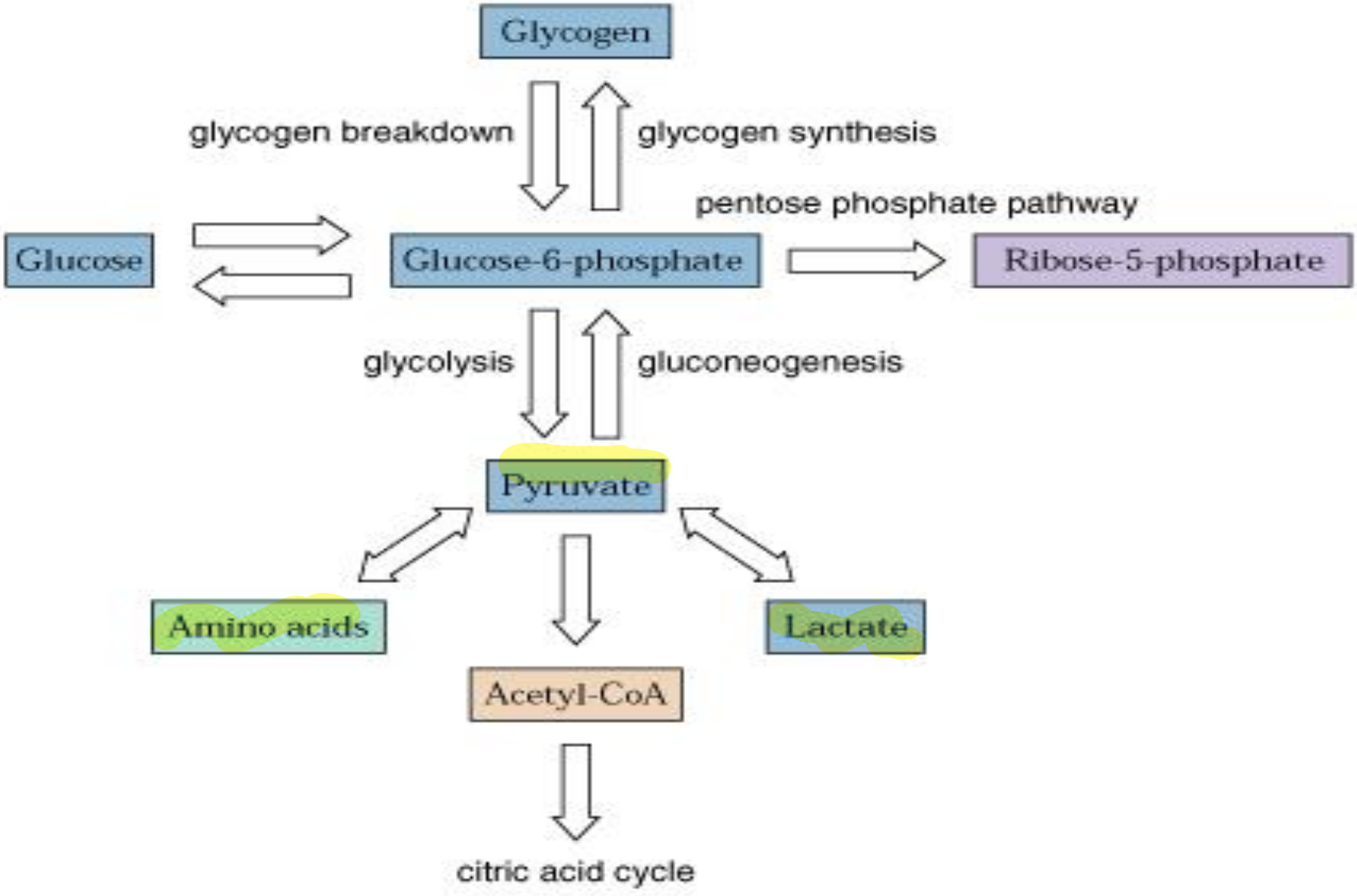
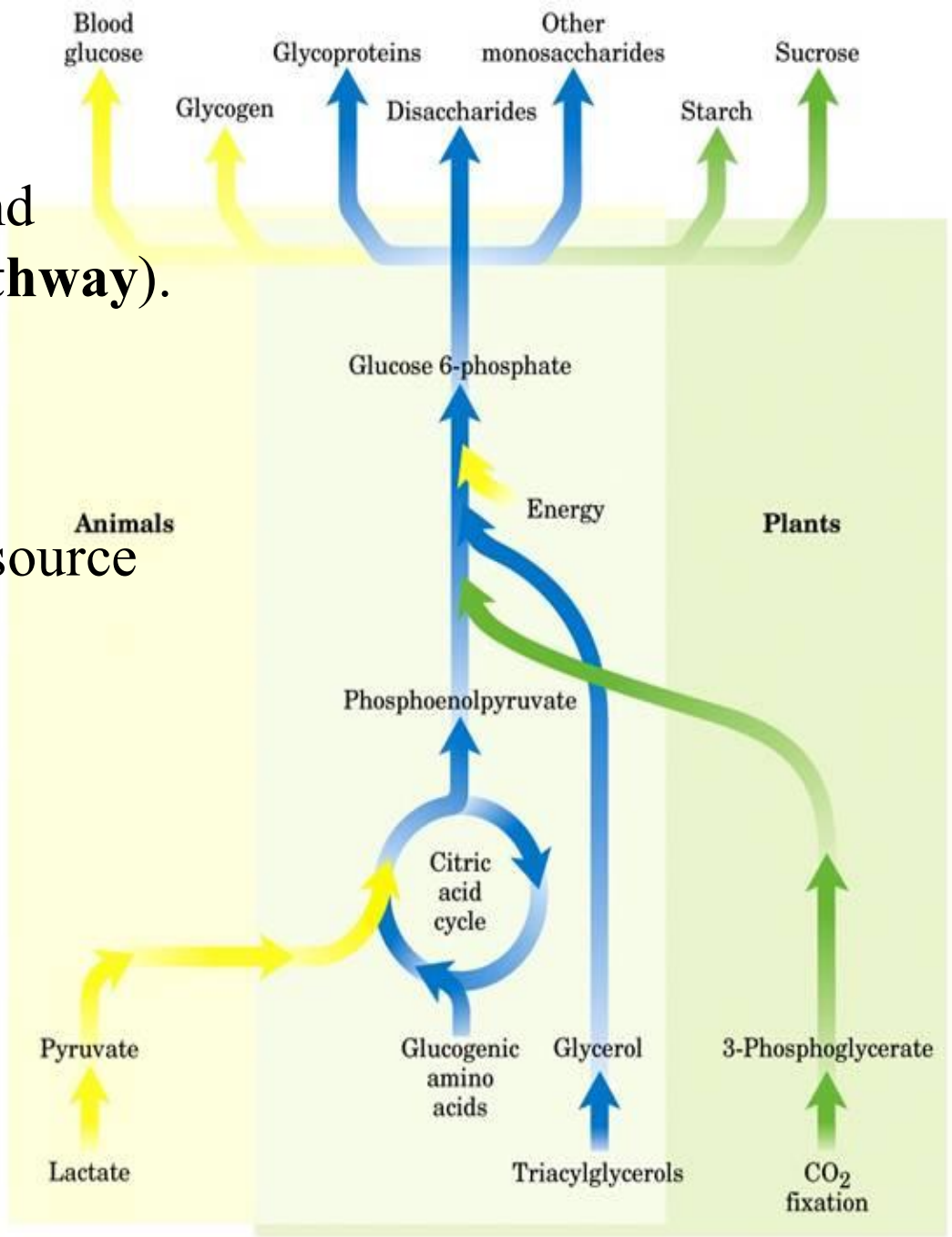
Gluconeogenesis

