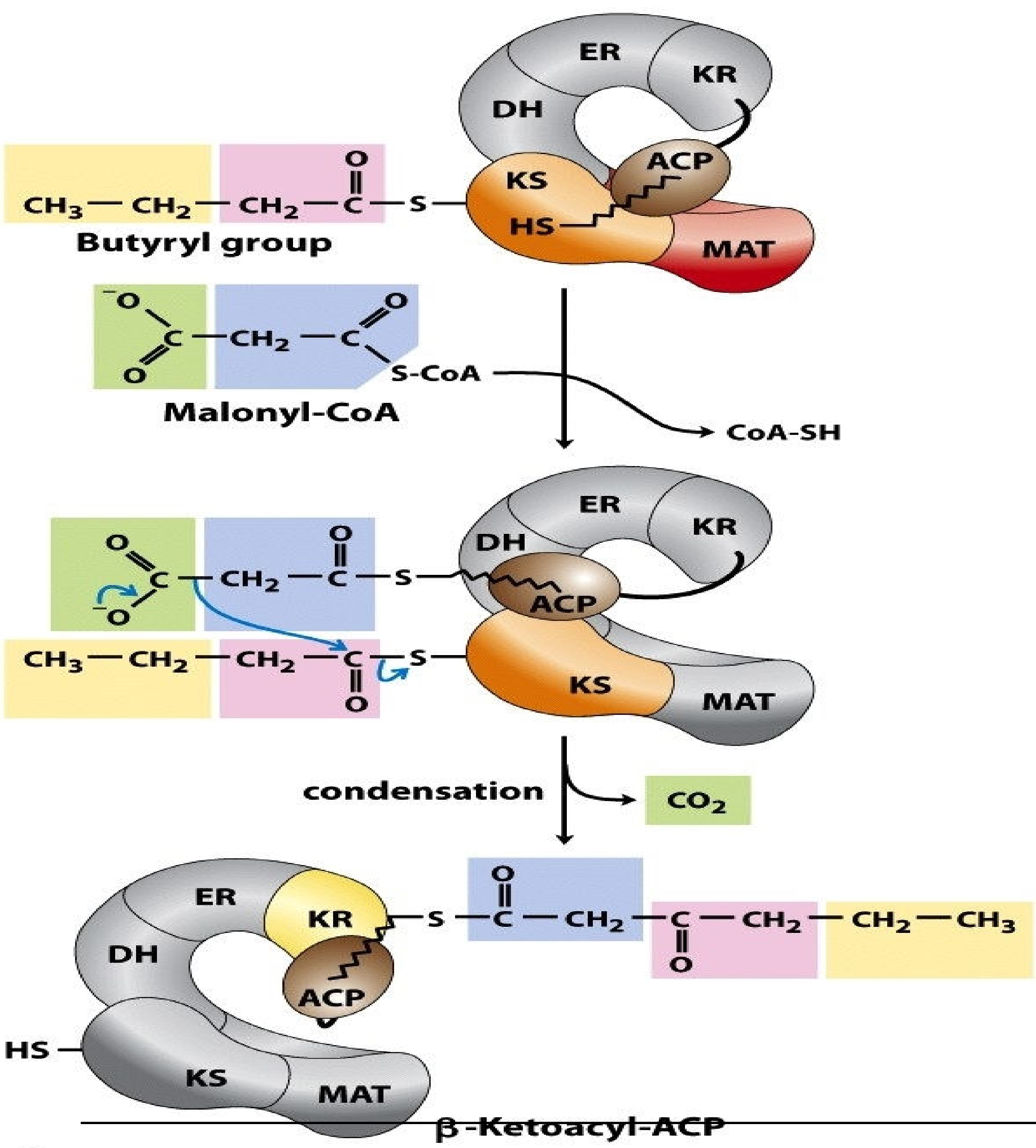
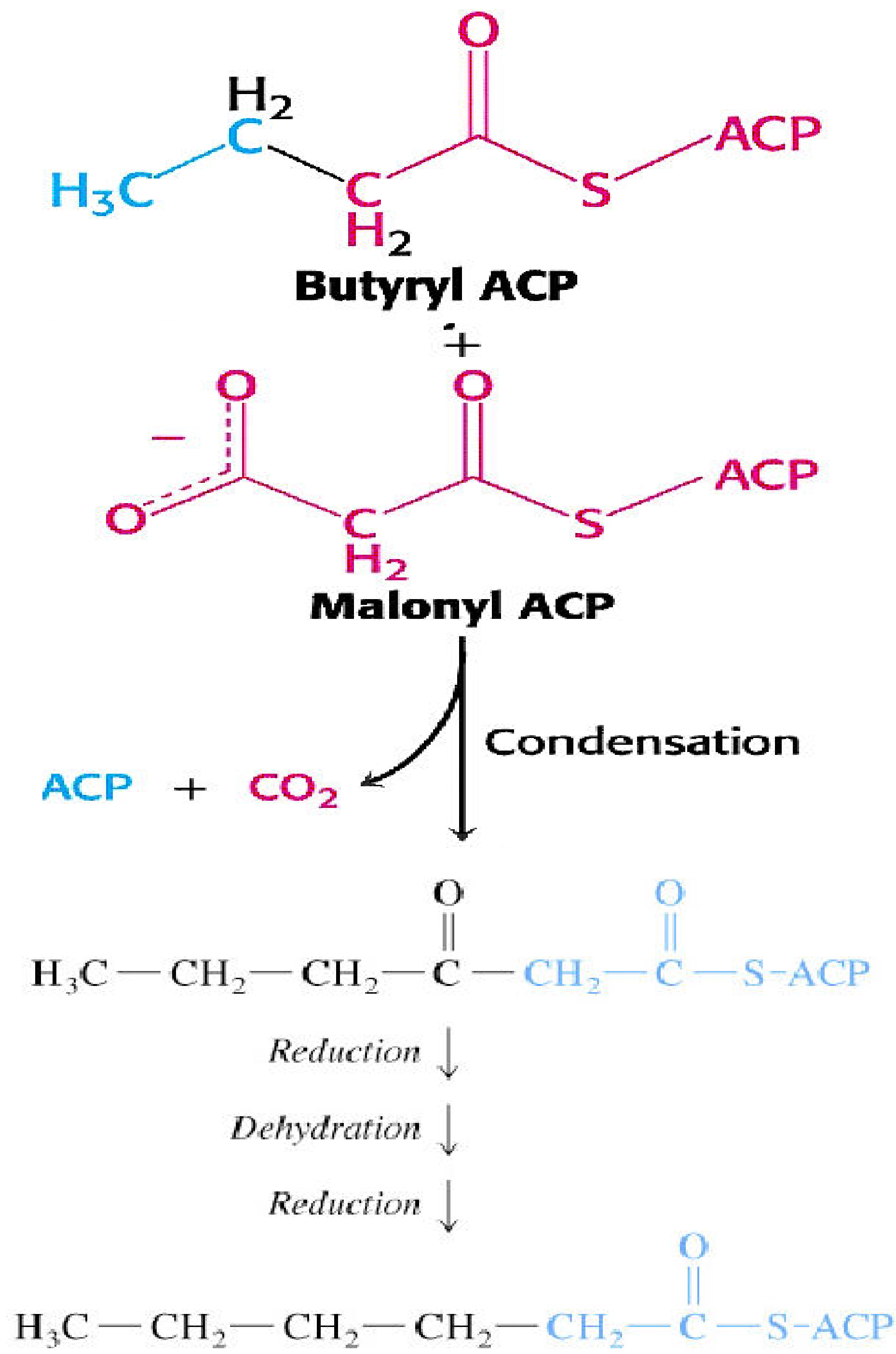


Figure 21-7
Lehninger Principles of Biochemistry, Fifth Edition

Next cycle begins

✓ Another Malonyl group is linked to ACP

Repetitions Of 6 More Cycles With 5 Steps



Fatty Acyl Synthase

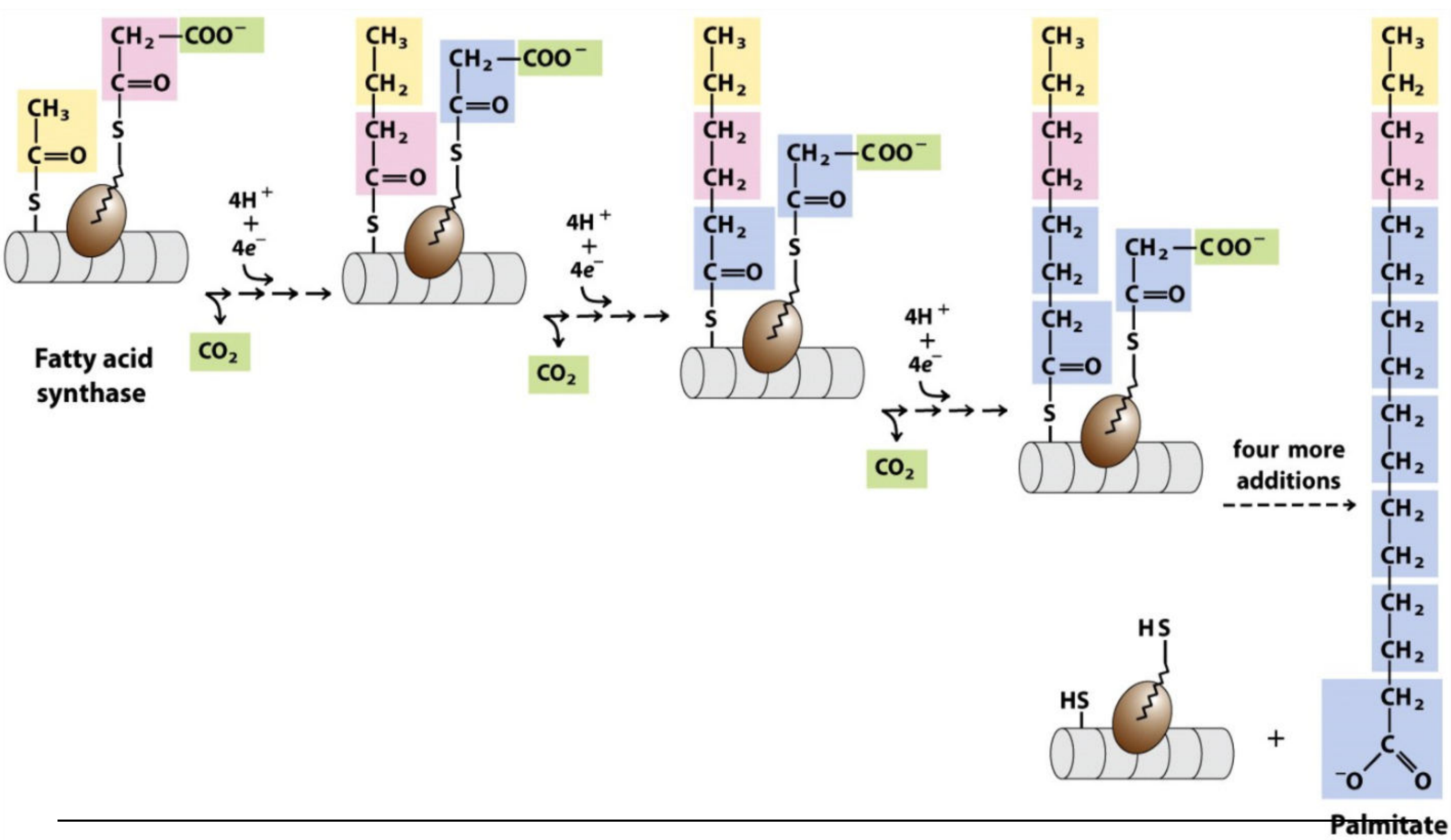
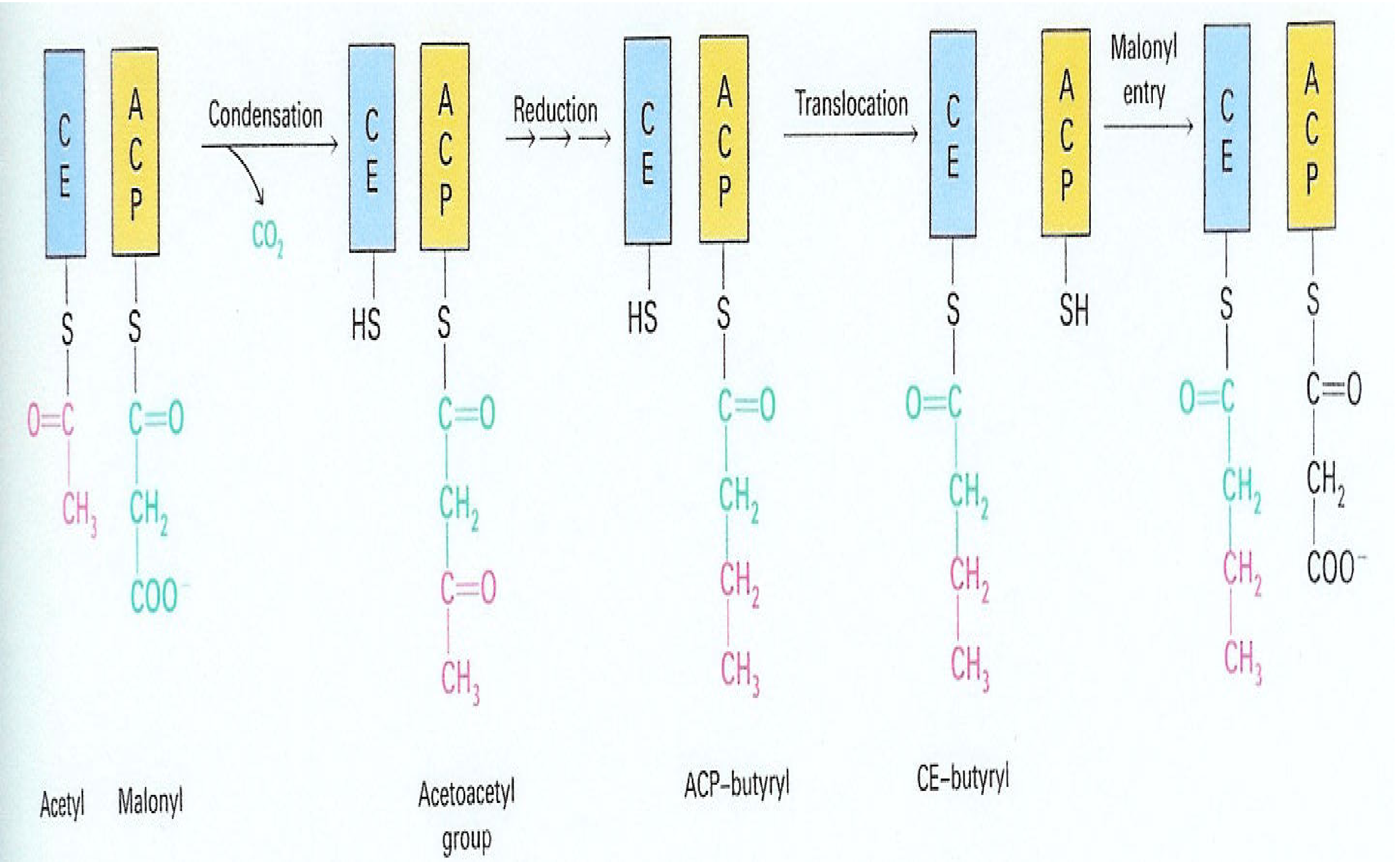


Figure 21-4
Lehninger Principles of Biochemistry, Fifth Edition

The Steps in the De Novo biosynthesis of fatty acid



- **Initiation To Form An Acyl Chain**
    - I. **Loading of Precursor** –Acetyl-CoA at SH-ACP
    - II. **Translocation of Acetyl –S-ACP** to SH-Condensing Enzyme (SH-CE)
    - III. **Entry of Malonyl-CoA and Loading of Malonyl to SH-ACP**
    - IV. **Condensation of the Acetyl and Malonyl with decarboxylation**
    - V. **Reduction Reaction** to transform beta Keto group to Hydroxyl
    - VI. **Dehydration Reaction** to transform Hydroxyl group to Enoyl
    - VII. **Reduction Reaction** to transform Enoyl
    - VIII. **Translocation of Butyryl** From S-ACP to SH-CE

- **Elongation and Growing of Acyl Chain**
    - –By Six Time Repetitions of Steps III-VIII
    - –Entry Of 6 Malonyl-CoA's at SH-ACP
    - –1 Malonyl-CoA in each cycle to ACP-SH

- **Cleavage of Fatty acid/ Palmitate**
    - –By Thioesterase activity to release Palmitate and FAS

- Following **transfer** of growing fatty acid from Phosphopantetheine to the Condensing Enzyme's Cysteine sulfhydryl.
- **Cycle begins again**, with another Malonyl-CoA.

- Elongation of Fatty Acyl chain occurs by addition of Malonyl-CoA after every cycle.
- Every time a new Malonyl – CoA enters and taken up by SH-ACP.

- **There are total 7 cycles** to utilize
- **1 Acetyl-CoA** and **7 molecules of Malonyl-CoA** and
- Elongate the Fatty Acid Chain to 16 Carbon Palmitate.

Remember

- At **Each turn one Molecule of Malonyl CoA enters**
- **Accepted by ACP-SH** to form **Malonyl – SACP.**
- Then repetitions of **Condensation ,Reduction , Dehydration and Reduction Reactions** takes place.

- **Decarboxylation of Malonyl-CoA and**
- **Reducing power of NADPH+H$^+$ drive fatty chain growth.**

- **Butyryl group** (C4) is **shifted to SH of Cysteine of β Keto Acyl Synthase.**
- **SH of ACP** is free for accepting second molecule of Malonyl CoA to form Malonyl-S-ACP.

- Steps of Condensation ,Reduction, Dehydration and Reduction repeats.

- **Aim of these steps is to convert a C=O group to CH2 group at β carbon of growing Acyl chain.**

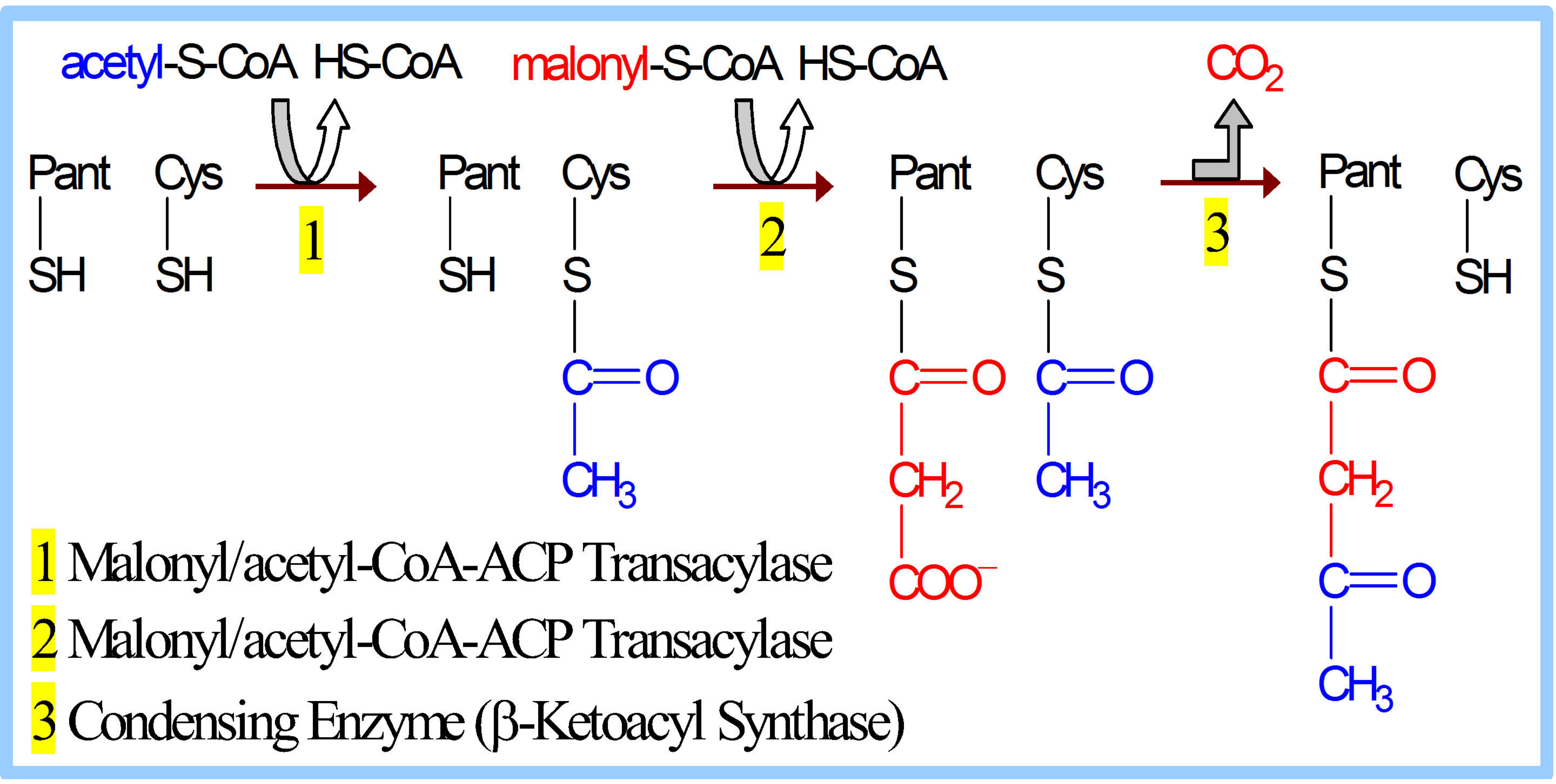
- After completion of total 7 cycles
- There is Palmitate synthesized and is carried by **S-ACP of FAS complex**(Palmitoyl-S-ACP)