Overview of Glucose Metabolism



Gluconeogenesis

- It is formation of glucose from non-carbohydrates precursors.
- Occurs in all animals, plants and microorganisms (**universal pathway**).
- Essential in mammals because **nerve cells, testes, medulla** and **RBCs** require glucose from blood as their major fuel source
- Important precursors of the glucose: lactate, pyruvate, glycerol and back bone of certain amino acids (or carbosteleton has no amino groups)
- Fasting requires all the glucose to be synthesized from these non-carbohydrate precursors.



Glyconeogenesis is reversibity of Glycolysis due to there is 3 steps that's irreversible :- 1 3 10

- Gluconeogenesis occurs largely in the liver, kidney, to little extent in renal cortex and intestine under certain condition.

Just - 2 - reaction < Pyruvate → oxaloacetate
oxaloacetate → PEP [phospho enol pyruvate]

- It occurs mostly in the **cytosol**, some reactions in the **mitochondria** and the last step occurs within the **endoplasmic reticulum** cisternae.

- It dose not occur by simple reversal of glycolysis.

- Most precursors must enter the TCA cycle at some point to be converted to oxaloacetate.
- Oxaloacetate is the starting material for gluconeogenesis.

- **Seven** of the of the glycolytic reactions are **reversible** and used in the gluconeogenesis but **three** of them are **irreversible** and should be bypassed by other four reactions.

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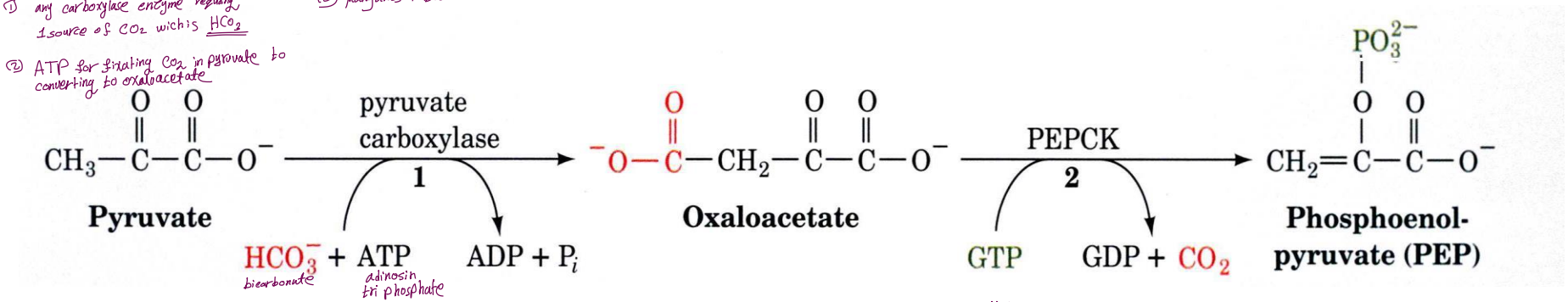
The three steps which should be bypassed in gluconeogenic pathway:

1- Pyruvate to PEP

2- Fructose 1,6- bisphosphate to fructose-6-phosphate

3- Glucose-6-Phosphate to glucose

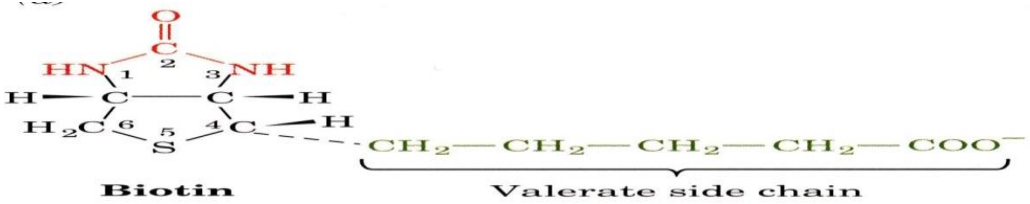
- Conversion of pyruvate to PEP requires two exergonic reactions mediated by the formation of oxaloacetate.

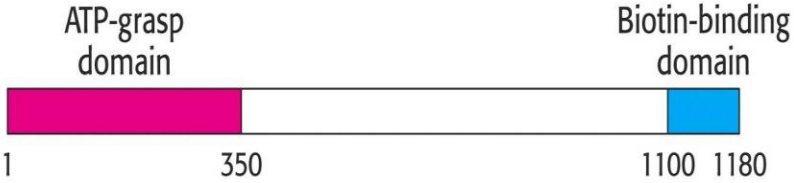


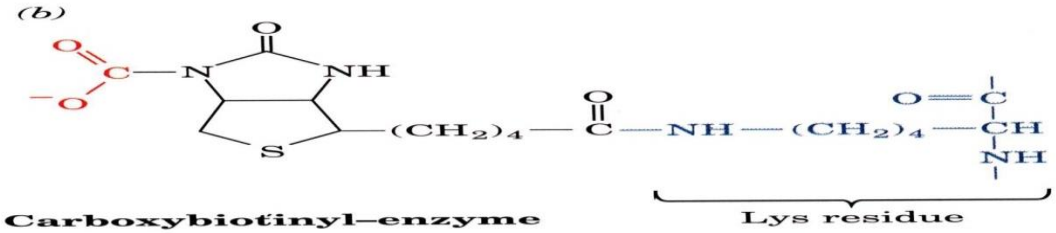
1- Pyruvate carboxylase requiring **biotin** as a cofactor, catalyses the irreversible **ATP-driven** formation of oxaloacetate from pyruvate and CO_2.

This enzyme found in the mitochondria of the **liver and kidney** but not of muscle.

2- PEP carboxykinase converts oxaloacetate to PEP that uses **GTP** as a phosphorylating agent.

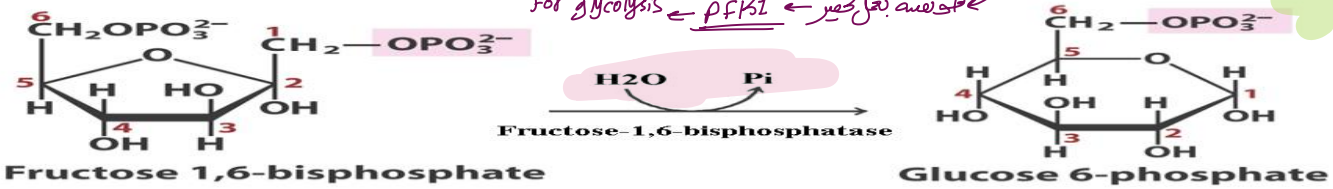






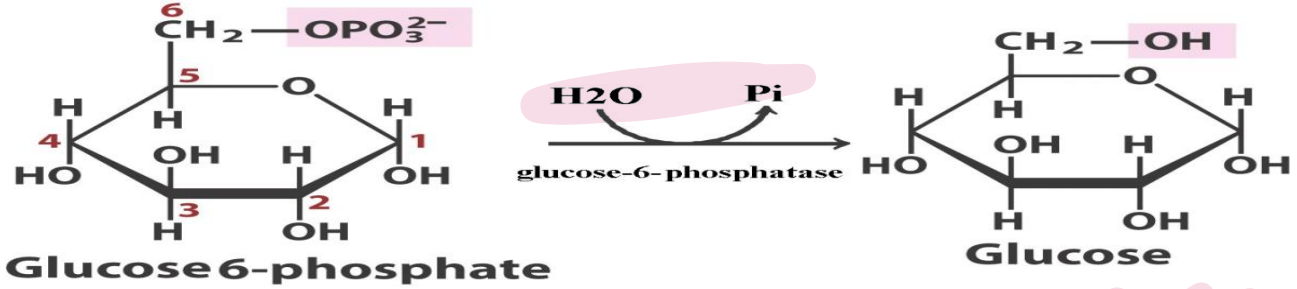
3- Hydrolysis of fructose-1,6-phosphate by fructose1,6-bisphosphatase bypasses the irreversible PFK-1 reaction.

- This reaction is an important **regulatory step** in gluconeogenesis and it occurs only in the **liver and kidney**.
- This enzyme is inhibited by high level of AMP which is a signal of an energy-poor state in the cell, while high ATP stimulates gluconeogenesis.
- It is inhibited also by **fructose 2,6-bisphosphate** which is an **allosteric modulator**, its level is affected by the circulating **glucagon**.

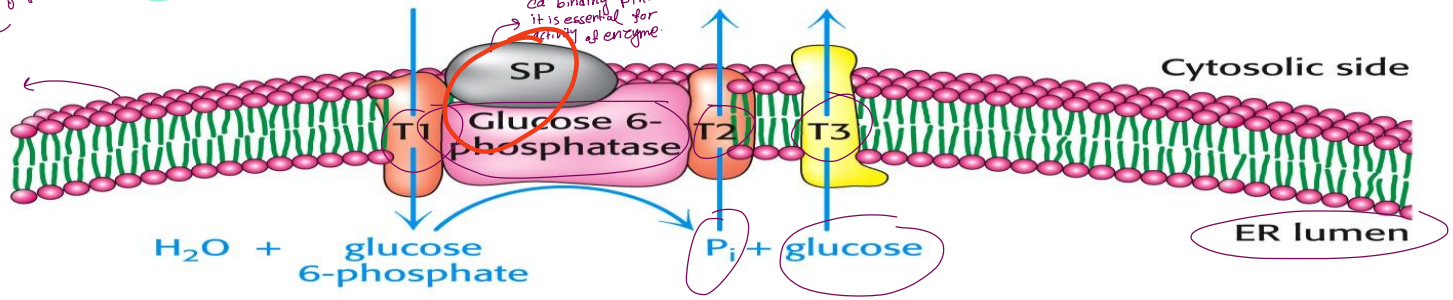


4- Hydrolysis of glucose-6-phosphate by glucose-6-phosphatase bypasses the irreversible hexokinase reaction.

- Glucose-6-phosphatase is only found in the **liver** to buffer blood glucose levels and the **kidney** but not in the **muscle and brain**.



- Ca^{2+}-binding stabilizing protein is essential for phosphatase activity.
- Glucose and P_i are then shuttled back to the cytosol by transporters.



- Generation of glucose from glucose 6-phosphate is involving several proteins: SP – Ca-binding protein.
  T1 transports G-6-P into the lumen of the ER
  T2 and T3 transport Pi and glucose, respectively back into the cytosol.