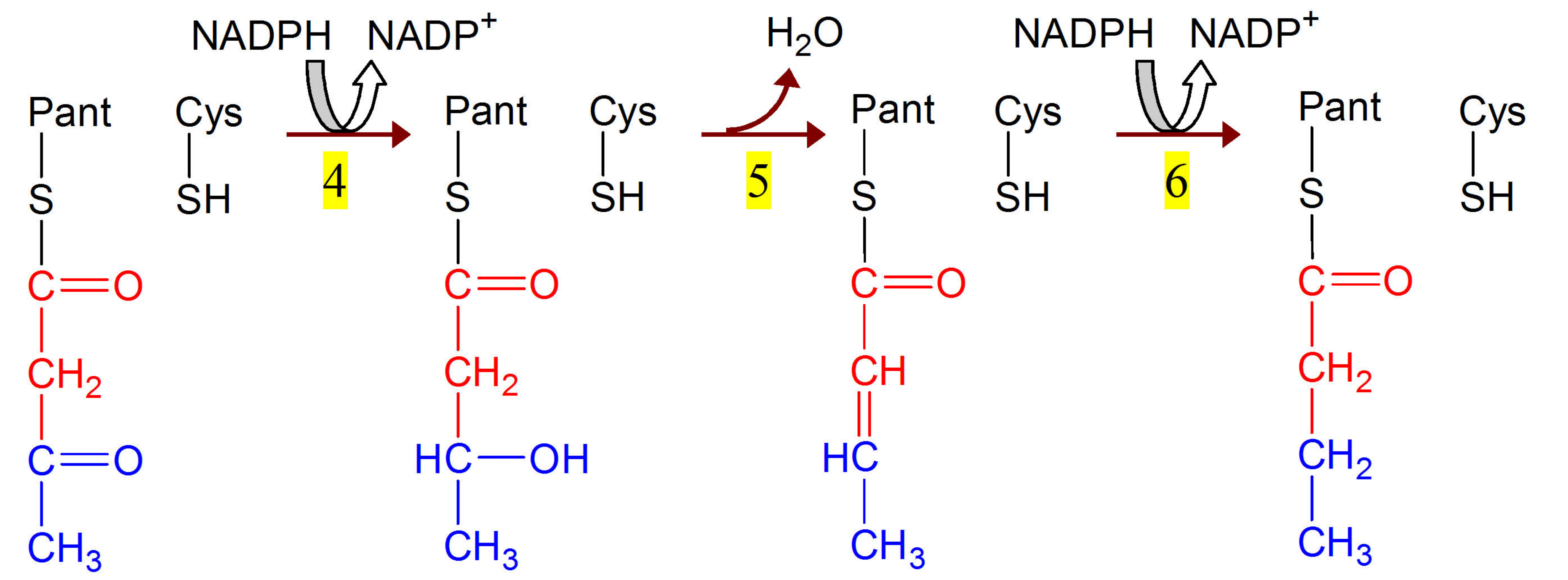
Cleavage Of Completely Biosynthesized Palmitate From ACP of FAS Complex By Catalytic Activity Of Thioesterase To Release Free Palmitate and FAS Complex

- Cleavage enzyme **Thioesterase** **cleaves Thioester linkage** and
- Releases **free Palmitic acid carried by S-ACP of FAS complex.**

- Since **FAS complex is a dimeric unit** having **two functional units.**

- During its operation at a time **two molecules of Palmitic acid are biosynthesized and released**.

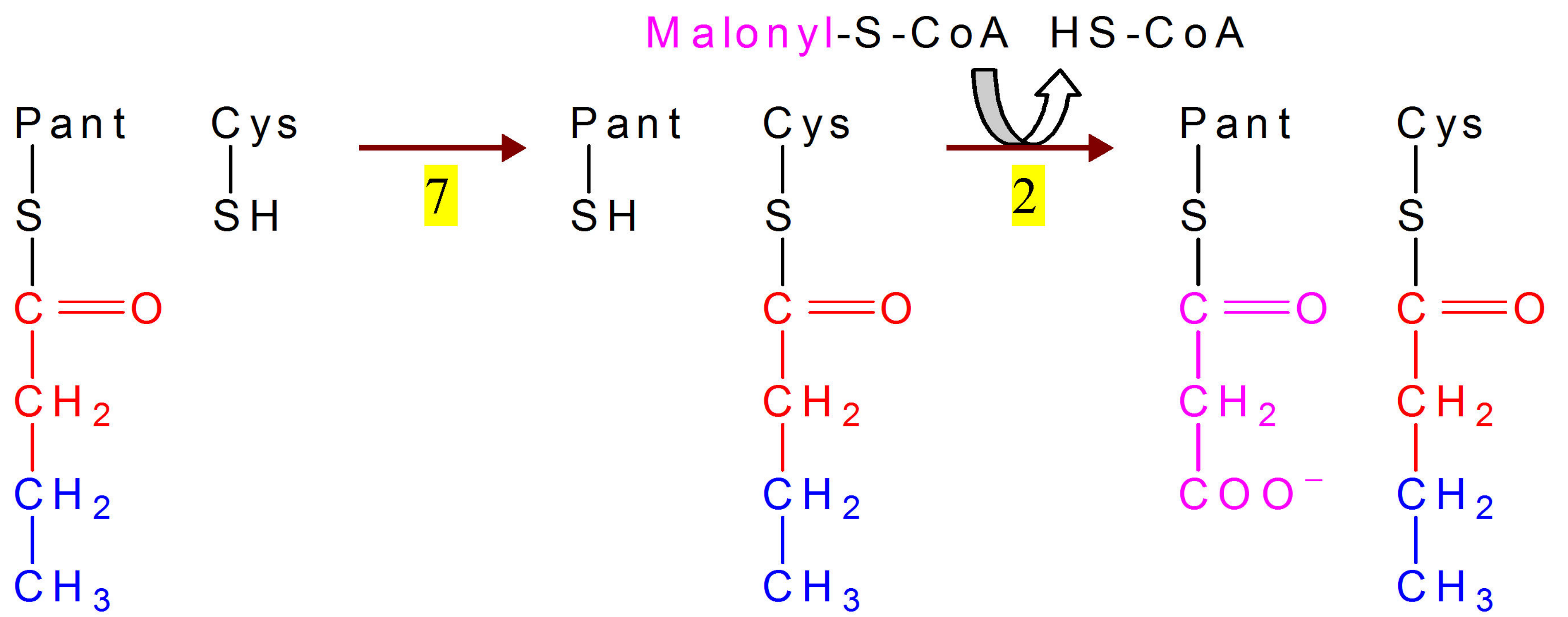




4 β-Ketoacyl-ACP Reductase

5 β-Hydroxyacyl-ACP Dehydratase

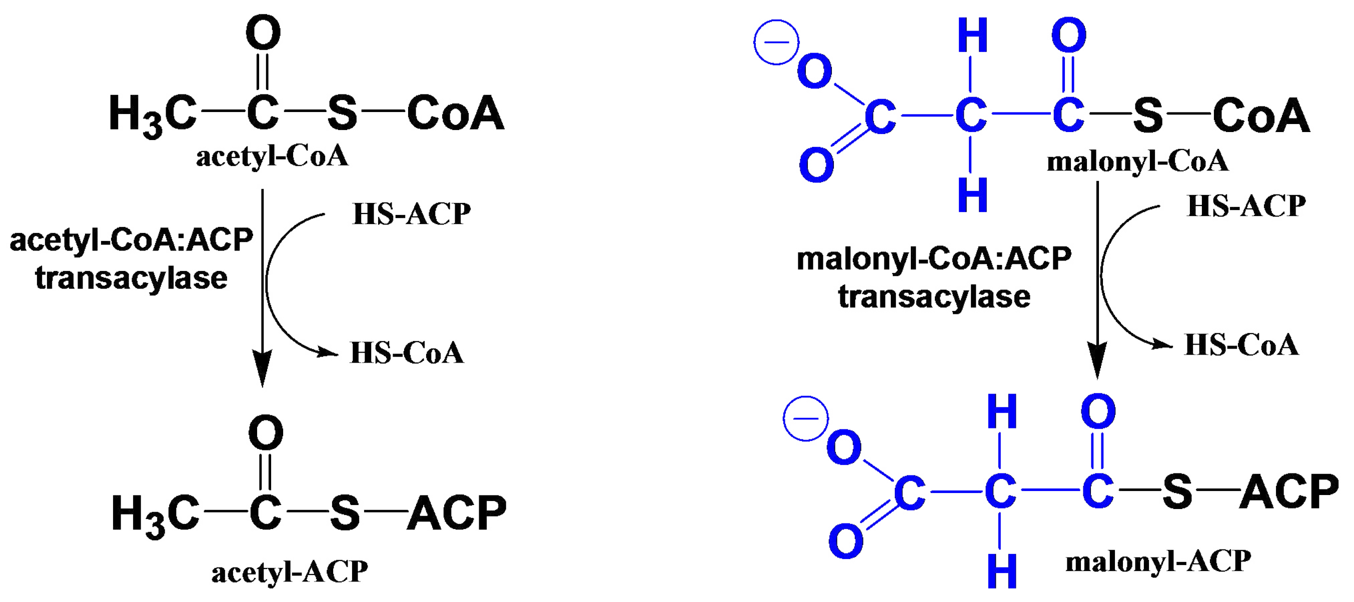
6 Enoyl-ACP Reductase

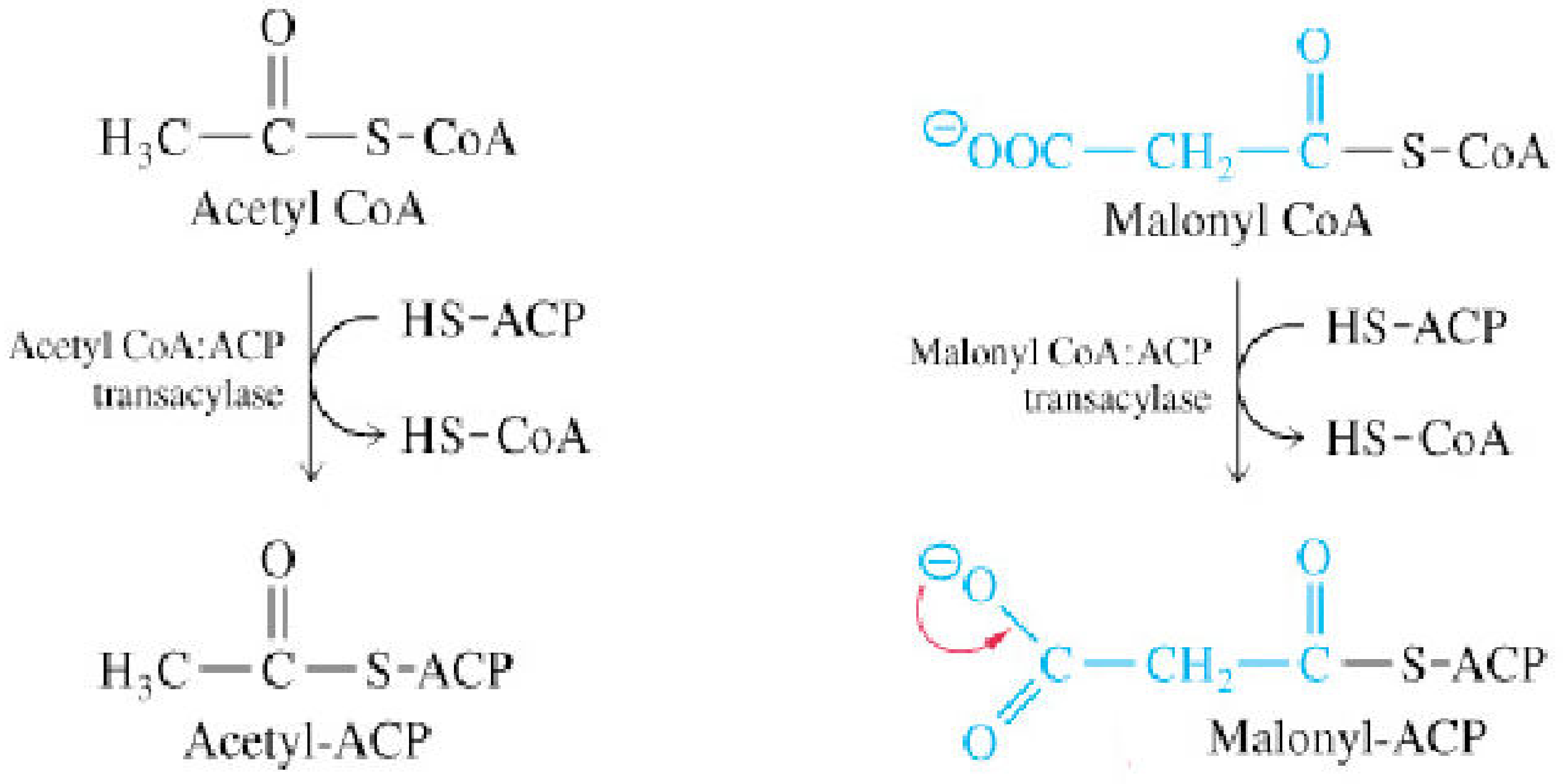


7 Condensing Enzyme

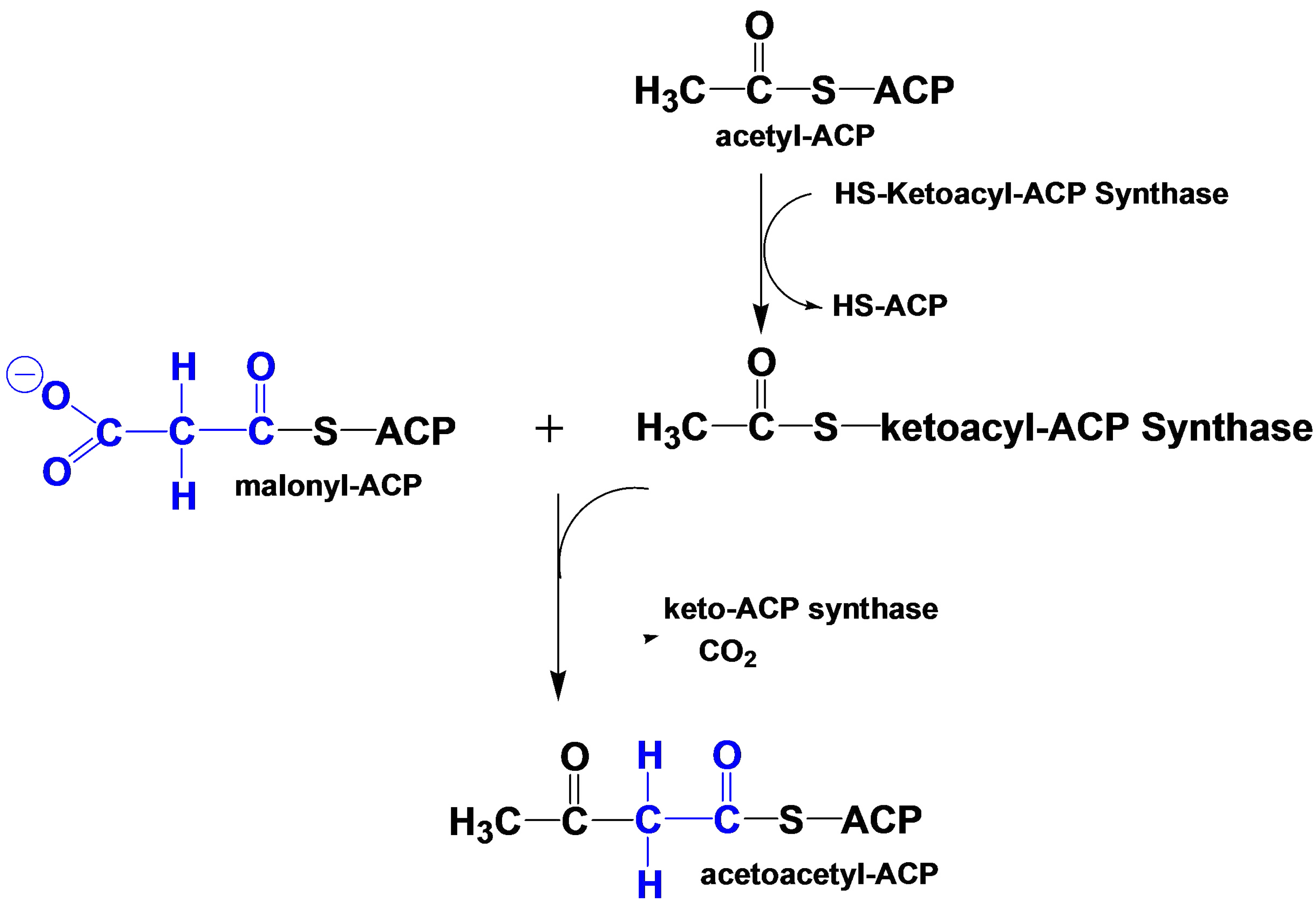
2 Malonyl/acetyl-CoA-ACP Transacylase (repeat).

Step 1: Loading Reactions

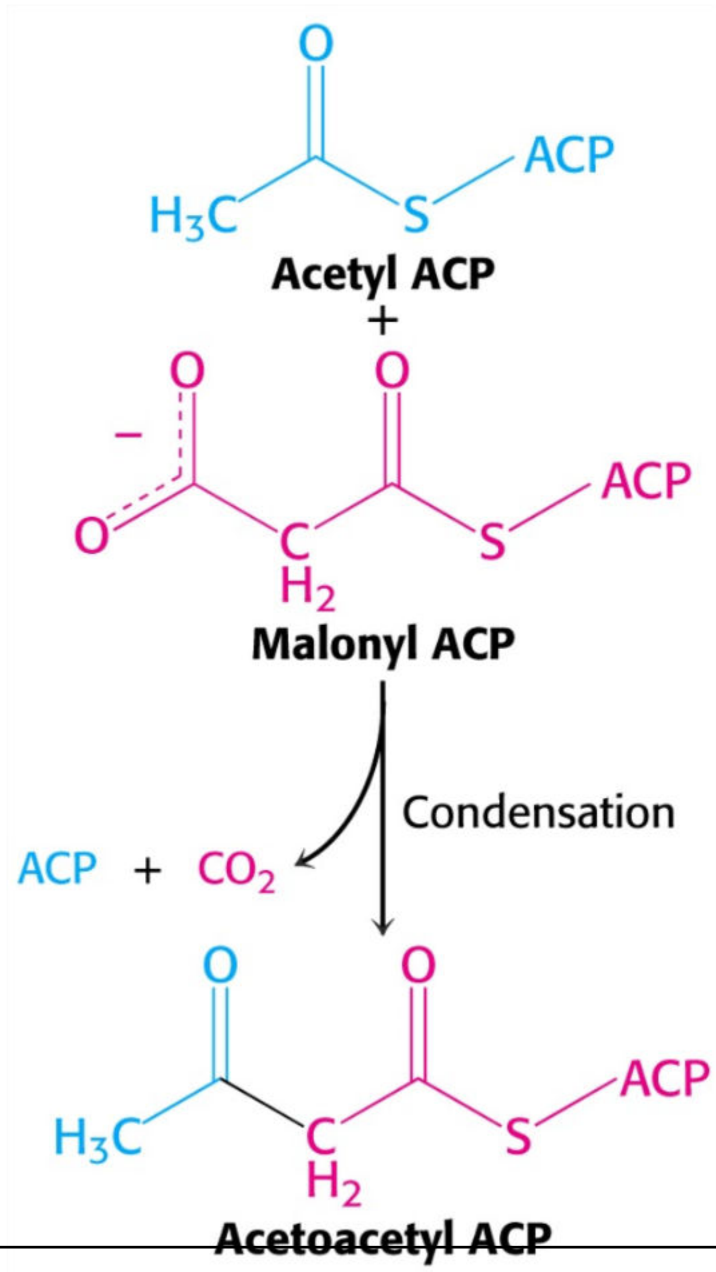




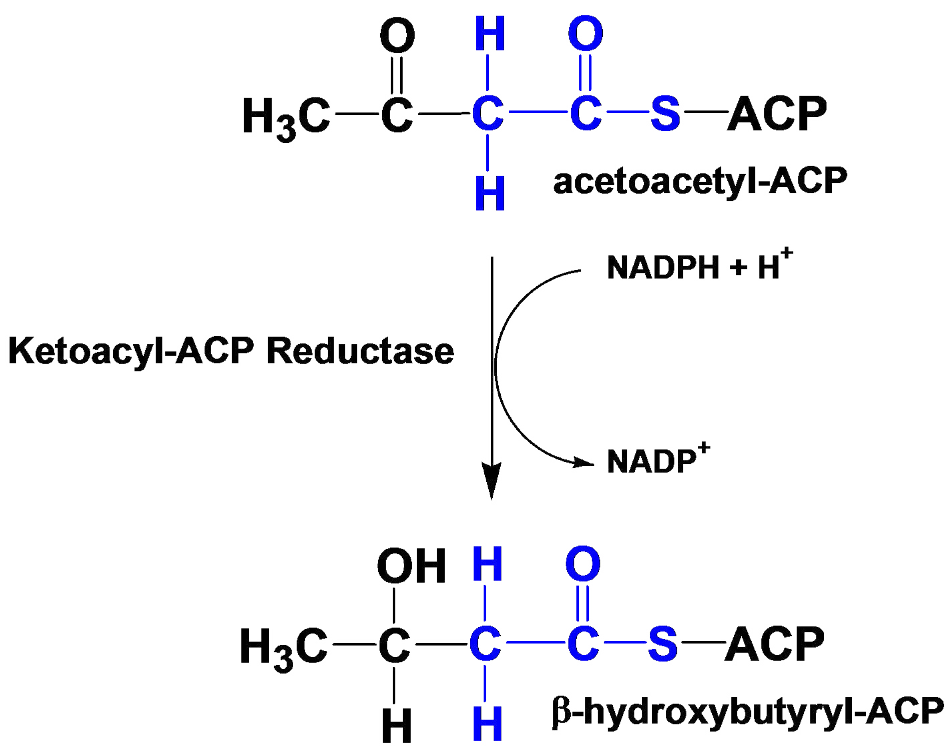
Step 2: Condensation Rxn



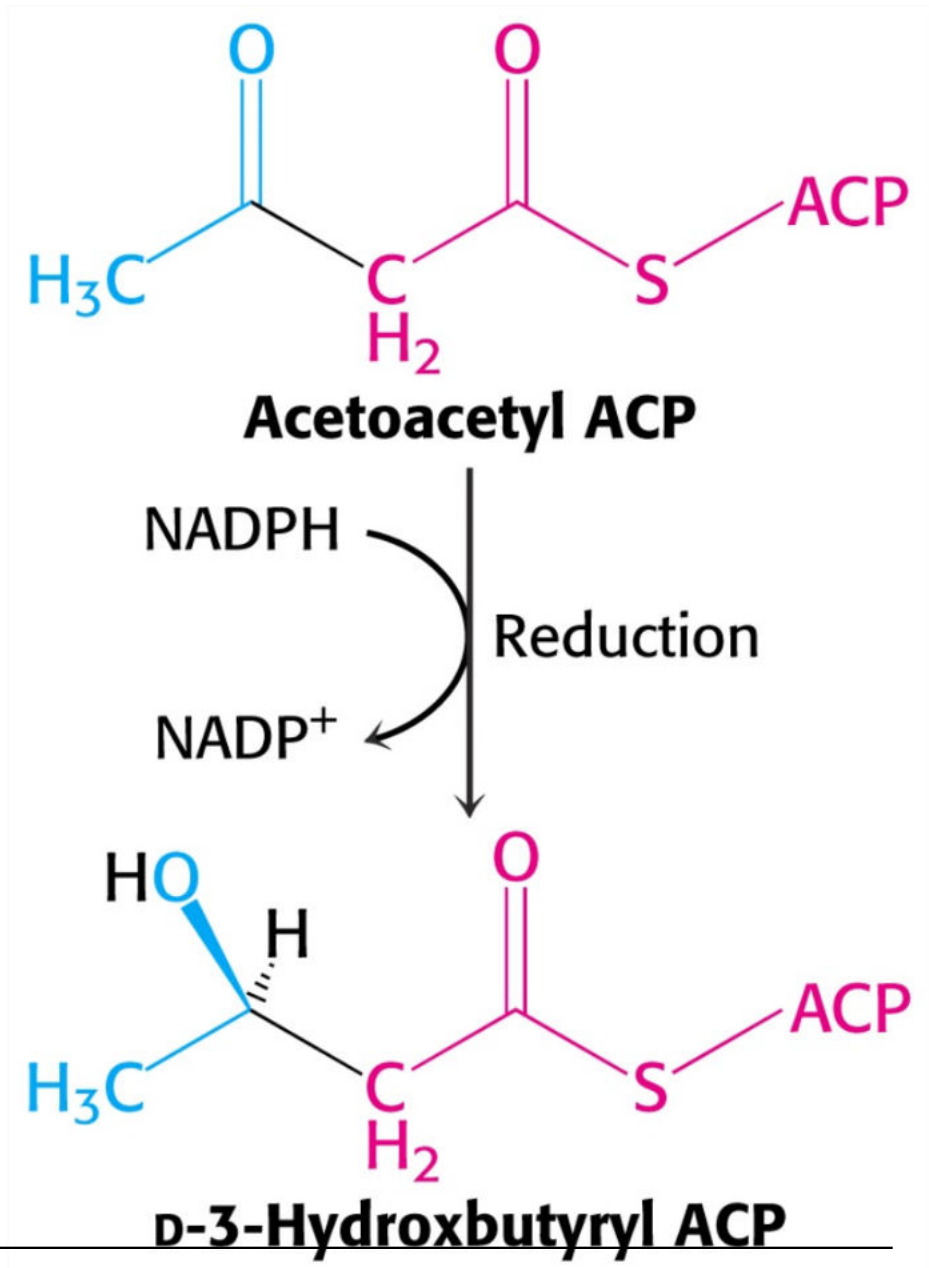
Condensation reaction



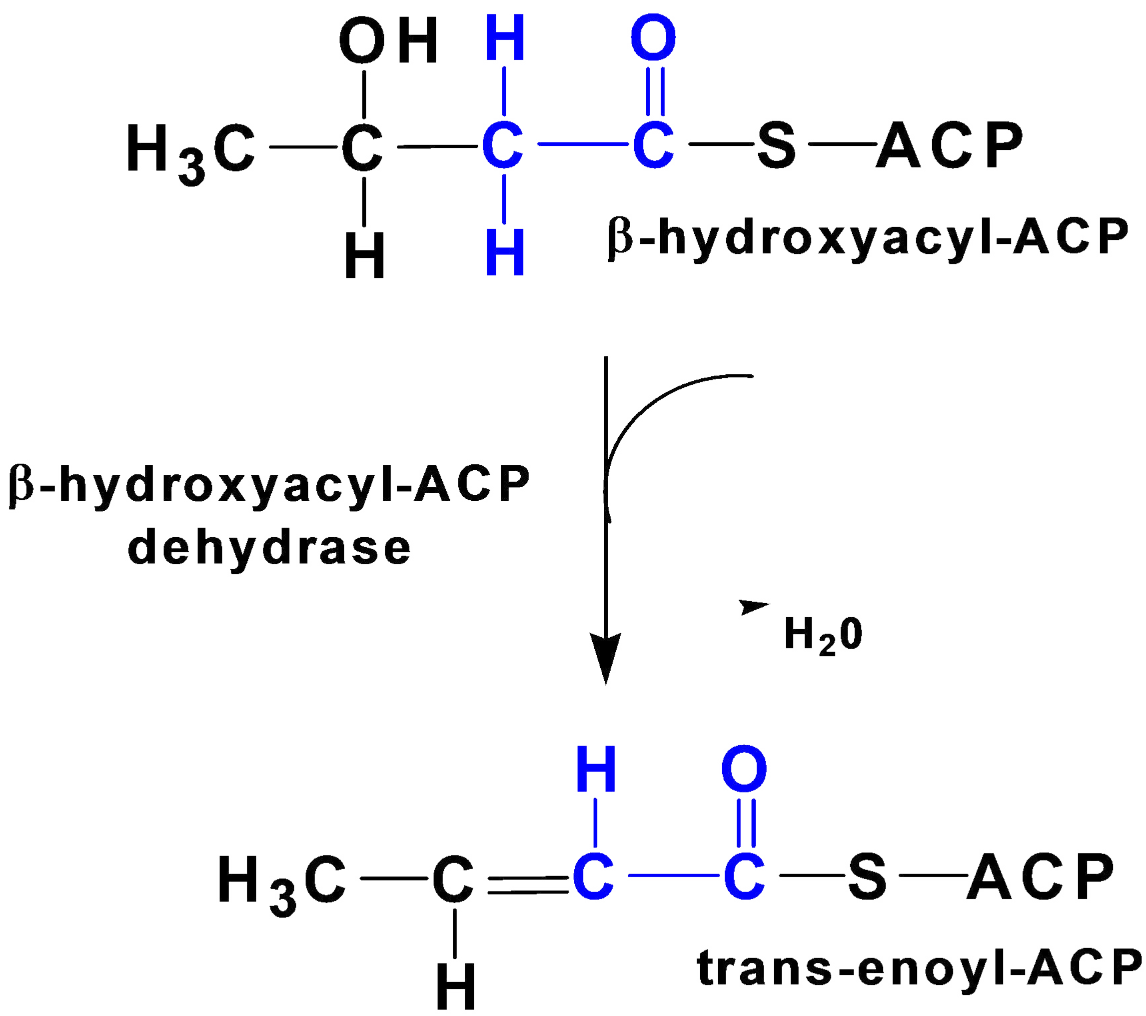
Step 3: Reduction



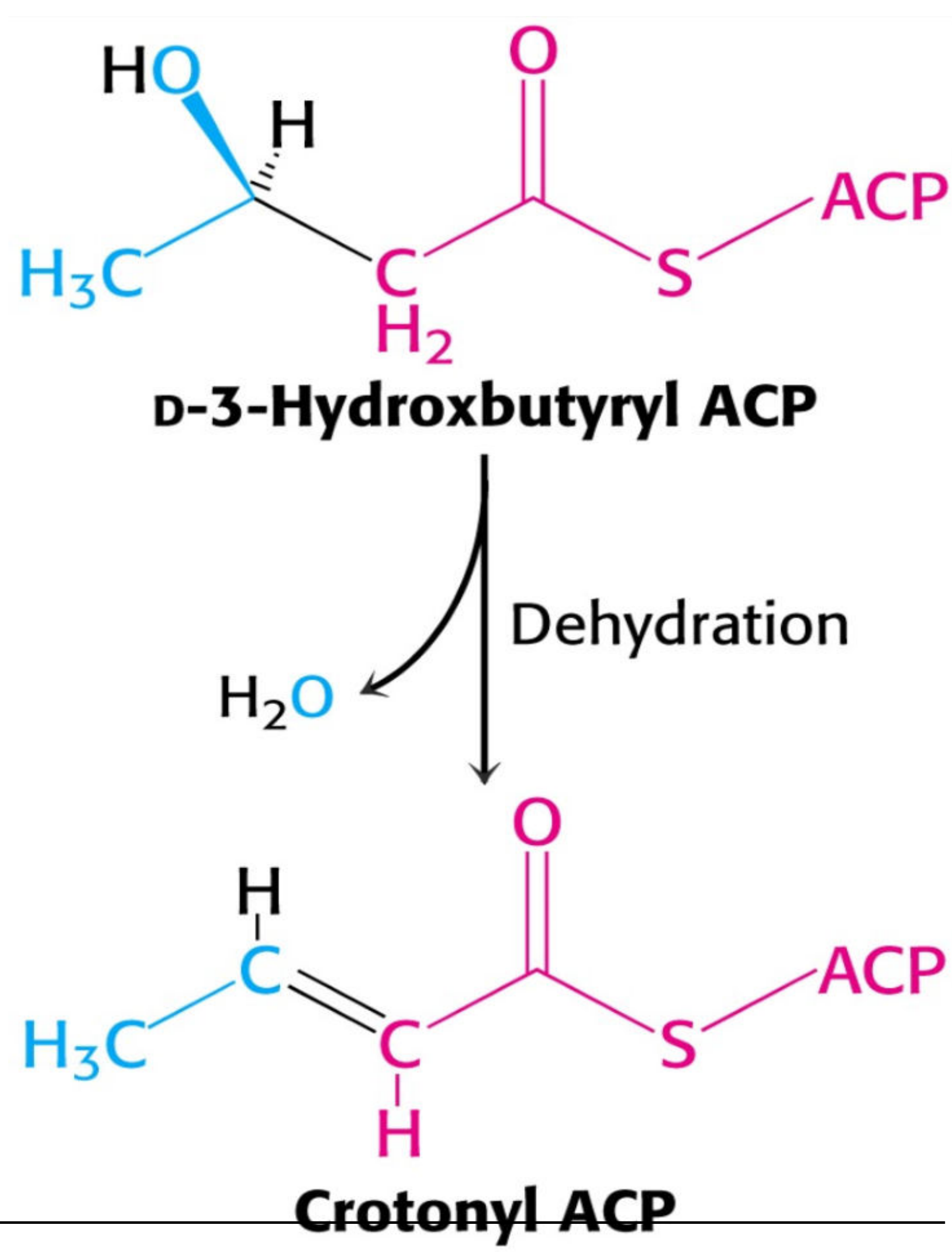
Reduction Reaction



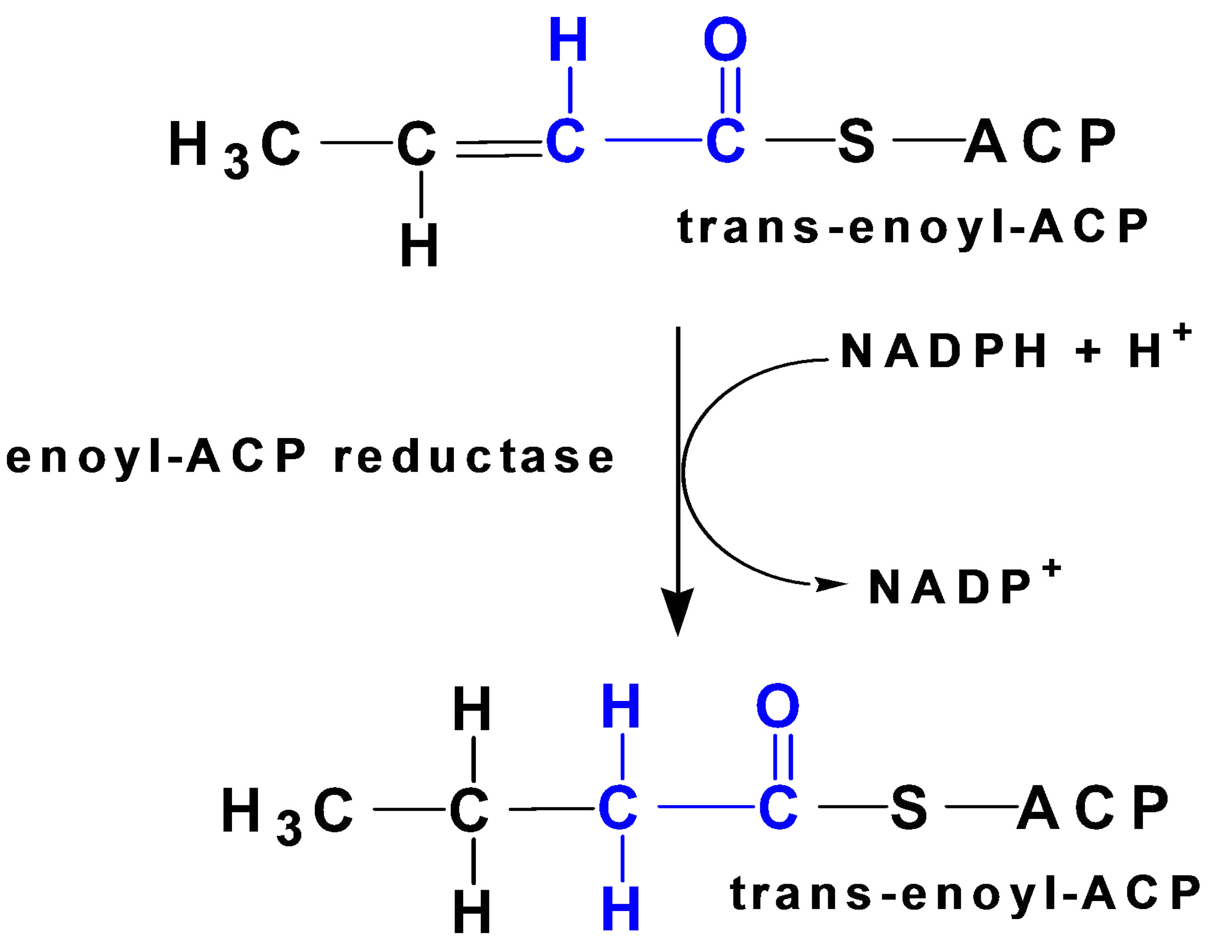
Step 4: Dehydration



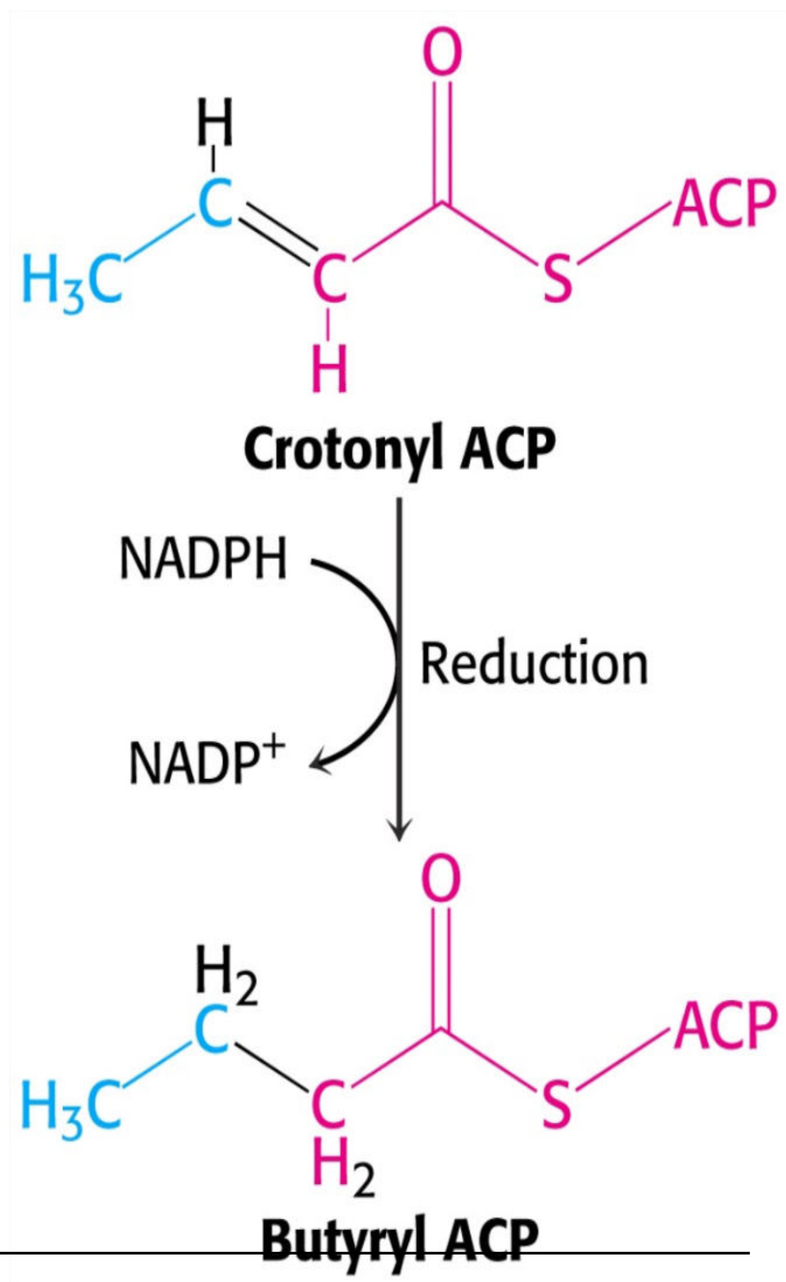
Dehydration



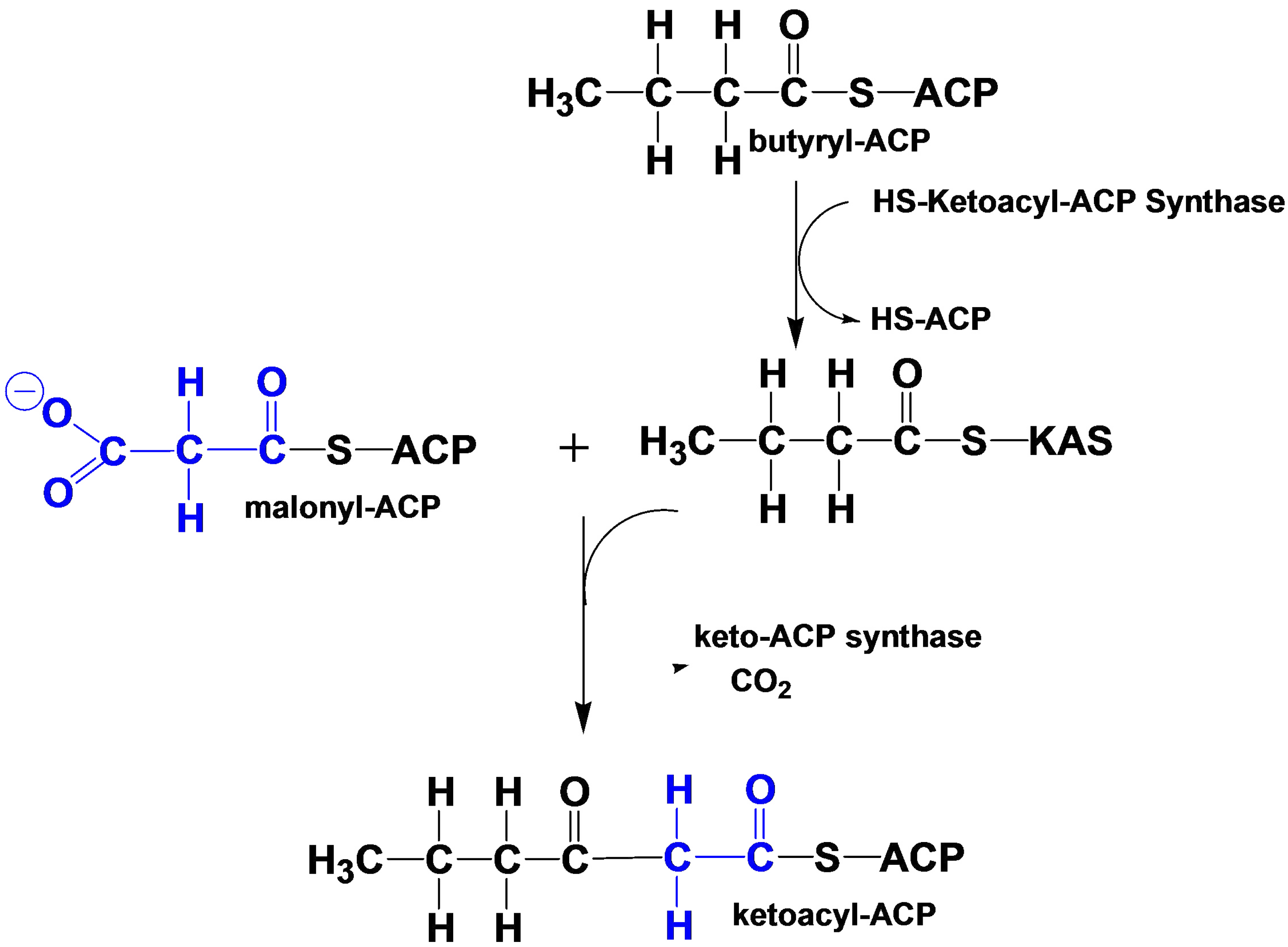
Step 5: Reduction



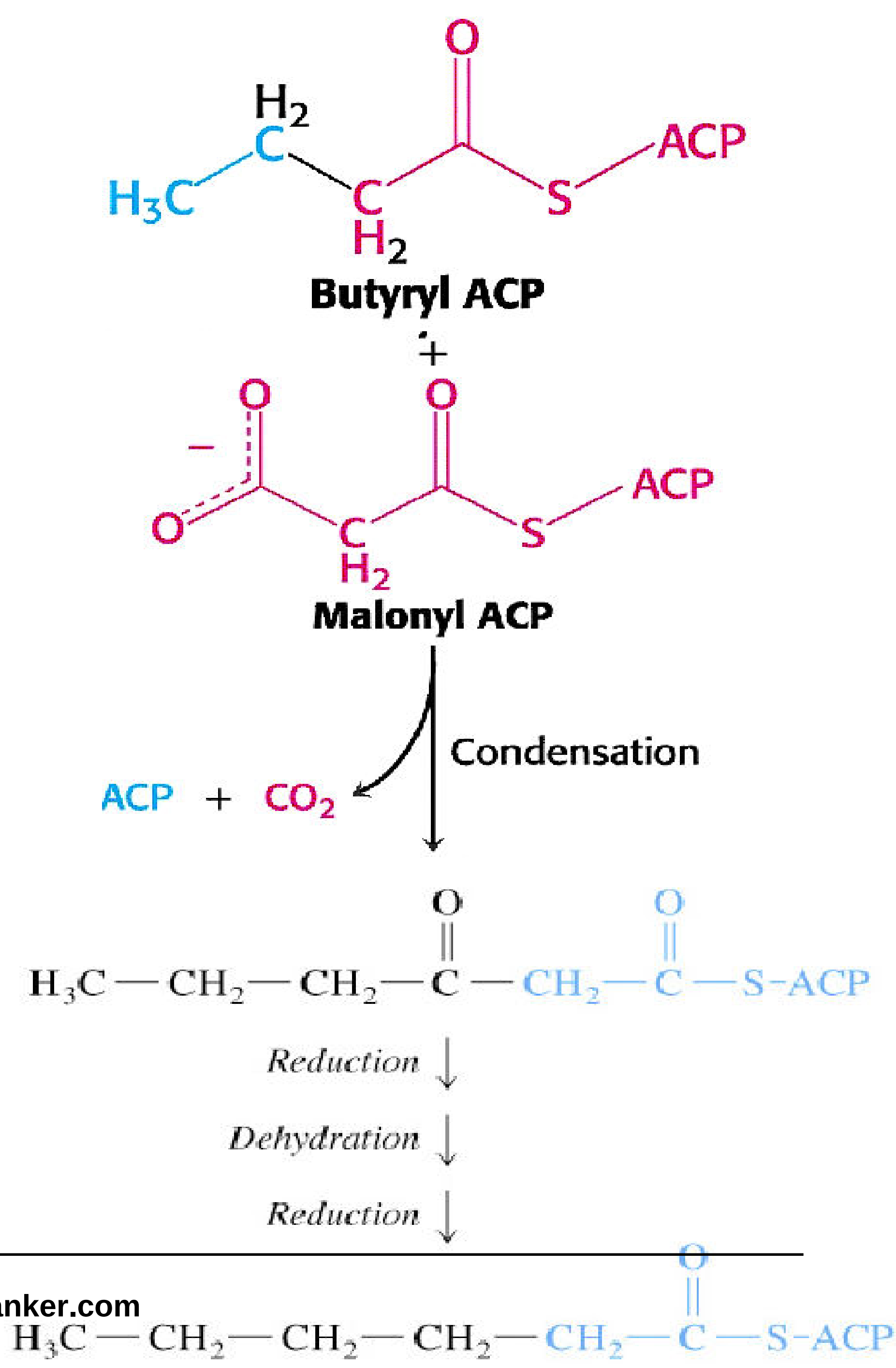
Reduction

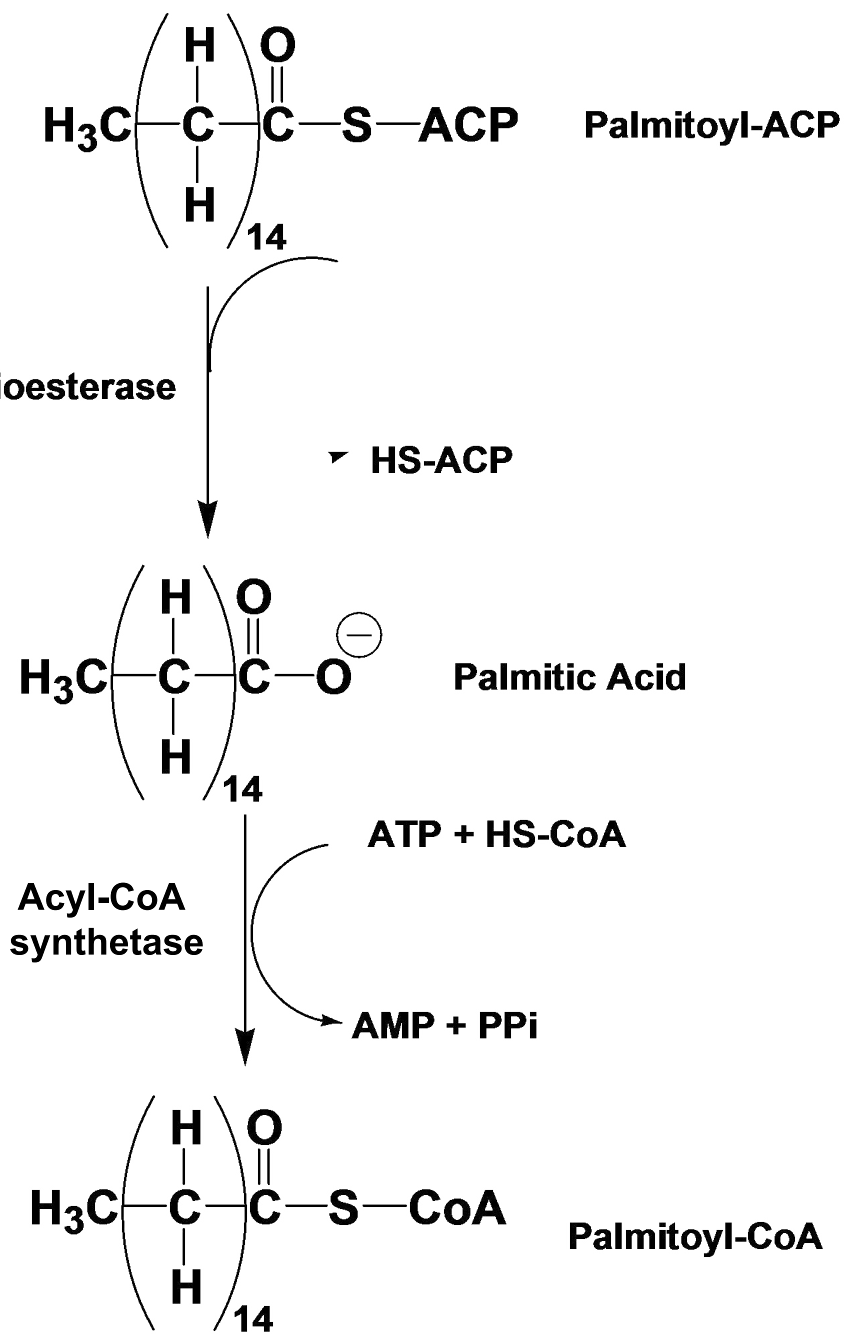


Step 6: Next condensation



Repetitions Of 7 Cycles





Termination of Fatty Acid Synthesis

Final reaction of FA synthesis is Cleavage

- Palmitoyl-ACP is hydrolyzed by a ***Thioesterase***

$$\text{Palmitoyl-ACP} \xrightarrow[\text{Thioesterase}]{H_2O} \text{Palmitate} + \text{HS-ACP}$$

Overall Reaction of Palmitate Synthesis from Acetyl CoA and Malonyl CoA

Acetyl CoA + 7 Malonyl CoA + 14 NADPH + 14 H^+

↓

Palmitate + 7 CO_2 + 14 $NADP^+$ + 8 HS-CoA + 6 H_2O

Principal reactions in fatty acid synthesis in bacteria

Step	Reaction	Enzyme
1	Acetyl CoA + HCO_3^- + ATP ⟶ malonyl CoA + ADP + P_i + H^+	Acetyl CoA carboxylase
2	Acetyl CoA + ACP ⇌ acetyl ACP + CoA	Acetyl transacylase
3	Malonyl CoA + ACP ⇌ malonyl ACP + CoA	Malonyl transacylase
4	Acetyl ACP + malonyl ACP ⟶ acetoacetyl ACP + ACP + CO_2	Acyl-malonyl ACP condensing enzyme
5	Acetoacetyl ACP + NADPH + H^+ ⇌ D-3-hydroxybutyryl ACP + $NADP^+$	β-Ketoacyl ACP reductase
6	D-3-Hydroxybutyryl ACP ⇌ crotonyl ACP + H_2O	3-Hydroxyacyl ACP dehydratase
7	Crotonyl ACP + NADPH + H^+ ⟶ butyryl ACP + $NADP^+$	Enoyl ACP reductase

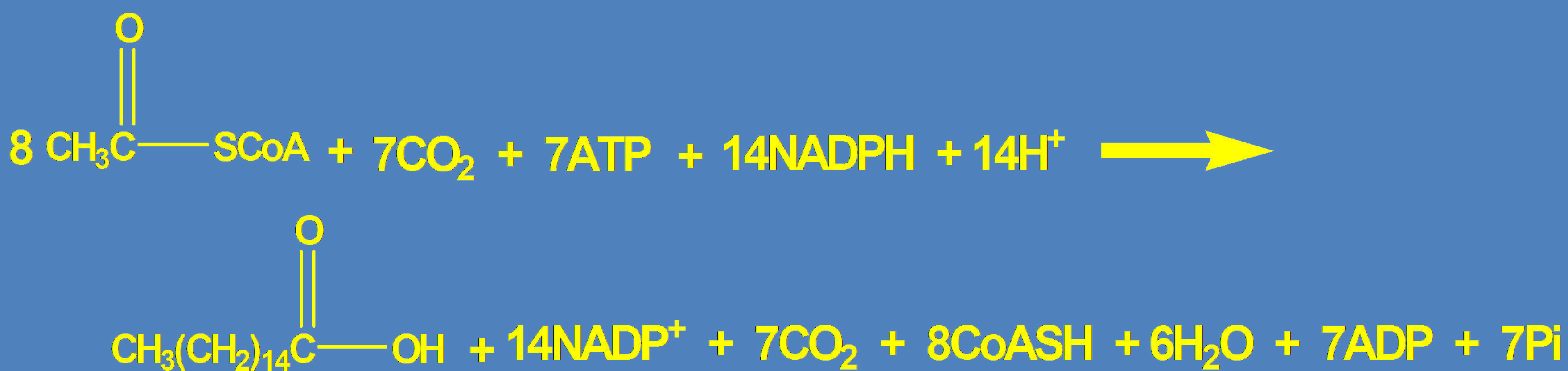
Summary based on Malonate as an input:

Acetyl-CoA + 7 Malonyl-CoA + 14 NADPH → Palmitate + 7 CO_2 + 14 $NADP^+$ + 8 CoA

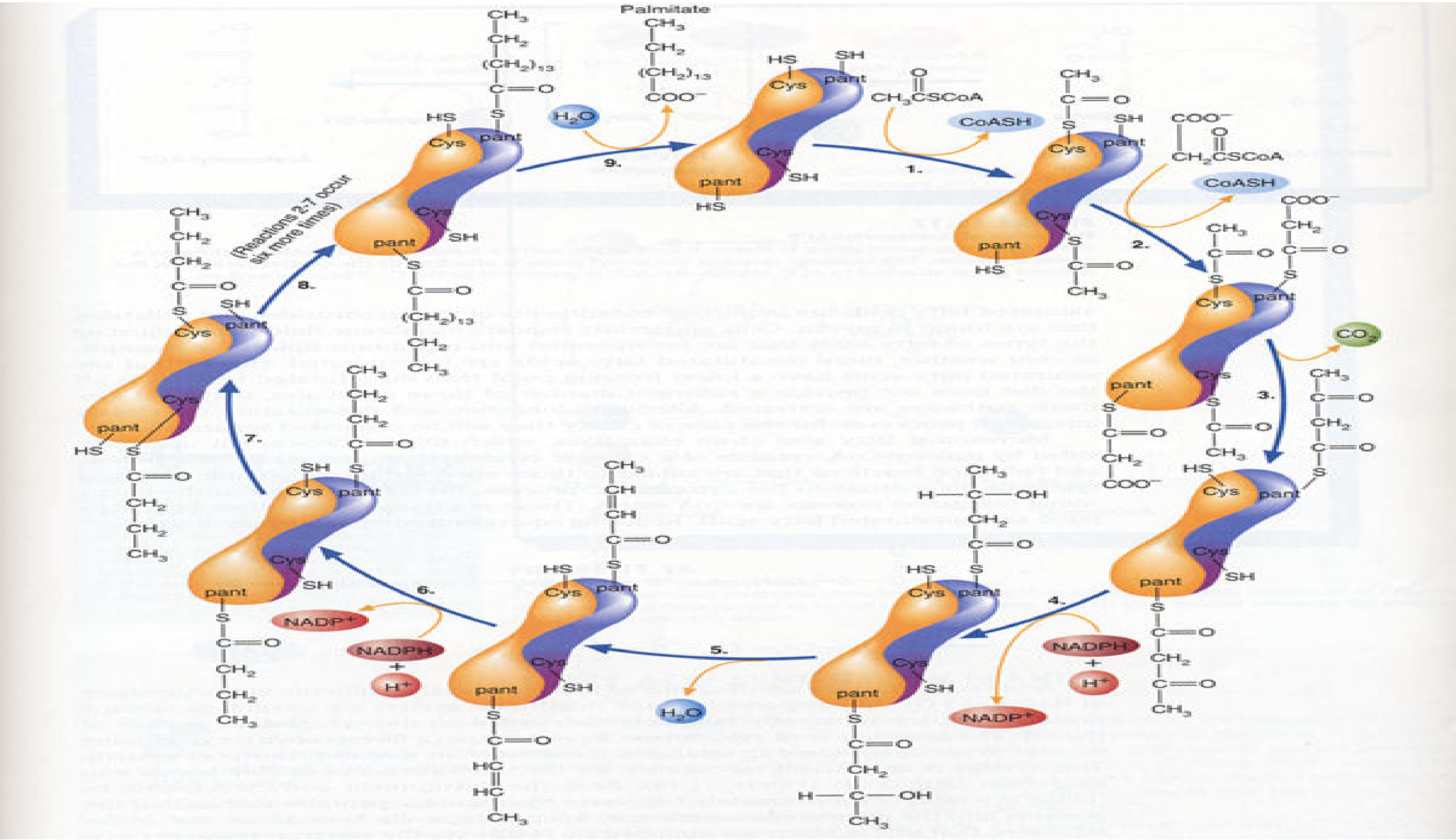