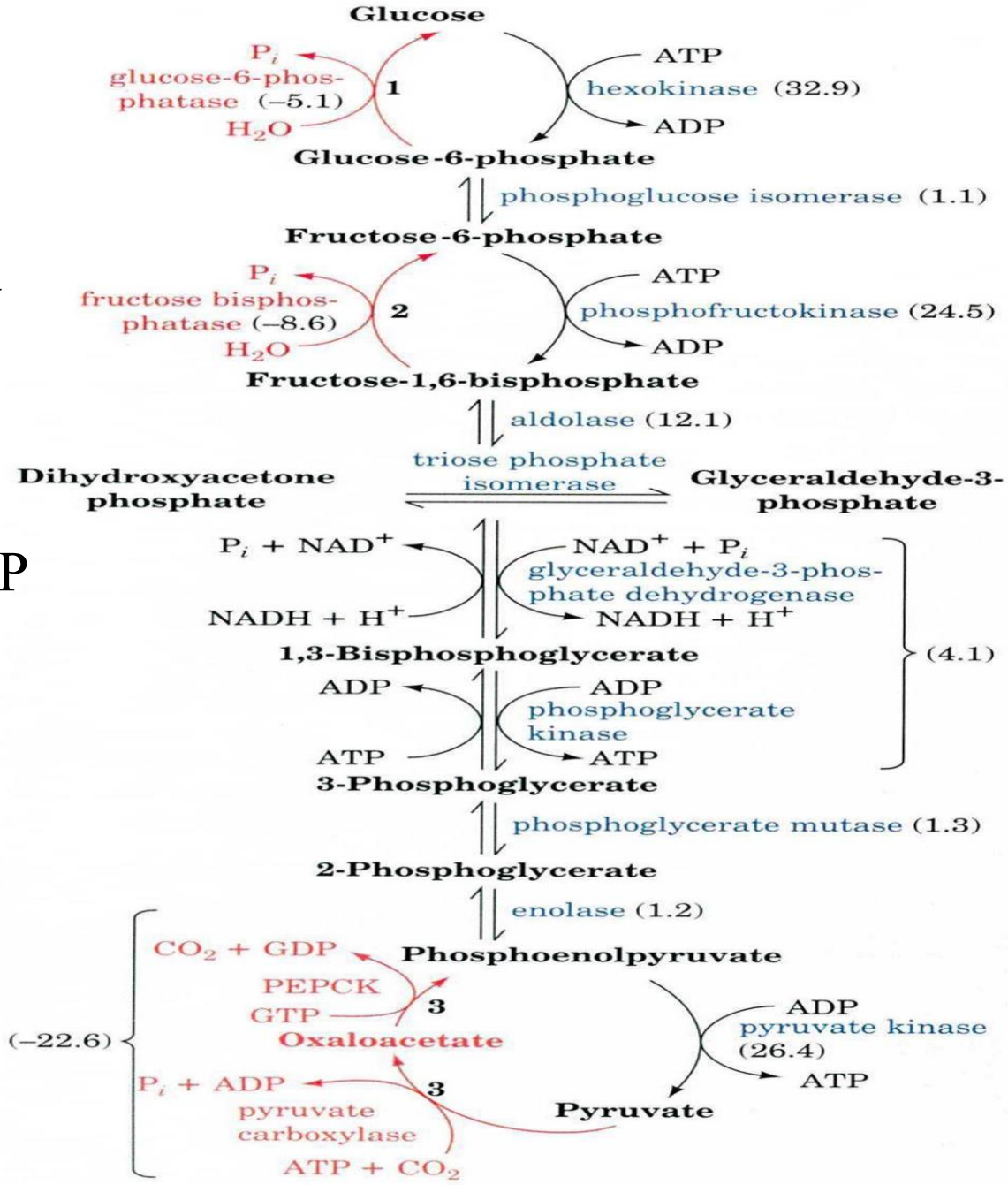
Gluconeogenesis

-The overall reactions of gluconeogenesis are:

2 Pyruvate +2 NAD + 4 H^+ + 4 ATP + 2 GTP + 6 H_2O

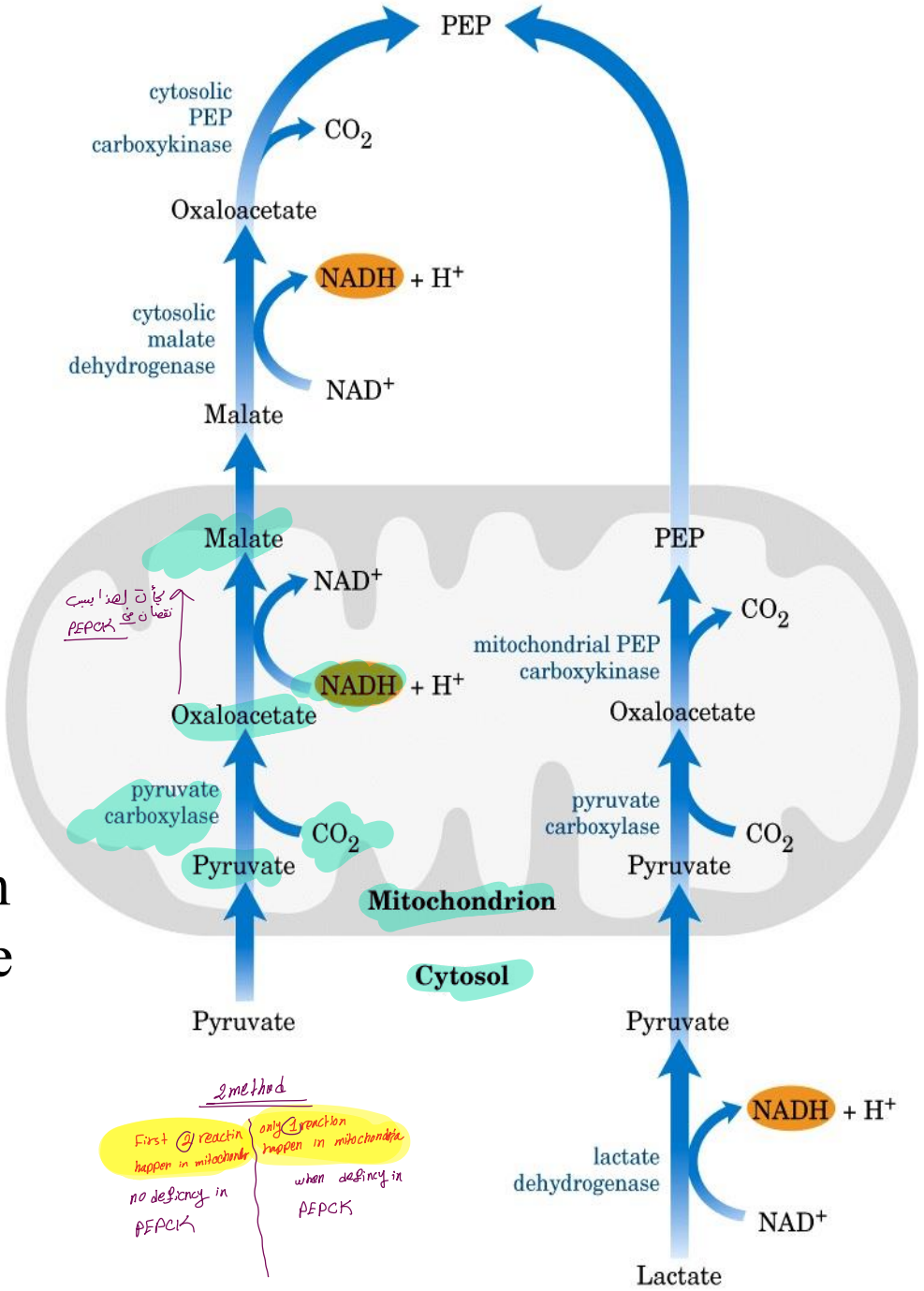
↓

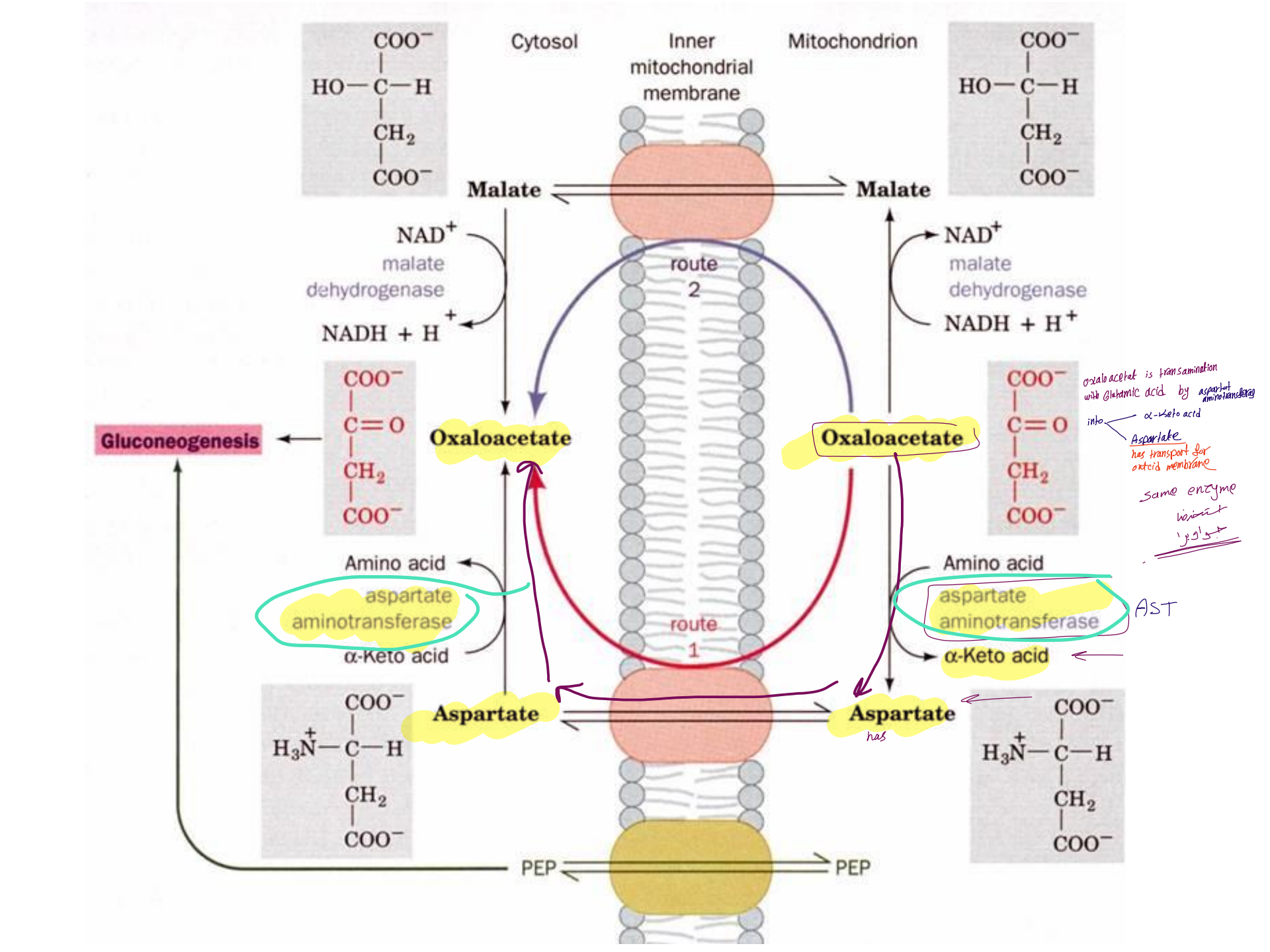
Glucose + 2 NAD^+ + 4 ADP + 2 GDP + 4 Pi



Transport between the mitochondria and the cytosol

- Generation of oxaloacetate occurs in the mitochondria only, but, gluconeogenesis occurs in the cytosol.
- PEPCK is distributed between both compartments in humans,
- Either PEP must be transported across the membranes or oxaloacetate has to be transported.
- PEP transport systems are seen in the mitochondria but oxaloacetate can not be transported directly in or out of the mitochondria.
- It can transported out of the mitochondria in form of Malate




Cytosol
Inner mitochondrial membrane
Mitochondrion
Malate
Malate
NAD+
malate dehydrogenase
NADH + H+
NAD+
malate dehydrogenase
NADH + H+
route 2
Gluconeogenesis
Oxaloacetate
Oxaloacetate
Amino acid
aspartate aminotransferase
α-Keto acid
Amino acid
aspartate aminotransferase
α-Keto acid
route 1
Aspartate
Aspartate
PEP
PEP
oxaloacetat is transamination with Glutamic acid by aspartat aminotransferase
into α-keto acid
Aspartate has transport for antcid membrane
same enzyme
AST

Regulation of gluconeogenesis

- To prevent the waste of a futile cycle, glycolysis and gluconeogenesis are reciprocally regulated.
- F-1,6-bisphosphatase is the most important control site in gluconeogenesis.

Reciprocal regulation by ATP/AMP

- AMP inhibits fructose-1,6-bisphosphatase but activates PFK-1
- ATP and citrate inhibit PFK-1 but activate fructose-1,6-biphosphatase
  - In high ATP/AMP ratio: stimulate gluconeogenesis
  - In low ATP/AMP ratio: stimulate glycolysis
- High levels of **ATP and alanine**, which signal that the energy charge is high and that building blocks are abundant, inhibit **pyruvate kinase**.
- **Pyruvate carboxylase** is activated by **acetyl CoA**.
- **ADP** inhibits **PEP carboxykinase** and **pyruvate carboxylase**.

- Gluconeogenesis is favored when the cell is rich in biosynthetic precursors and ATP.

Reciprocal regulation by fructose-2,6-biphosphate:

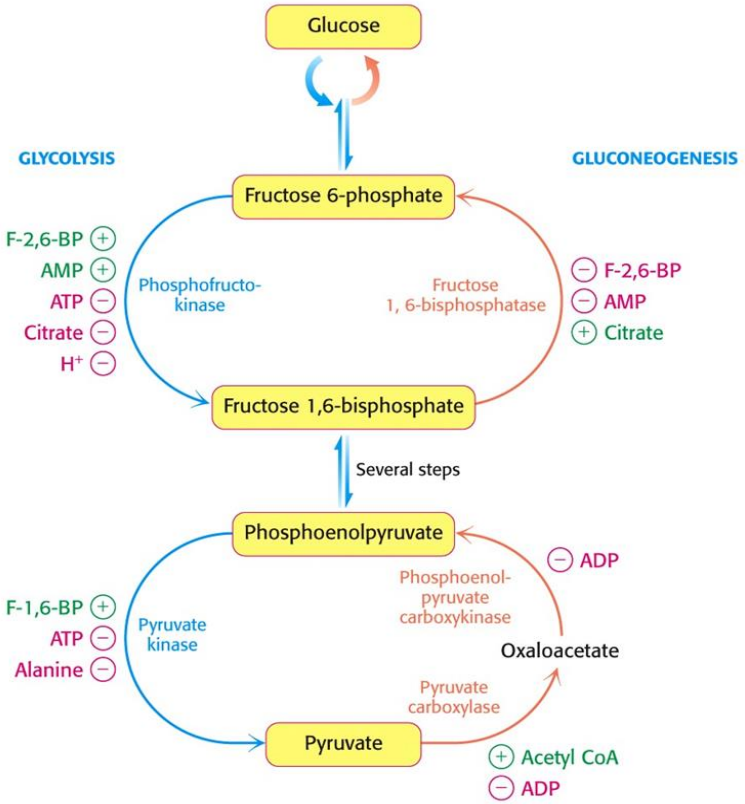
- Fructose-2,6-biphosphate stimulates glycolysis by activating PFK-1 and inhibits gluconeogenesis through the inhibition of fructose-1,6-biphosphatase.
- During starvation, gluconeogenesis predominates because the level of F-2,6-BP is very low.

Reciprocal regulation by hormones

PFK-1: induced in feeding by insulin and repressed in starvation by glucagon

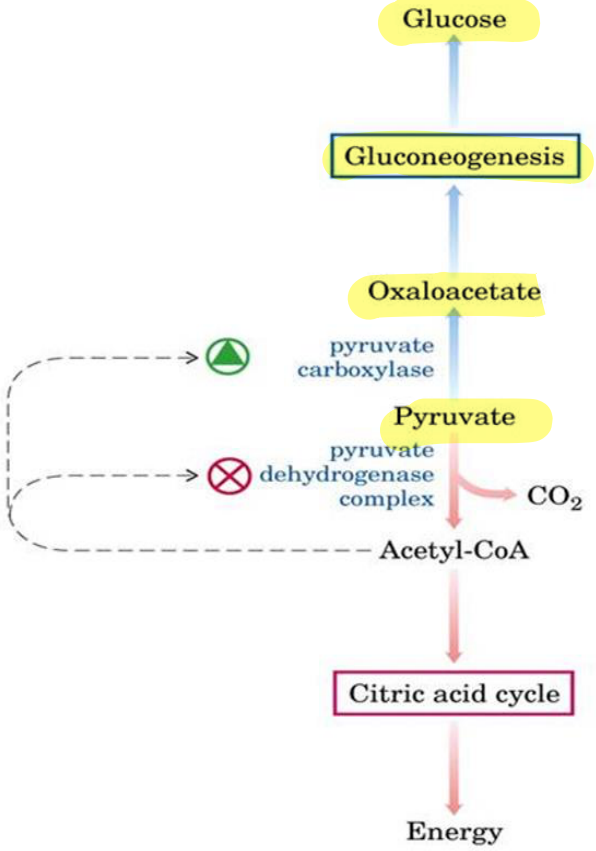
Fructose-1,6-bisphosphase: repressed in feeding by insulin and induced in starvation by glucagon

So: Insulin activates glycolysis but inhibits gluconeogenesis; Glucagon activates gluconeogenesis but inhibits glycolysis.



Acetyl-CoA regulates pyruvate carboxylase

- The increase in oxaloacetate concentration → the activity of the TCA cycle.
- Acetyl-CoA is an allosteric activator of pyruvate carboxylase.
- At low levels of acetyl-CoA, pyruvate carboxylase is largely inactive and pyruvate is oxidized in TCA cycle.
- However, when ATP and NADH concentrations increased, oxaloacetate goes to glucose.