Fatty acid synthesis occurs in **cytosol**. Acetyl-CoA generated in mitochondria is transported to cytosol via a shuttle mechanism involving **Citrate**.

Stoichiometry for Palmitic Acid Synthesis

From acetyl-CoA



Diagrammatic View of Fatty Acid Biosynthesis



Energetics Of De Novo Synthesis Of Fatty Acids

- De Novo Fatty acid biosynthesis is an **Anabolic process involving use of ATPs.**

- **Total 23 ATPs are utilized** during biosynthesis of **one molecule of Palmitate.**

- **2 ATPs** are used for **1 Acetyl-CoA** translocation through **Citrate transport** system
  - **For 8 Acetyl CoA translocation uses 16 ATPs**

- **1 ATP** each is used for **Acetyl CoA Carboxylation** to Malonyl CoA.
  - To form **7 Malonyl CoAs 7 ATPs** are utilized.

- **16+7 =23 ATPs Net utilized**

Regulation Of Fatty Acid Biosynthesis

Nutritional Status Regulates Lipogenesis

- High Carbohydrate
- High Lipid Diet
- Acyl-CoA Inhibits Pyruvate Dehydrogenase

Enzyme Acetyl-CoA Carboxylase Is a Regulatory ,Key Enzyme Of De Novo Fatty acid Synthesis.

- **Committed Step of Fatty Acid Synthesis**
  - Carboxylation of Acetyl CoA to Malonyl CoA
  - **By Acetyl CoA Carboxylase - Biotin**

- **Carboxylation of Acetyl-CoA to form Malonyl-CoA**

- **Is an Irreversible, committed step in Fatty acid biosynthesis**

Modes Of Regulation Of Acetyl CoA Carboxylase of FA Biosynthesis

Acetyl-CoA Carboxylase is regulated by 3 modes:

1. Hormonal Influence
2. Allosteric Control
3. Covalent Modification

1. Hormonal Influence

- **ACC is an Inducible Enzyme:**
  - –Induced by **Insulin**
    - **Insulin activates ACC**
  - –Repressed by **Glucagon**
  - **–Glucagon inhibits ACC**

2. Allosteric Modifiers

- Citrate Activates Acetyl-CoA Carboxylase (Feed Forward)
- Fatty Acyl-CoAs inhibit Acetyl-CoA Carboxylase

Allosteric modification of Acetyl-Co A Carboxylase

- **Activated by**: Citrate
- **Inhibited by:** Long Chain Fatty Acid

- Body with **high levels of cellular Citrate**
- Stimulate De novo biosynthesis of Fatty acids.

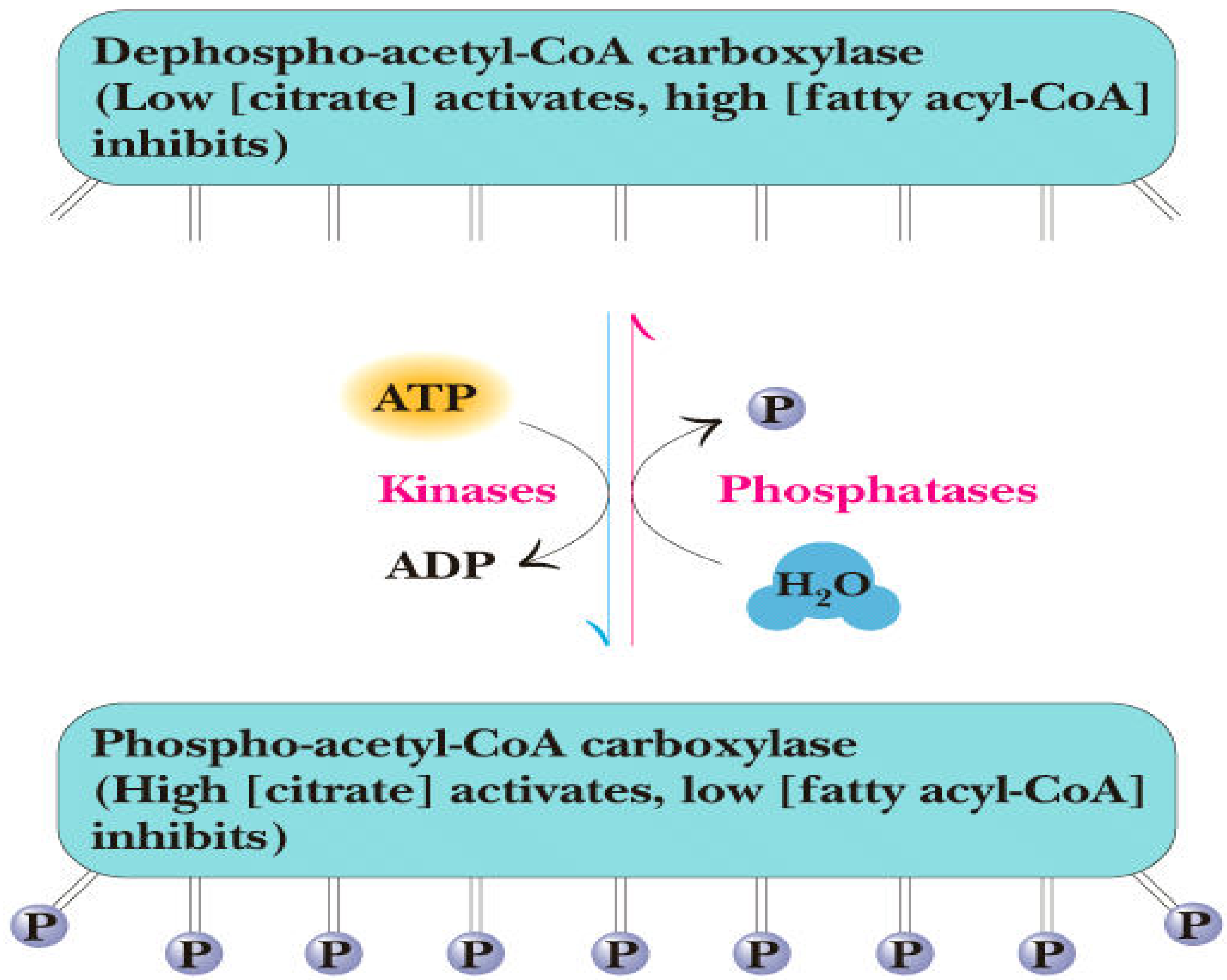
- Body on a **high fat diet** experience **little** if any **de novo fatty acid synthesis.**

3. Covalent Modification Of Acetyl-CoA Carboxylase(ACC)

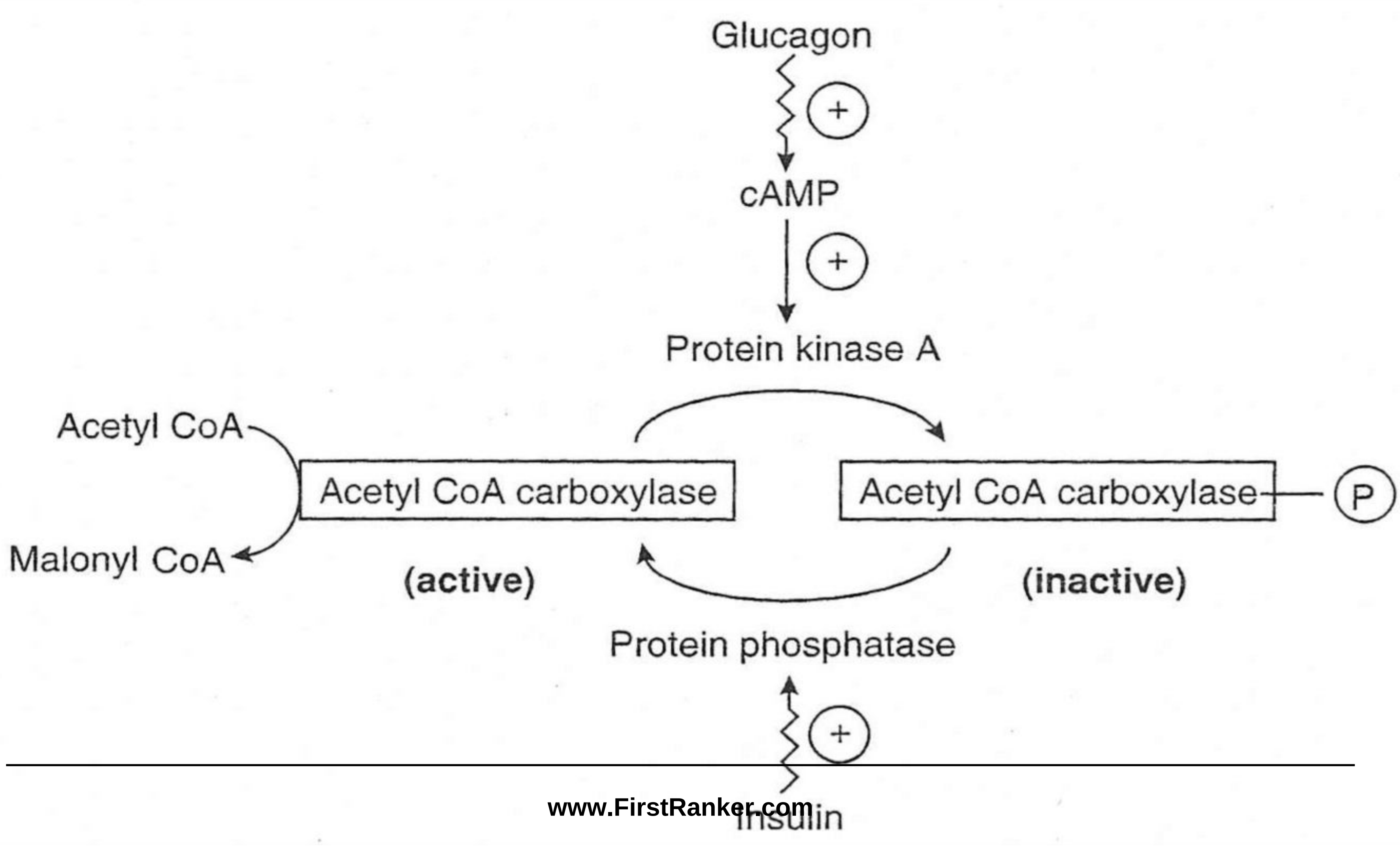
- **ACC is Activated by : Dephosphorylation**

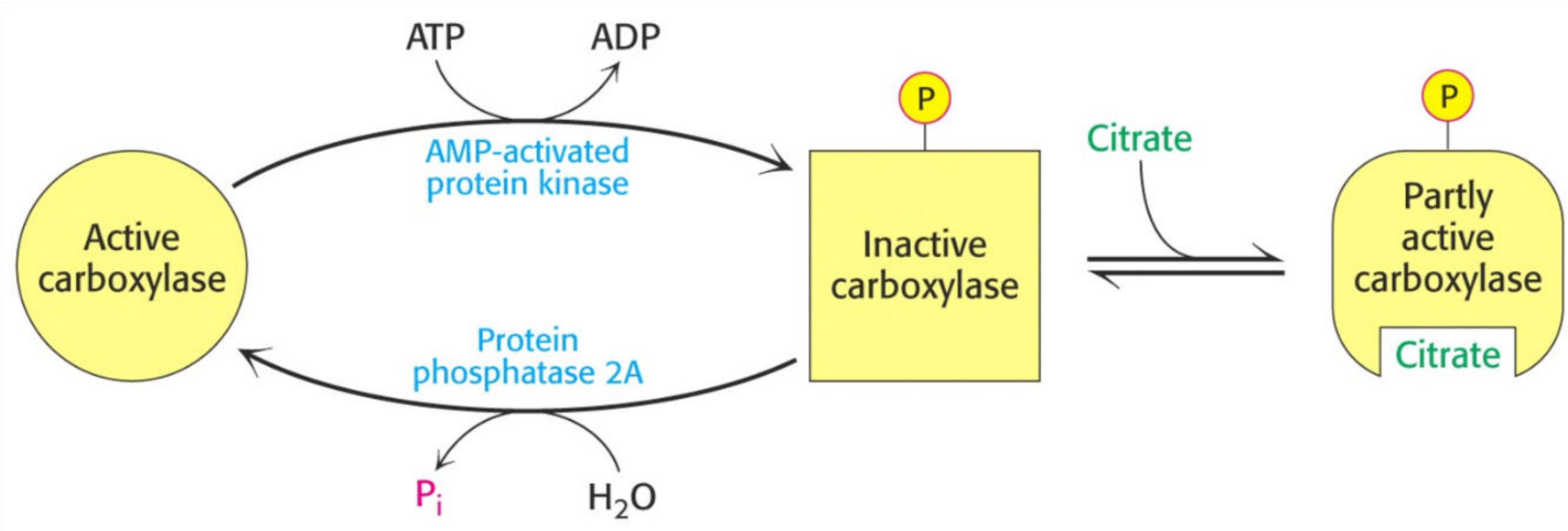
- **ACC is Inhibited by: Phosphorylation**

Covalent Modification Of ACC



Covalent Regulation OF Acetyl CoA Carboxylase

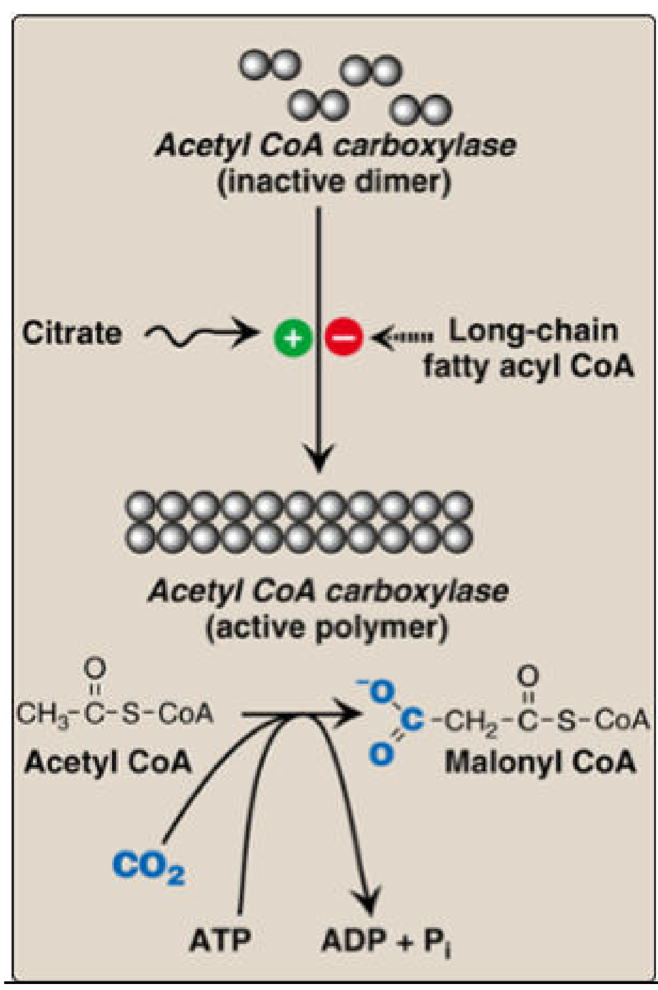


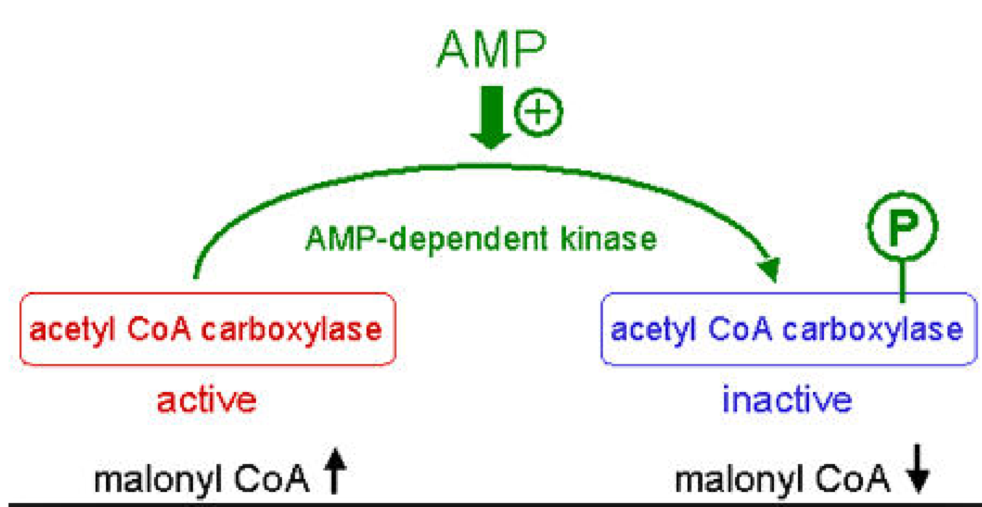


- **Activation of ACC**
- **In a well Fed state**
  - **Insulin** induces **Protein Phosphatase**
  - **Activates ACC by De phosphorylation**

- **Inactivation of ACC**
- **In a Starved state**
  - Glucagon increases **cAMP**
  - Activates **Protein kinase A**
  - Inactivates ACC by **Phosphorylation**

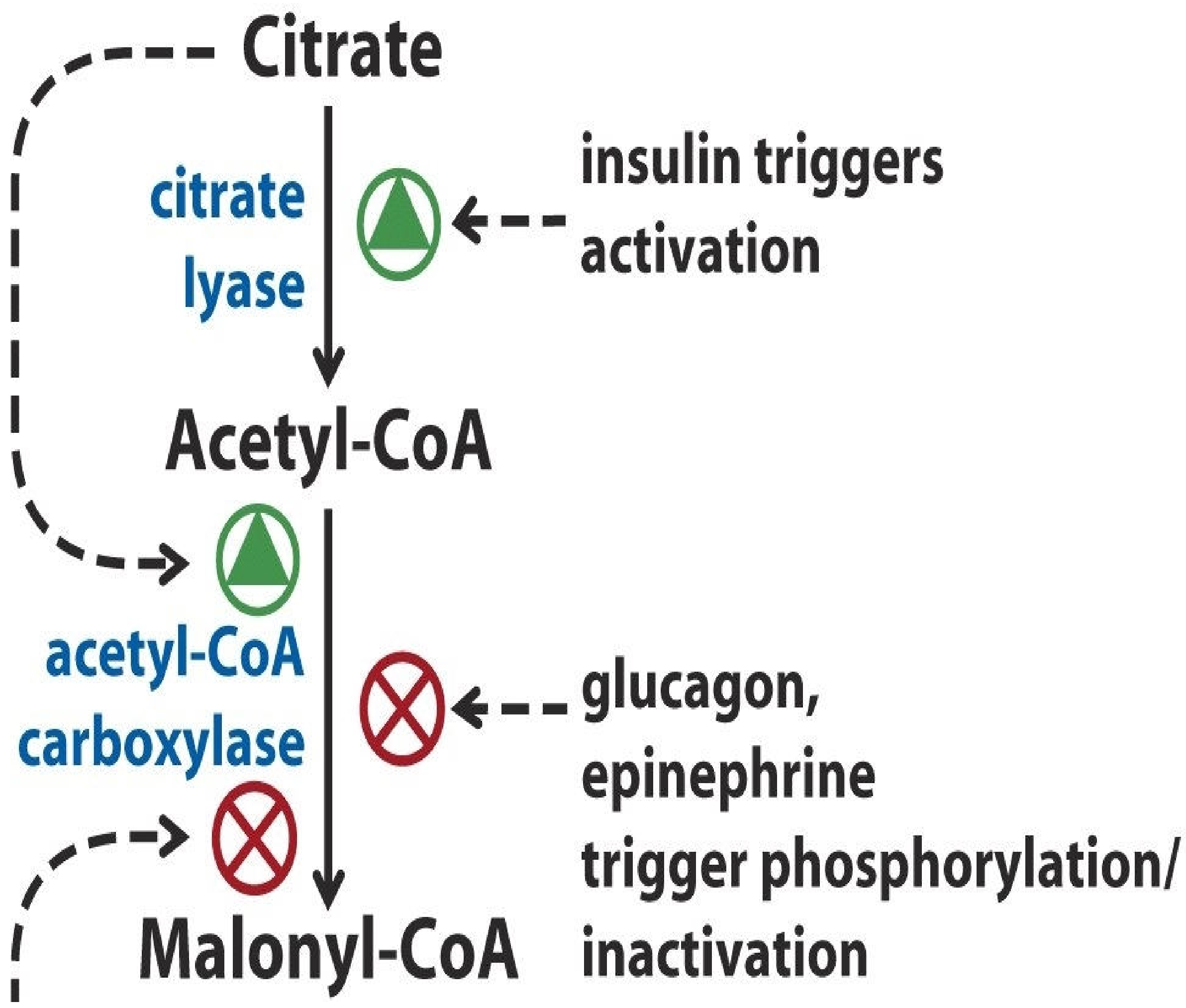
Acetyl-CoA Carboxylase





glucagon, epinephrine trigger phosphorylation/ inactivation

Control of Fatty Acid Synthesis



Remember

Lipogenesis Is Inhibited
In
Type I Diabetes Mellitus And
Obesity

Biosynthesis and Degradation of Fatty Acid are Reciprocally Regulated

Very Well Coordination And Regulation Of Lipolysis And Lipogenesis Is A Healthy Lipid Metabolism

Both Lipogenesis And Lipolysis Should Be Kept In Dynamism For Good Health

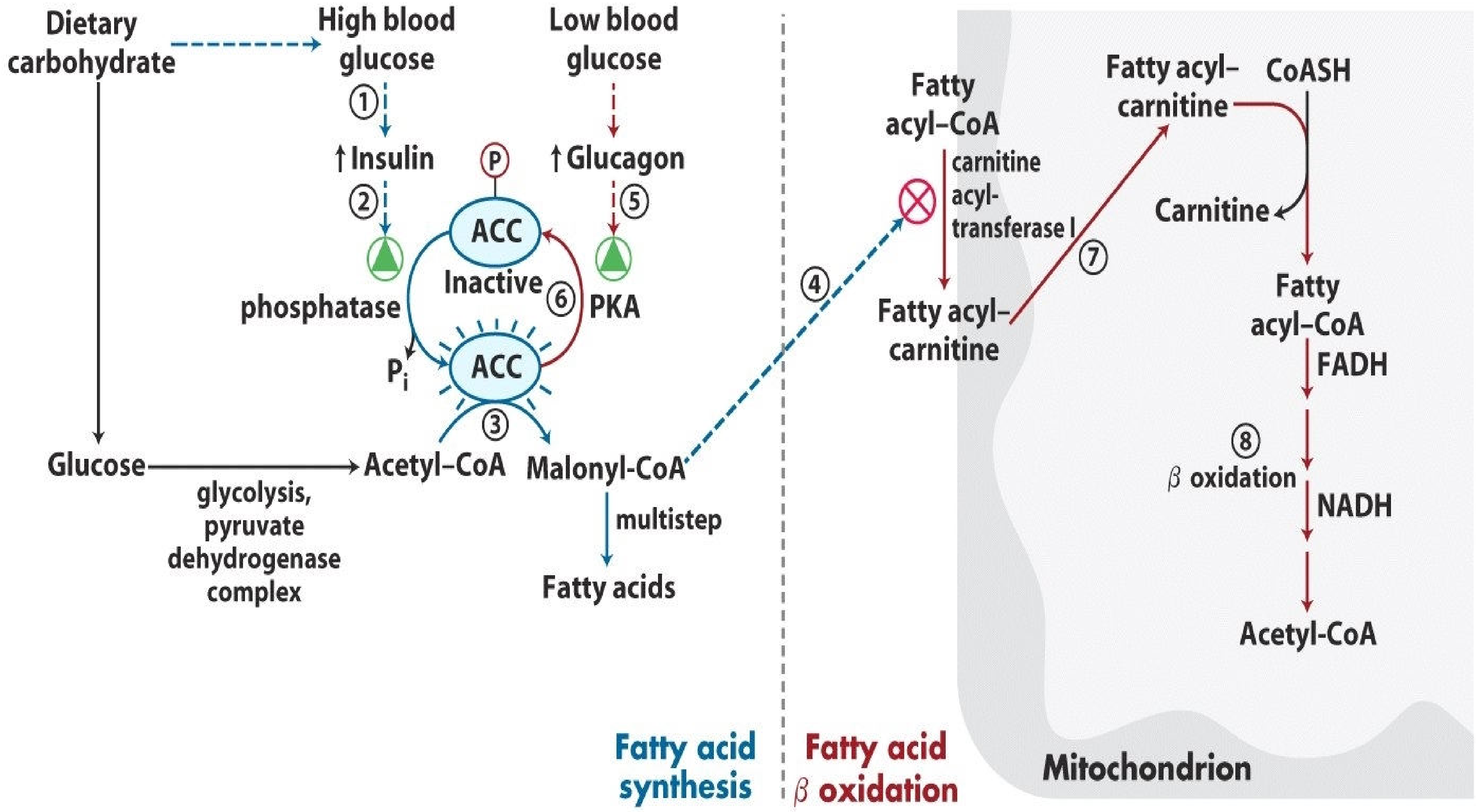
Well Regulated Lipolysis And Lipogenesis Prevent From Lipid Associated Disorders