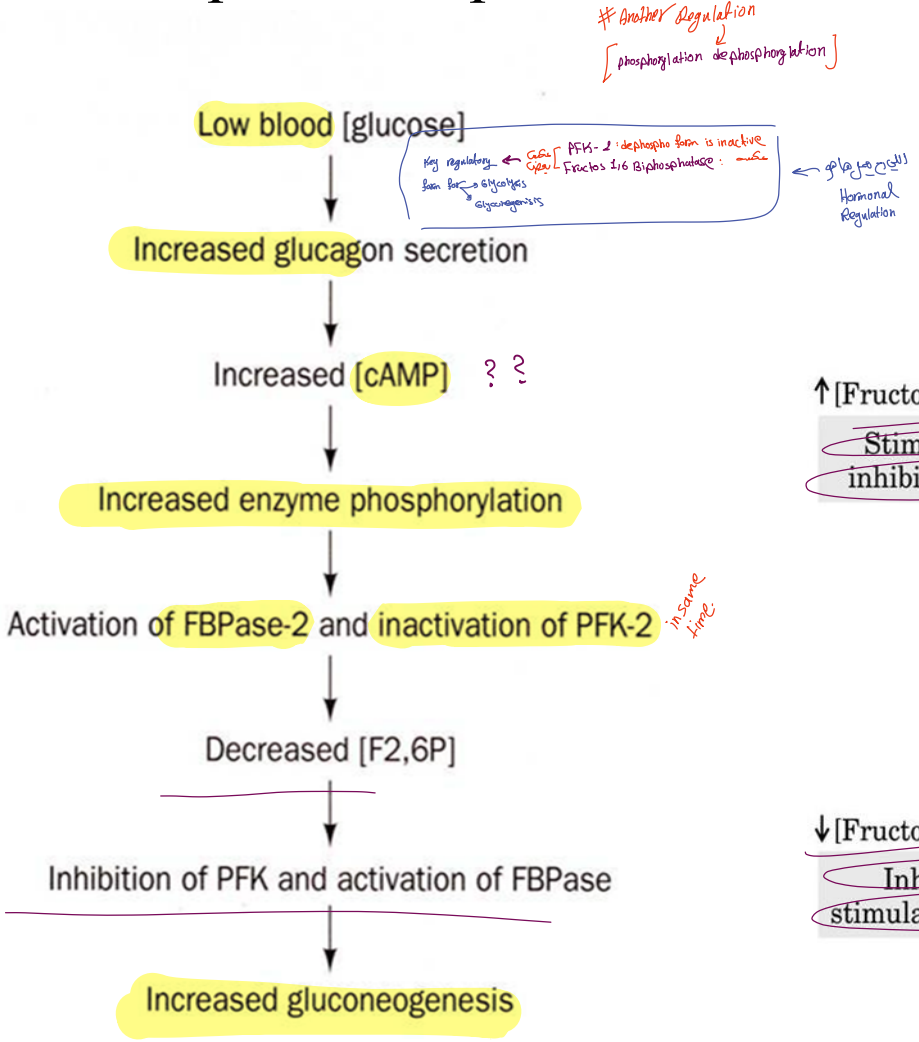


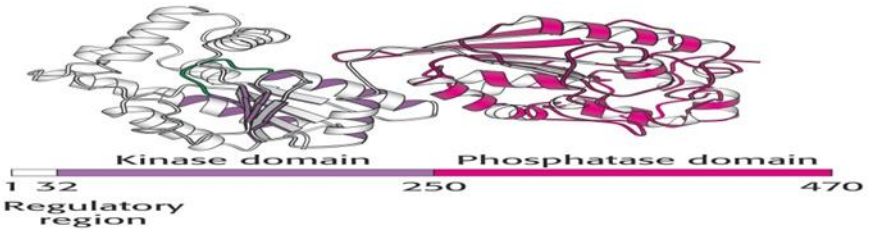
Allosteric activation by acetyl CoA

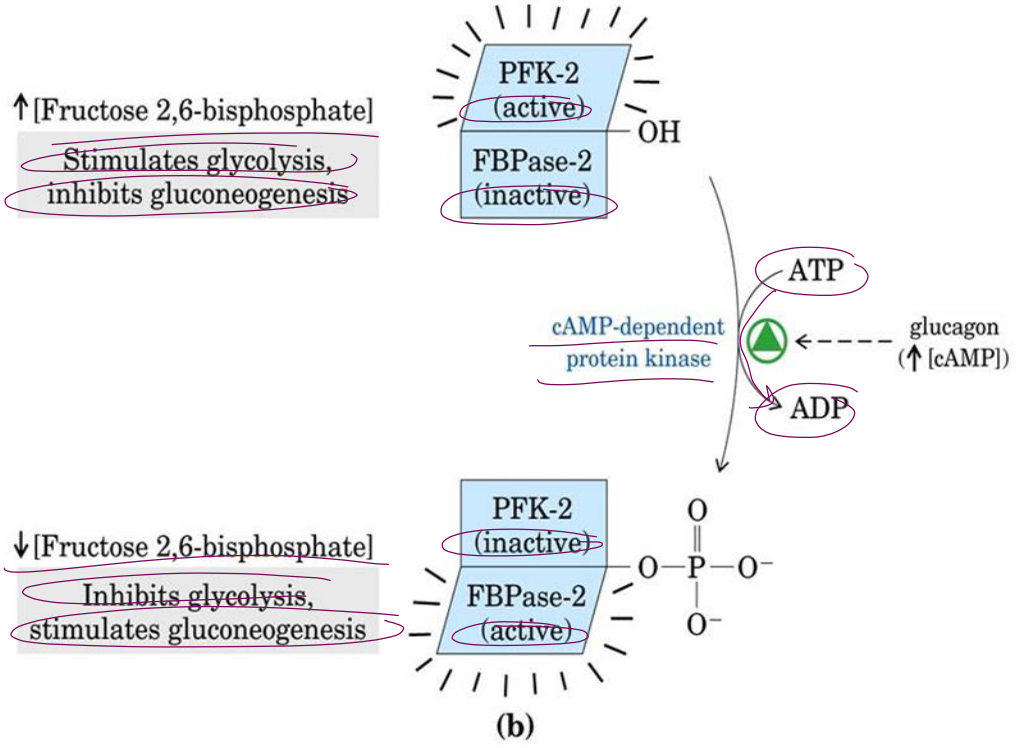
- During starvation → excessive lipolysis → excessive oxidation of fatty acid into acetyl CoA → accumulation of acetyl CoA → activation of pyruvate carboxylase → activation of gluconeogenesis.

Substrate availability:

- The availability of gluconeogenic precursors like glucogenic amino acids → ↑ the hepatic gluconeogenesis.
- ↓ Insulin / glucagon ratio favor the mobilization of amino acids from muscle protein to provide their skeletons for gluconeogenesis.







PEP carboxykinase ↑

F 1,6-bisphosphatase ↑

Glycolytic enzymes ↓ (pyruvate kinase)