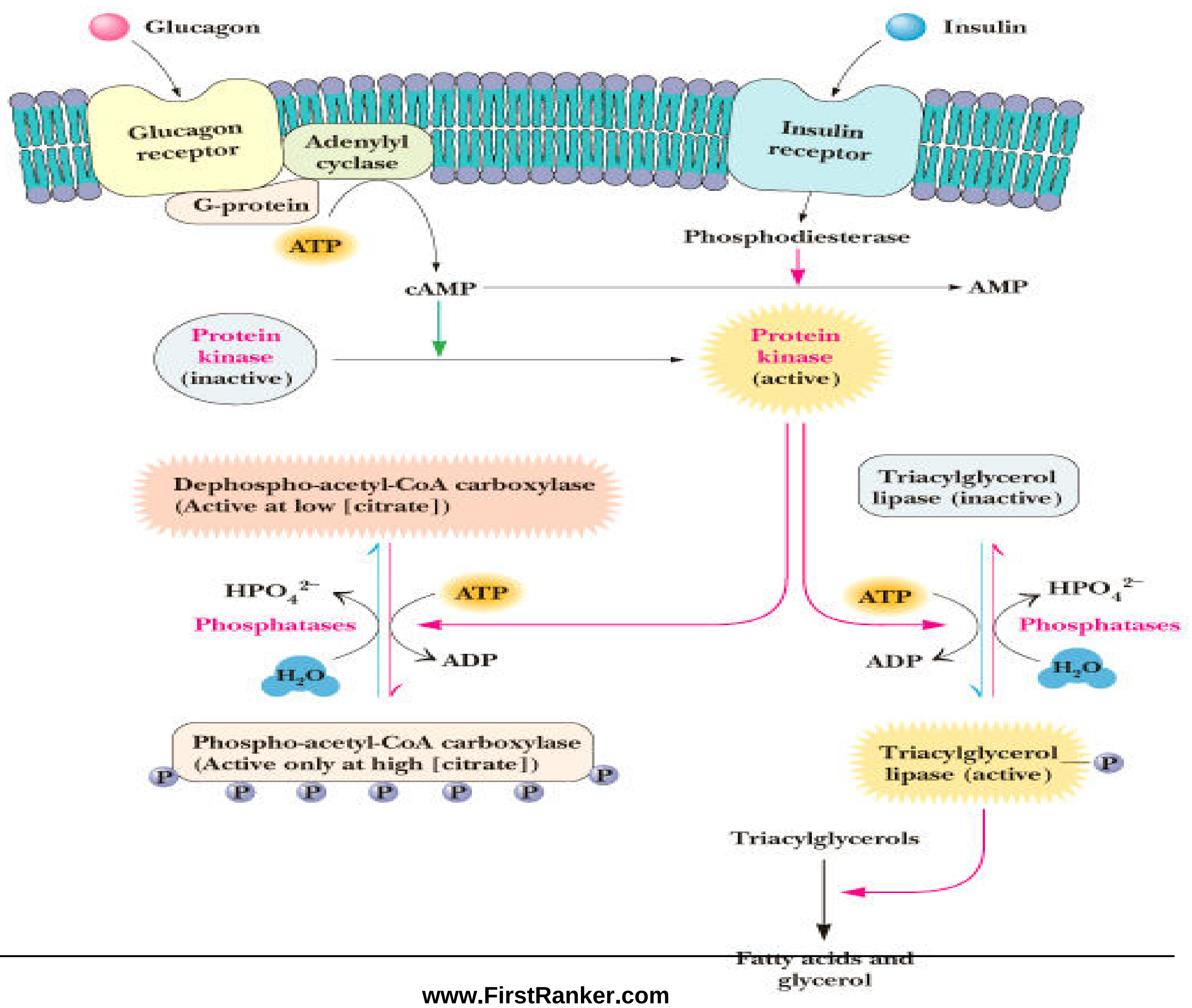
– **During Starvation**
– Epinephrine & Glucagon Stimulate Lipolysis
– Brings degradation of FA

– **Well Fed state**
- ❖ Insulin inhibits Lipolysis
- ❖ Insulin Stimulates Fatty acid biosynthesis.

- **ACC also influences degradation of Fatty acids.**
  – **Malonyl CoA** inhibits **Carnitine Acyltransferase I activity.**
  – This **limits Beta oxidation of Fatty acids** in Mitochondrial Matrix.

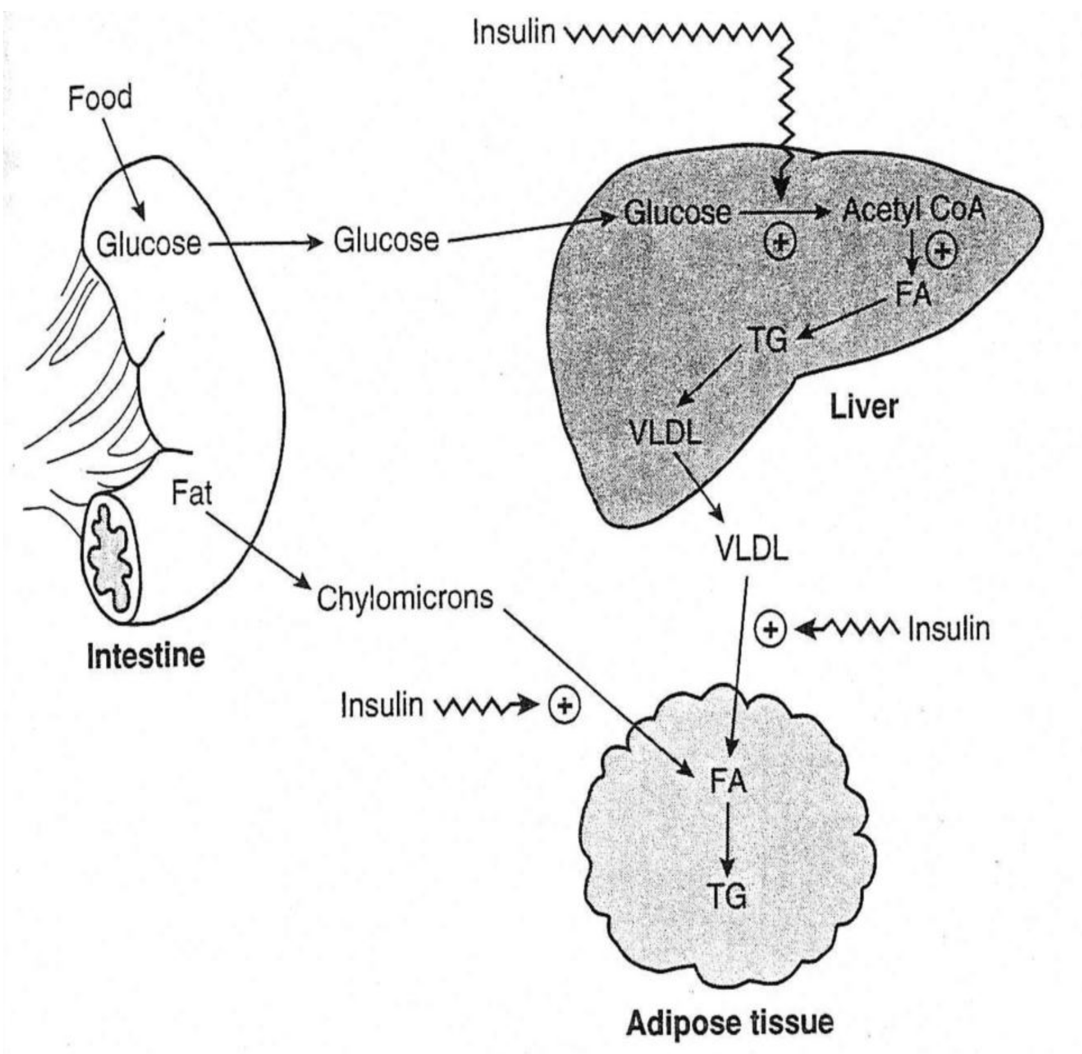
Reciprocal Control





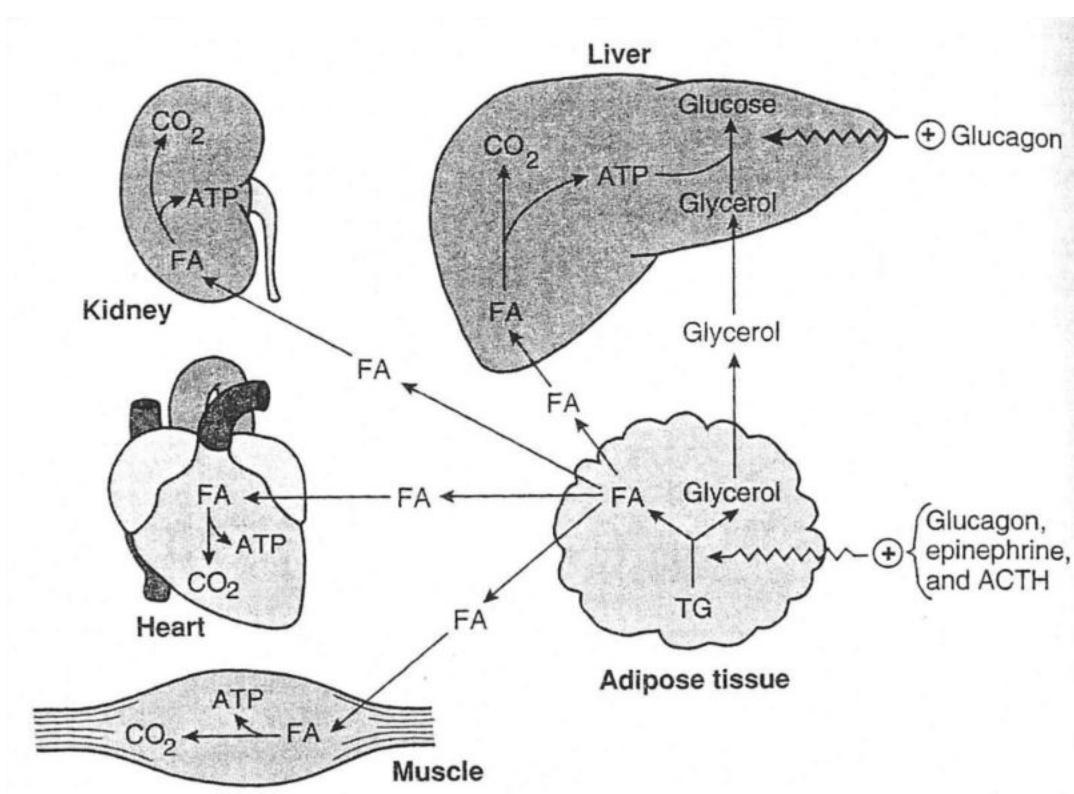
Overview of Fatty Acid Metabolism: Insulin Effects

- Liver
  - Increased fatty acid synthesis
    - Glycolysis, PDH, FA synthesis
  - Increased TAG synthesis and transport as VLDL
- Adipose
  - Increased VLDL metabolism
    - lipoprotein lipase
  - Increased storage of lipid
    - Glycolysis



Overview of Fatty Acid Metabolism: Glucagon/Epinephrine Effects

- Adipose
- Hormone-sensitive lipase Increased
  - Increased TAG mobilization
- Increased FA oxidation
  - All tissues Except CNS and RBC



Post-Synthesis Modifications Of Biosynthesized Fatty Acids

- **C16 Saturated fatty acid (Palmitate) is product which may undergo:**
  - Elongation
  - Unsaturation
  - Incorporation to form Triacylglycerols
  - Incorporation into Acylglycerol phosphates to form Phospholipids

Chain Elongation Of Fatty Acids

Occurs In
Mitochondria
And
Smooth Endoplasmic Reticulum

Elongation Of Fatty Acids
In Microsomes /Mitochondria
To
Synthesize Long Chain Fatty Acids

- Palmitate biosynthesized by **De Novo Biosynthesis in Cytosol by the activity of FAS Complex**

- Is further **elongated** to more higher Fatty acid either in **Mitochondria /Endoplasmic reticulum.**

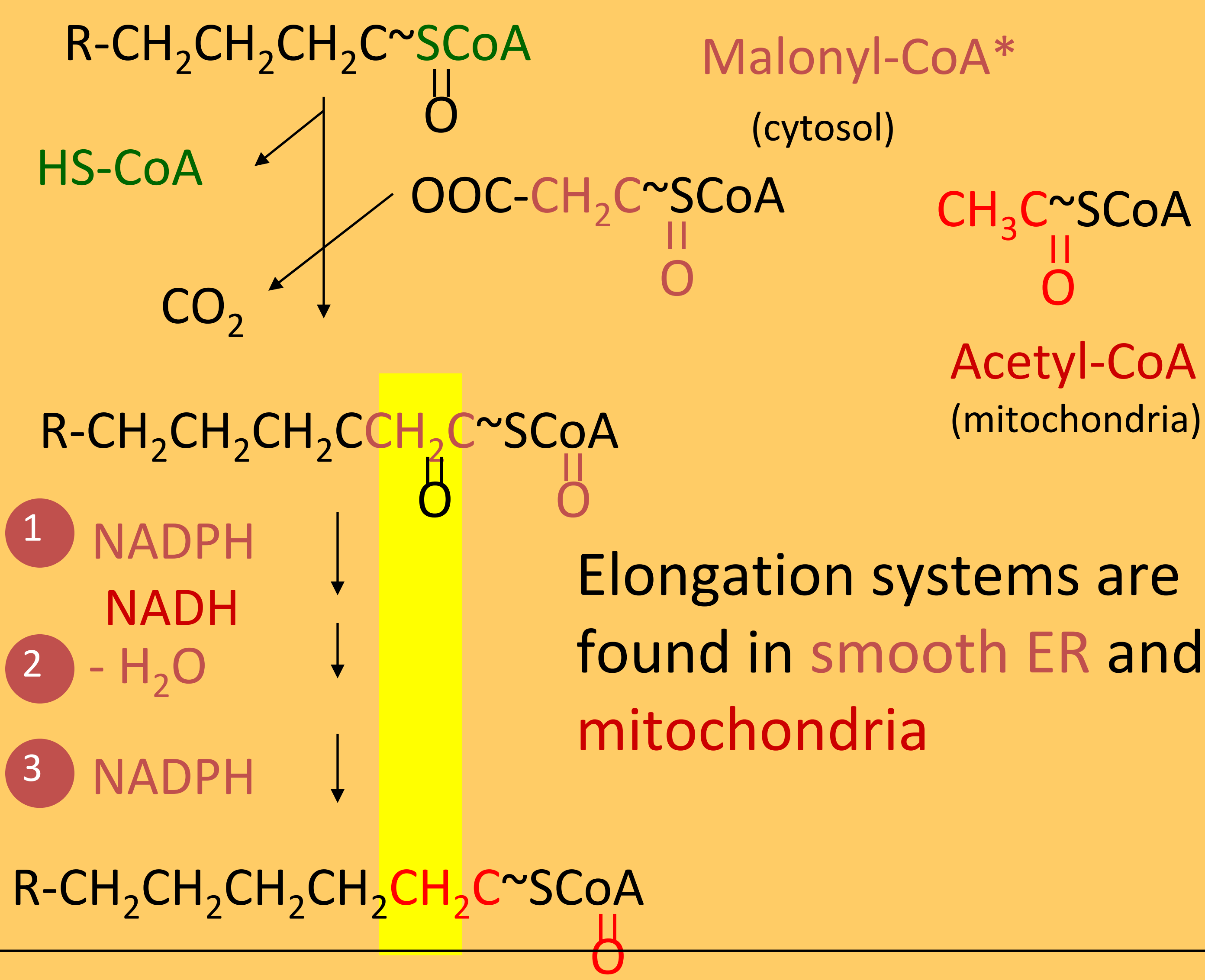
Mitochondrial Chain Elongation

- Here **Acetyl-CoA is successively added** to Fatty acid chain lengthened
- In presence of reducing equivalents **NADPH+ H^+**
- Steps are **almost reversal of Beta Oxidation of Fatty acids.**

Microsomal/ER Chain Elongation Of Fatty Acid

- This is **more predominant way** of **Fatty acid Chain Elongation**.
- It involves **successive addition of Malonyl-CoA** with the participation of **NADPH+ H+** and enzyme **Elongases.**

Elongation of Chain (Two Systems)



Elongation systems are found in smooth ER and mitochondria

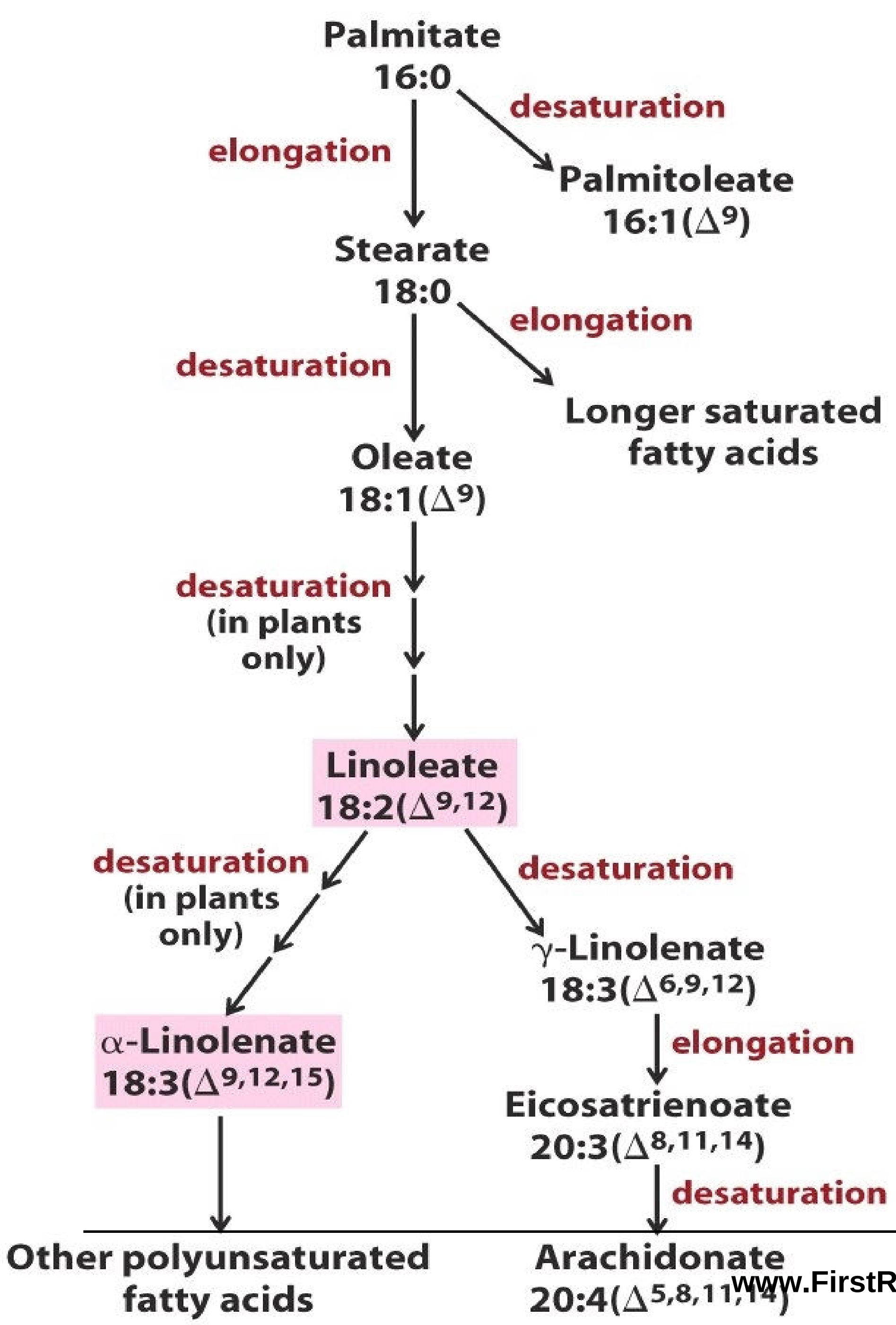
Synthesis Of Unsaturated Fatty Acids

Mammals can Biosynthesize Long Chain And Monounsaturated Fatty acids

Using Elongation And Desaturation

Desaturation of Fatty Acid Chain In Microsomes

- Enzyme **Fatty Acyl-CoA Desaturase** which is a Flavoprotein
- Helps in **creating double bonds** and forming **Mono Unsaturated Fatty acids.**



Palmitic acid modifications

- Cell makes a pool of palmitic acid that it can elongate and/or desaturate in the ER.
- Elongation system is very similar to synthesis: 2C units added from malonyl-CoA.

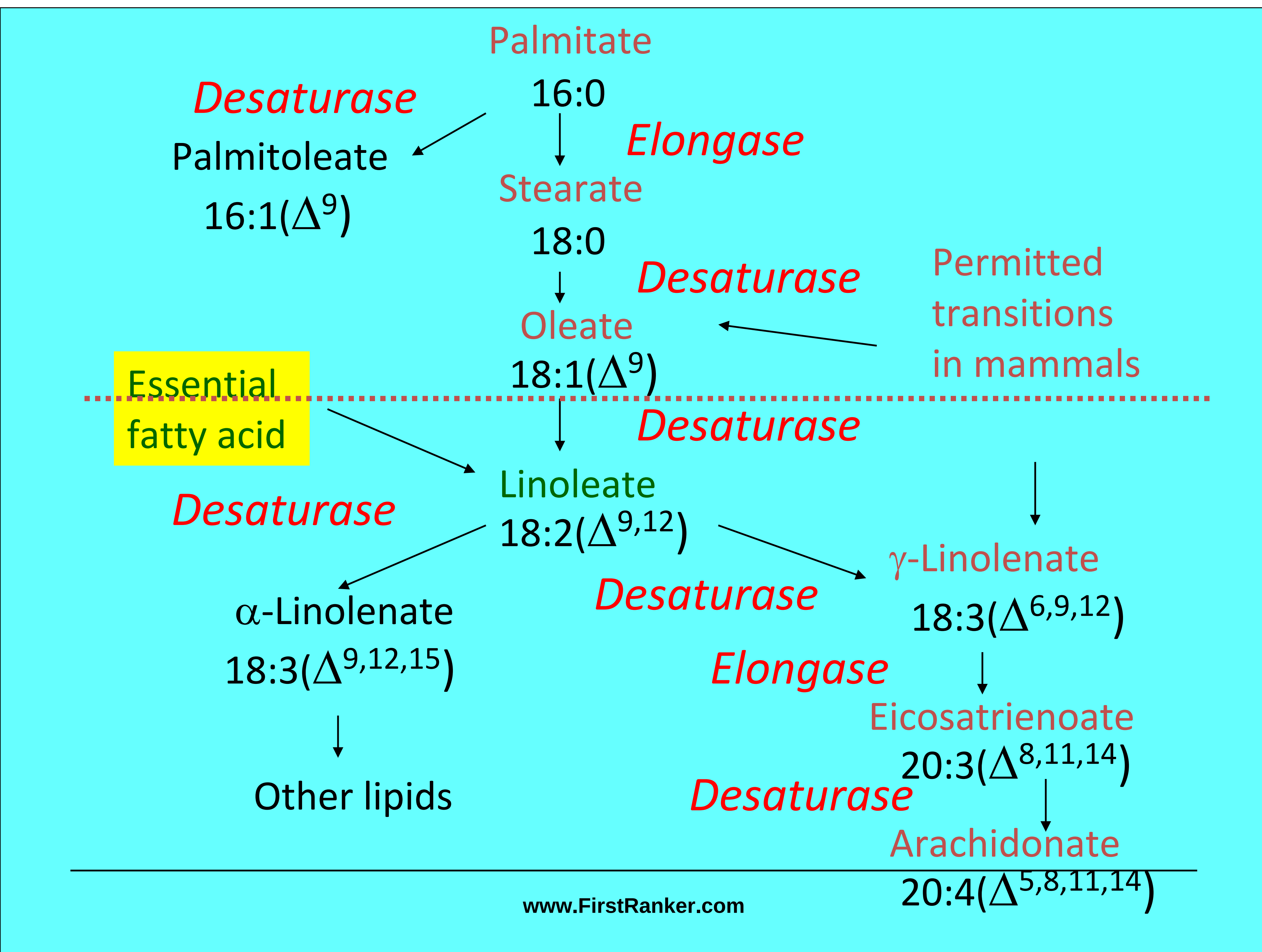
- **Palmitic acid and Stearic acid** on **Desaturation**
- **Forms corresponding MUFAS Palmitoleic and Oleic acid respectively.**

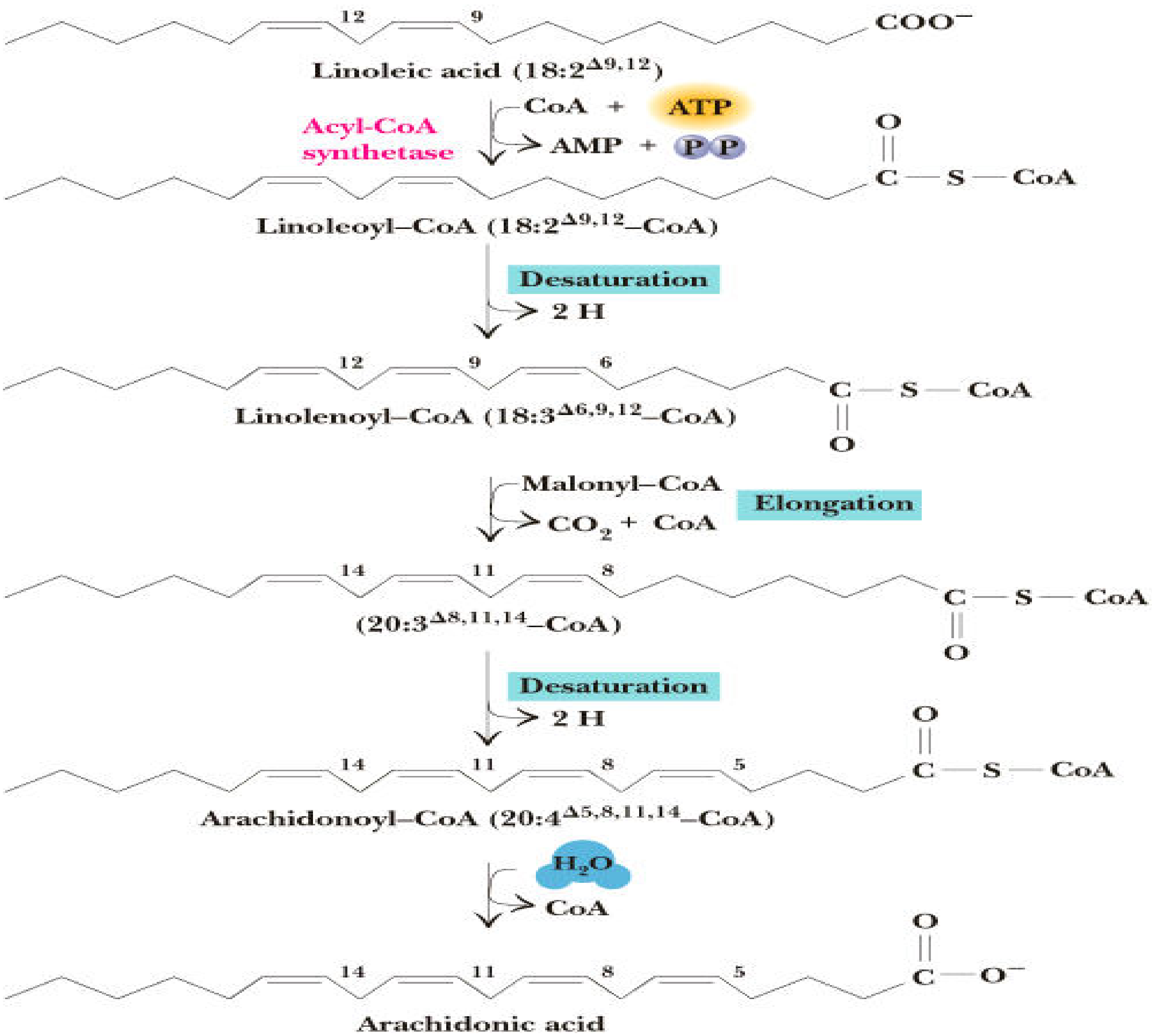
- Human body **lack ability** to **introduce double bonds beyond carbon 9 and 10 of Fatty acids**.

- Hence body **cannot biosynthesize Linoleic and Linolenic acid** and become dietary essential Fatty acids.

- However **Linoleic Acid by Chain Elongation and Desaturation**

- **Forms Arachidonic acid** in Human body.



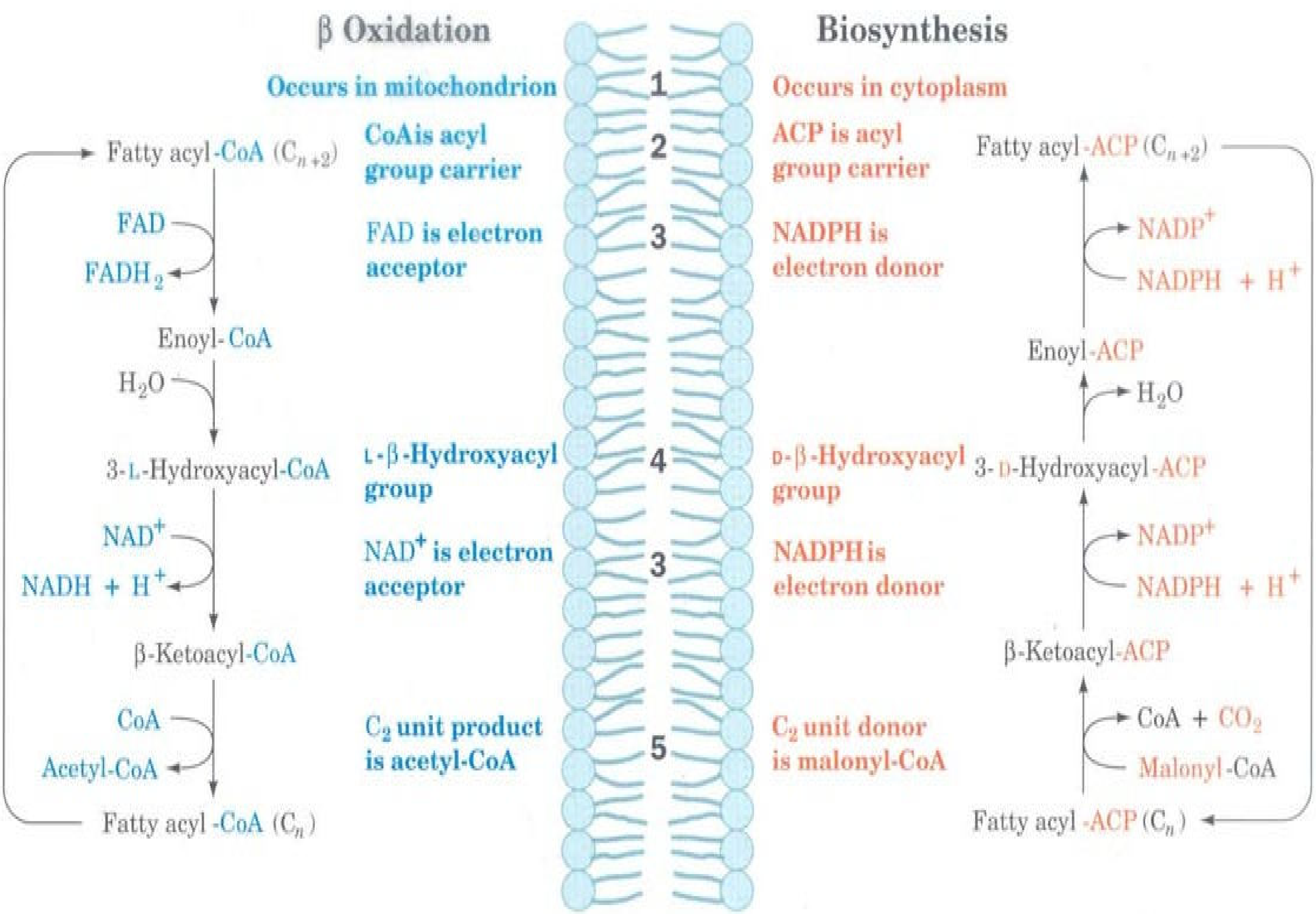


Differences Between Beta Oxidation Of Fatty Acid And De Novo Biosynthesis Of Fatty Acids

Biosynthesis and Degradation Pathways are Different

- Major differences between Fatty acid breakdown and biosynthesis are as:

Beta Oxidation Palmitic acid Pathway	De Novo Biosynthesis Palmitic acid Pathway
Catabolic /Oxidative	Anabolic /Reductive
Occurs In Mitochondria	Occurs In Cytosol
Acetyl CoA is an end product	Acetyl CoA is a precursor
Beta Carbon **CH2 is** transformed to **C=O**	Beta Carbon **C=O** is converted to **CH2**
Generates 106 ATPs	Utilizes 23 ATPs
Coenzymes FAD and NAD^+ are involved	Coenzymes NADPH $+H^+$ is involved
CoA is an Acyl Carrier	ACP is an Acyl Carrier



Fatty Acid Synthesis

- C=O → -CH2

Fatty Acid Beta Oxidation

- CH2 → C=O

Triacylglycerol (TAG) Biosynthesis

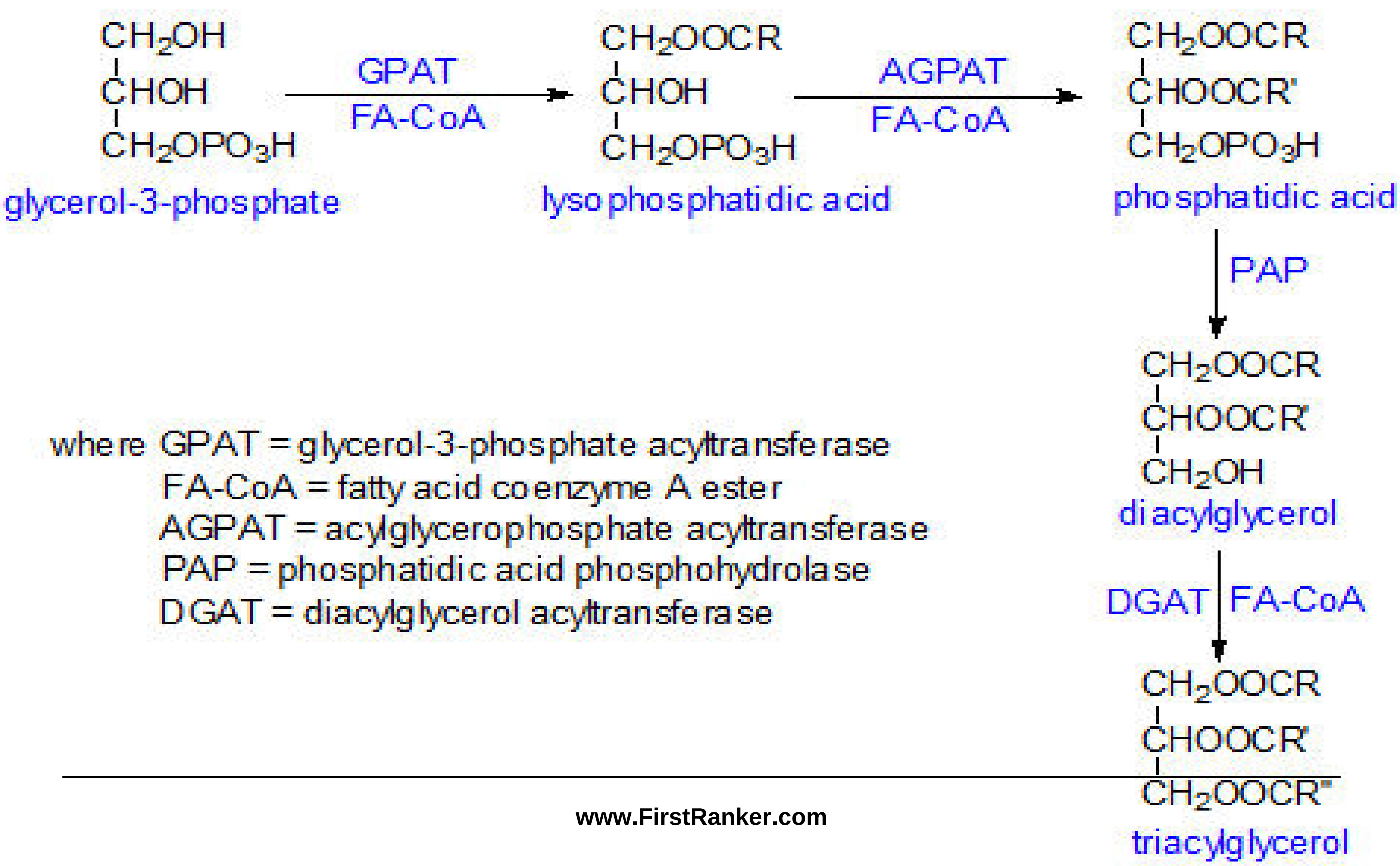
Site For TAG Biosynthesis

- TAG biosynthesis predominantly occurs in **Liver and Adipocytes**

TAG Biosynthesis
Takes Place In
Smooth Endoplasmic Reticulum

- **TAG biosynthesis takes place after De Novo Biosynthesis of Fatty acids.**
- **Fatty acids** and **Glycerol** are **activated** before TAG biosynthesis.
- Fatty acids are activated to **Acyl CoA by Thiokinase**
- Glycerol is activated to **Glycerol-3-Phosphate by Glycerol Kinase.**

Phospholipids are Common Intermediates of TAG ,Phospholipids and Glycolipid Biosynthesis

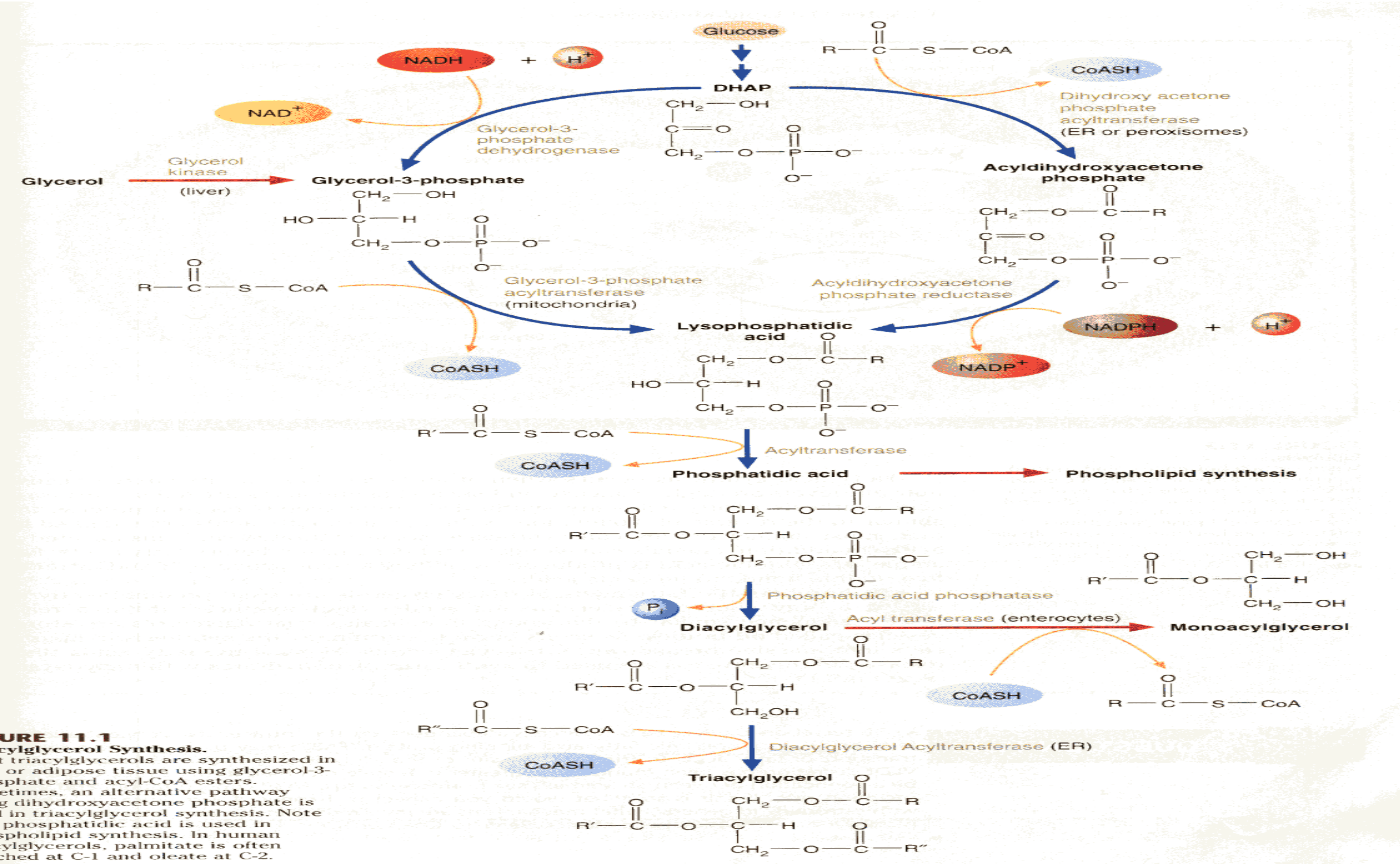


where GPAT = glycerol-3-phosphate acyltransferase
FA-CoA = fatty acid coenzyme A ester
AGPAT = acylglycerophosphate acyltransferase
PAP = phosphatidic acid phosphohydrolase
DGAT = diacylglycerol acyltransferase

- An Acyl chain is transferred to Glycerol by **Acyl Transferase** producing **Lysophosphatidic acid.**
- Lysophosphatidic acid is transformed to **Phosphatidic acid** on addition of one more Acyl chain.

- Phosphate group is removed from Phosphatidic acid to **generate Diacylglycerol**.
- The **addition of third Acyl chain to Diacylglycerol** finally **results in Triacylglycerol**.
- Usually a **mixed type of TAG** is synthesized in the body.

Triacylglycerol Synthesis

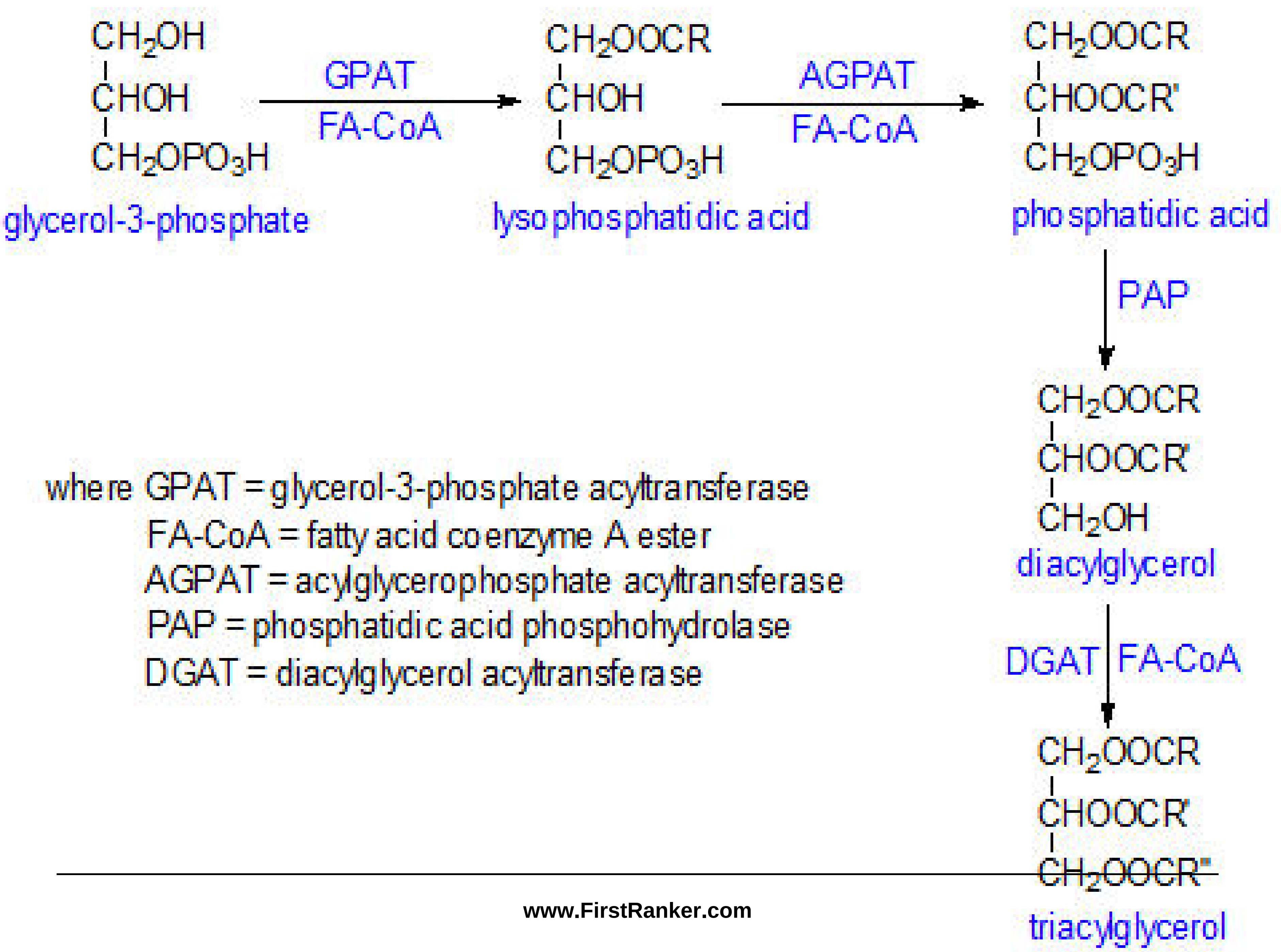


URE 11.1
cylglycerol Synthesis.
t triacylglycerols are synthesized in
or adipose tissue using glycerol-3-
sphate and acyl-CoA esters.
etimes, an alternative pathway
g dihydroxyacetone phosphate is
l in triacylglycerol synthesis. Note
phosphatidic acid is used in
spholipid synthesis. In human
ylglycerols, palmitate is often
ched at C-1 and oleate at C-2.

Phospholipid Biosynthesis

Glycerophospholipid Synthesis

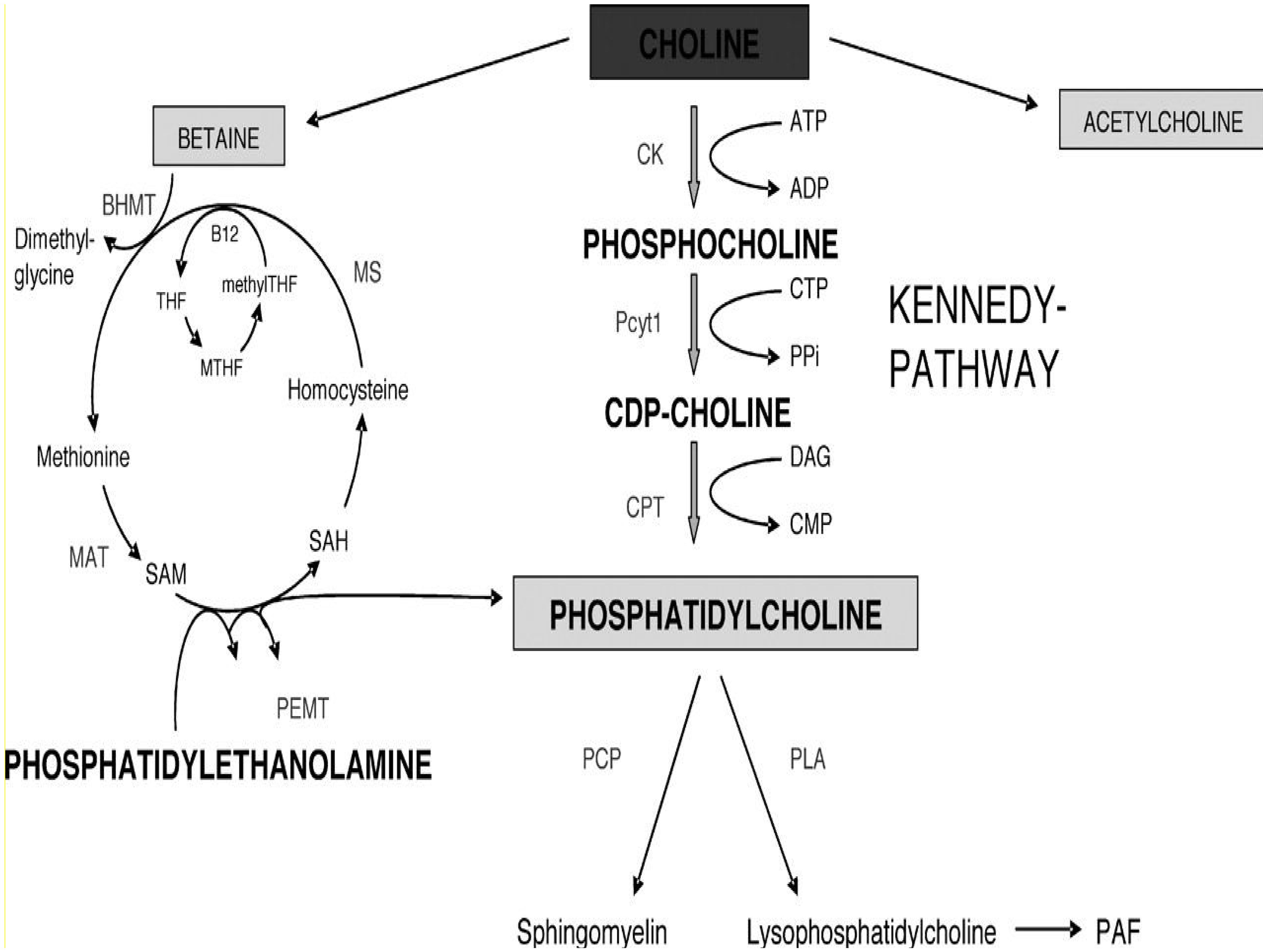
- Glycerophospholipids are biosynthesized **from Phosphatidic acid and Diacylglycerol.**
- These are also intermediates of TAG biosynthesis.



Synthesis OF Lecithin and Cephalin

- Nitrogenous bases **Choline** and **Ethanolamine** are **activated by CTP**
- To form **CDP-Choline and CDP-Ethanolamine.**
- These then **added to Phosphatidic acid** to **form Lecithin and Cephalin respectively.**

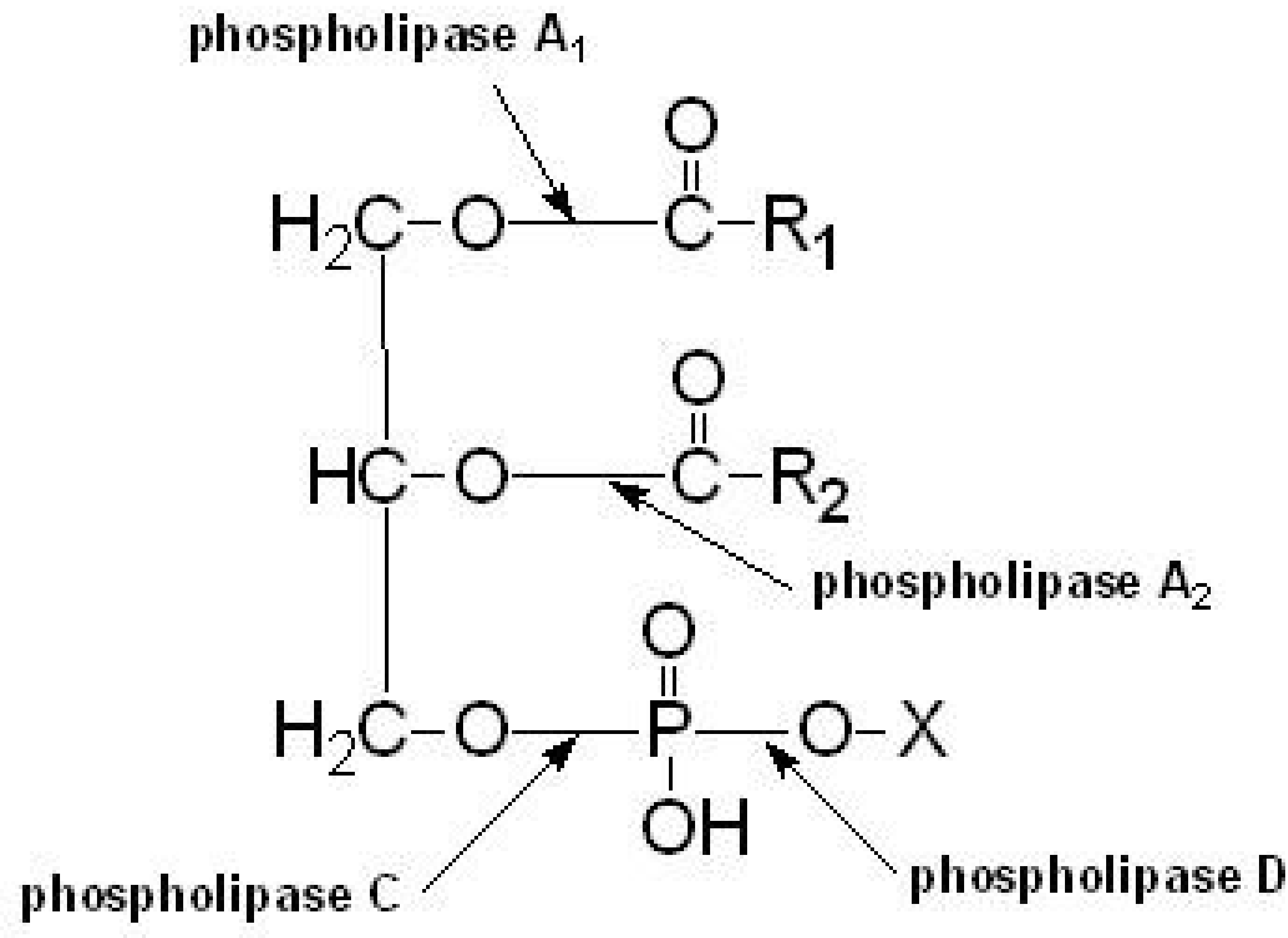
- **Addition of Serine /Inositol** to Phosphatidic acid forms **Phosphatidyl Serine** and **Phosphatidyl Inositol**



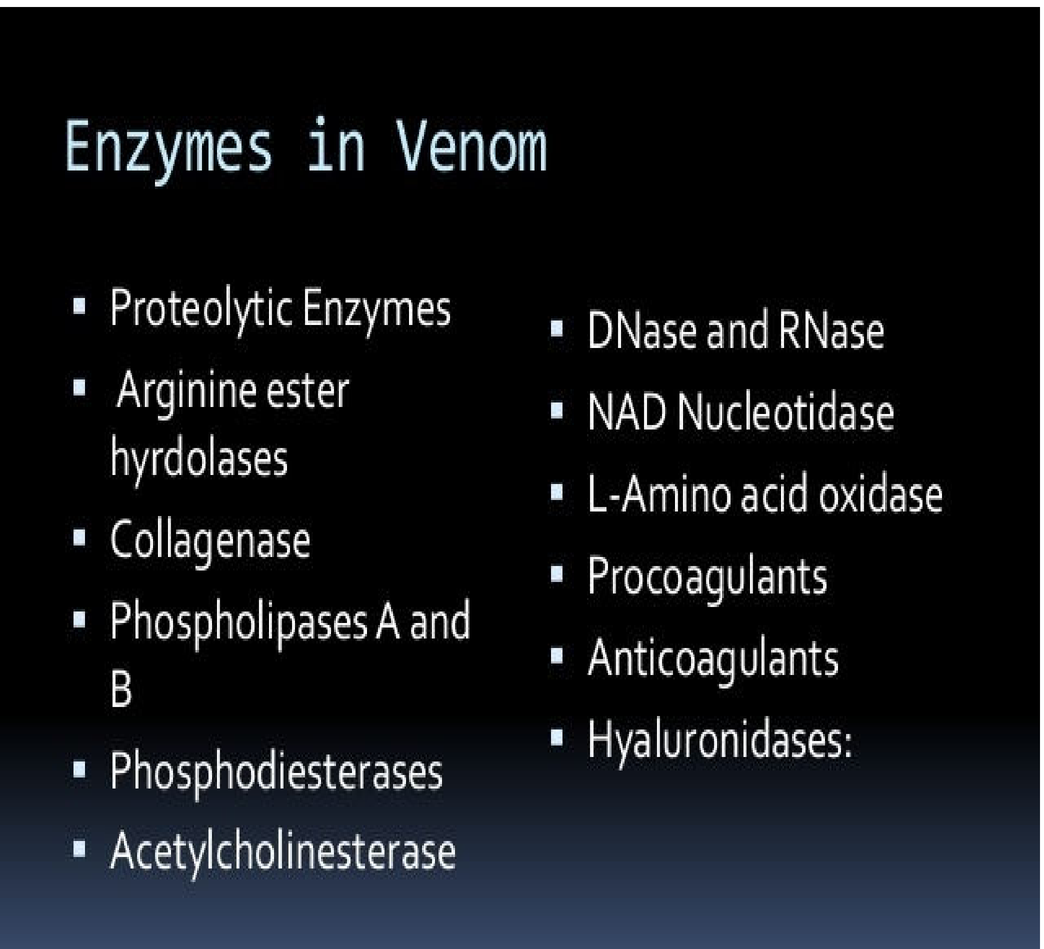
Degradation Of Phospholipids By Phospholipases

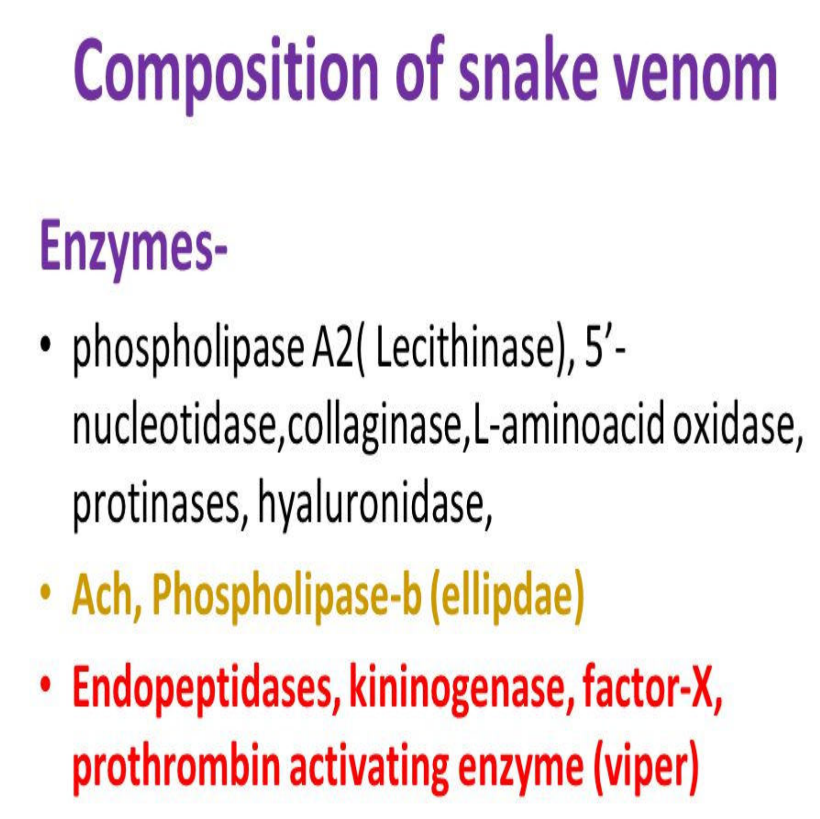
OR

Different Types Of Phospholipases



Phospholipases are Rich In Poisonous Snake Venoms





How and Why Snake Venom Is Toxin?

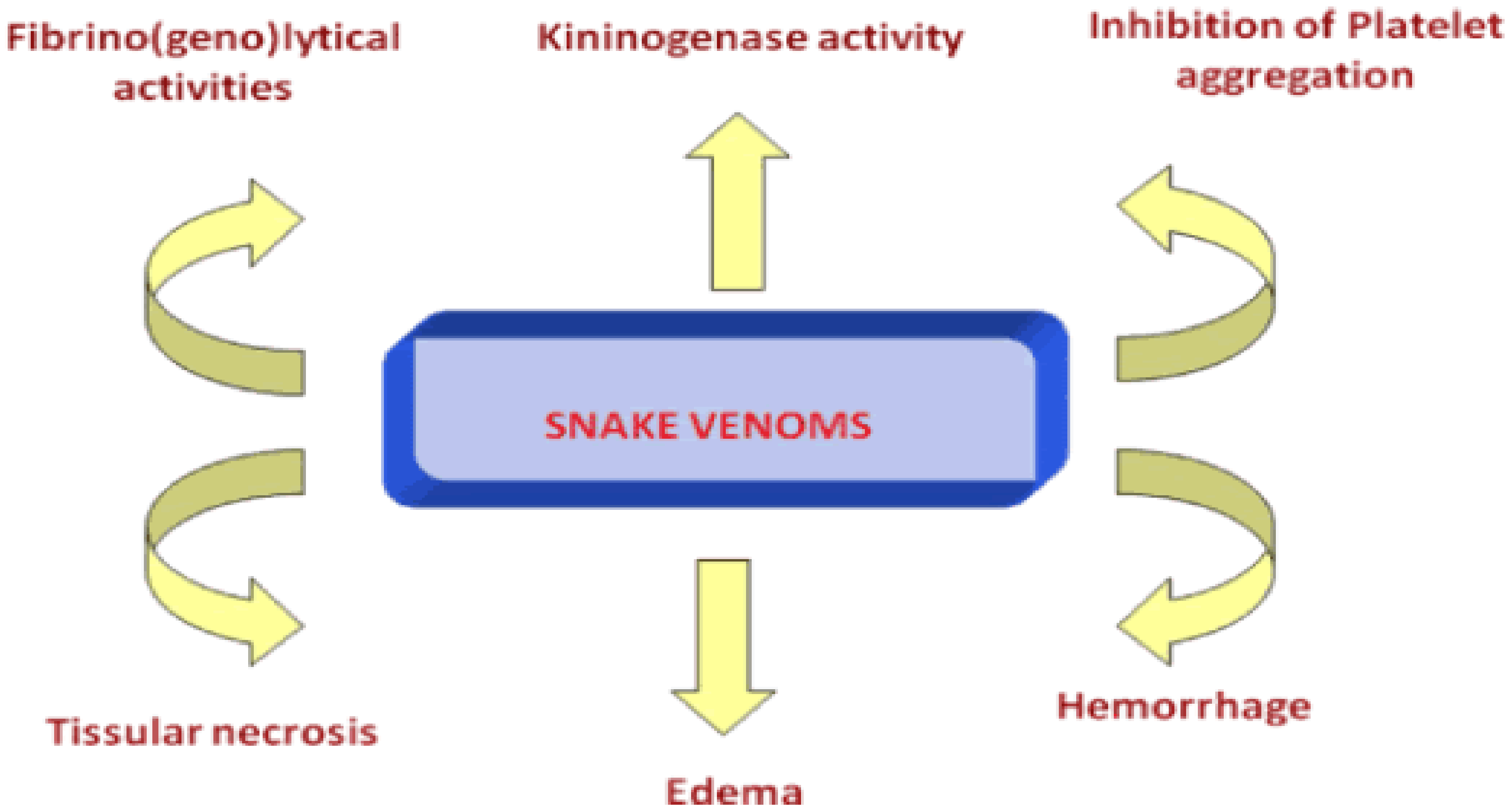


Figure 1: Pathophysiological effects induced by snake venoms.