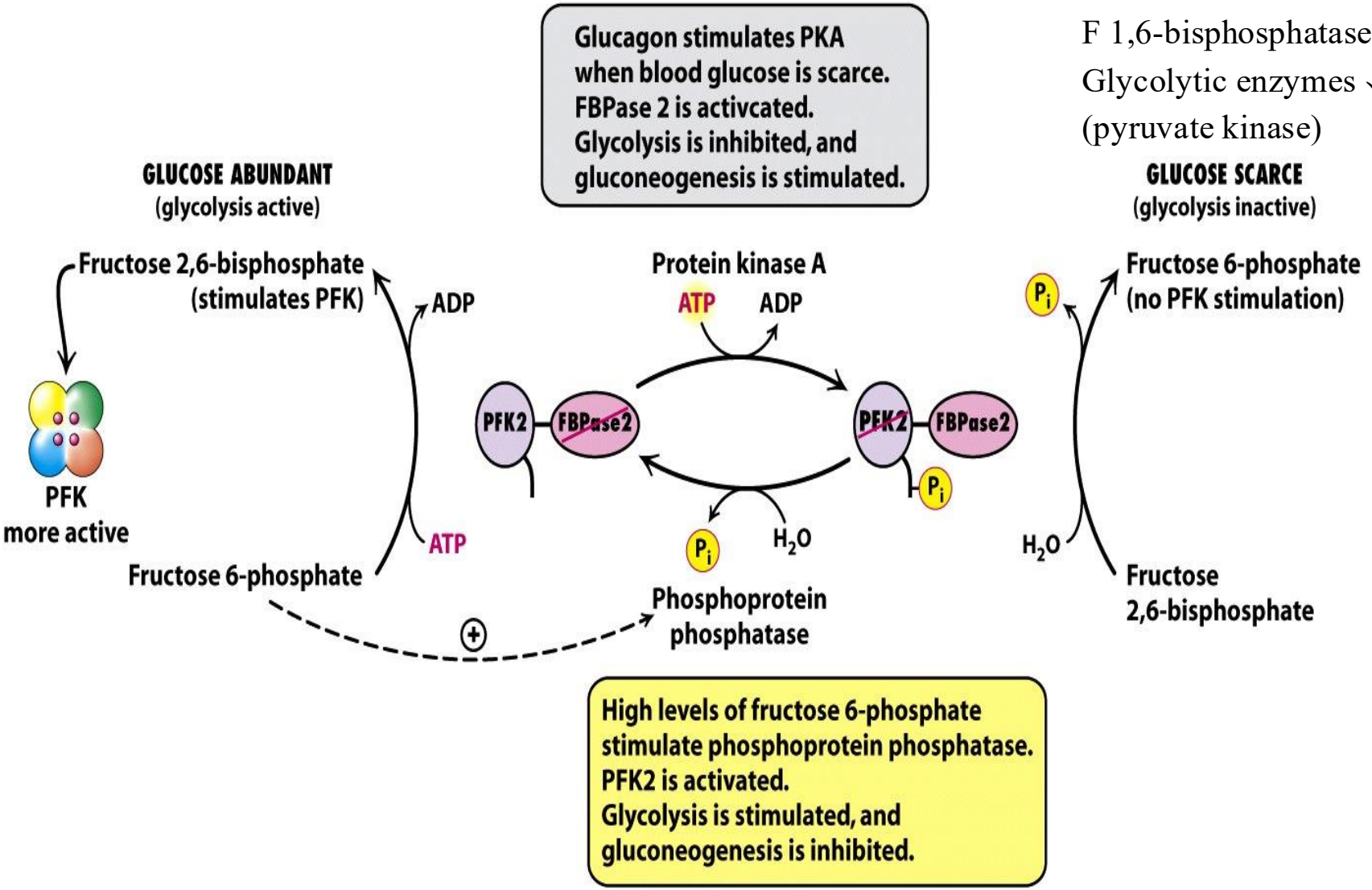
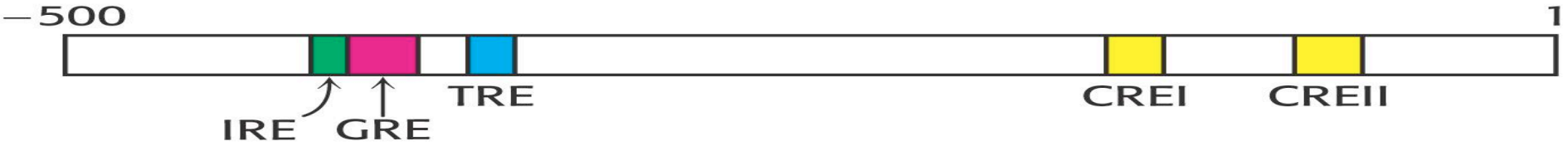


Figure 16-30
Biochemistry, Sixth Edition

Hormones

- Affect the expression of the gene of the essential enzymes
  - Change the rate of transcription
  - Regulate the degradation of mRNA
- Phosphorylation control (~ s); allosteric control (~ms); transcription control (~ h to d)



The promoter of the PEP carboxykinase (OAA→PEP) gene:

IRE: insulin response element;

GRE: glucocorticoid response element

TRE: thyroid response element

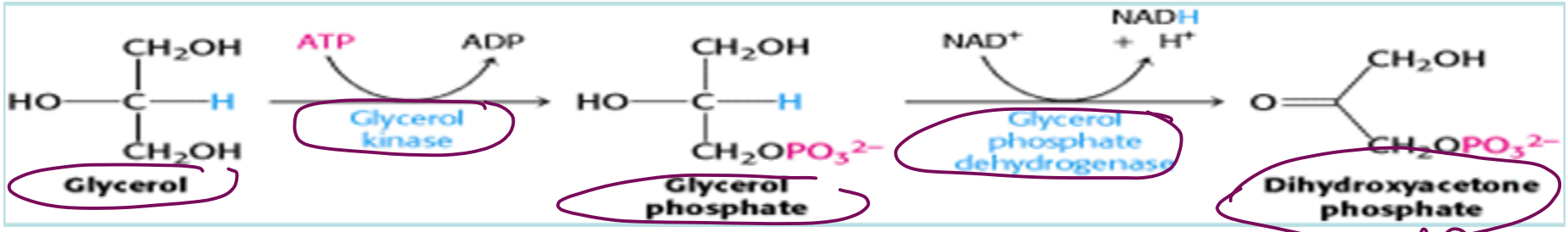
CREI and II: cAMP response elements

any intermediat of Glycolysis / TCA is substrate for Glyconeogenesis.

Substrates for gluconeogenesis

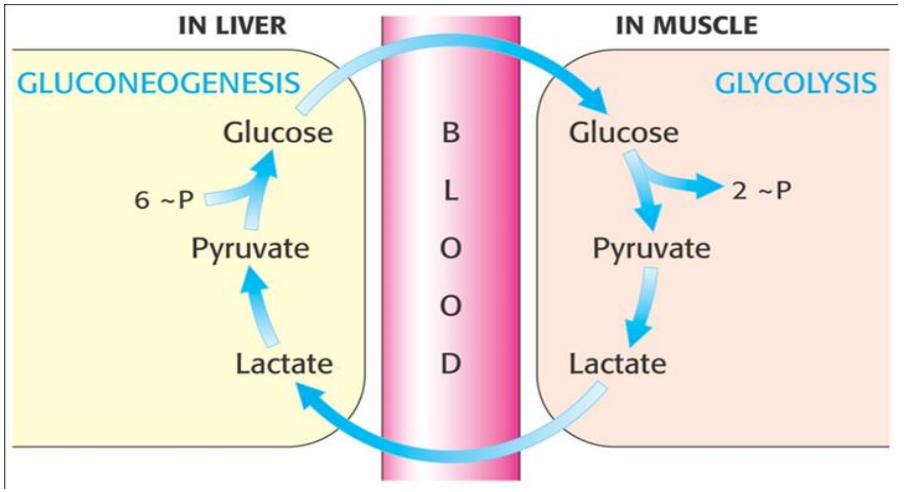
-Include all intermediates of glycolysis and TCA cycle, glycerol, lactate and the α-keto acids obtained from deamination of glucogenic A.A.s.

- **Glycerol**: obtained from the hydrolysis of the triglycerides in adipose tissue, travels to liver which is phosphorylated and metabolized.



- DHAP is converted into glyceraldehyde 3-P by triose isomerase.

- **Lactate**: released from the RBC and exercising muscle, carried to the liver by the blood and converted to glucose and released again to blood through Cori cycle.

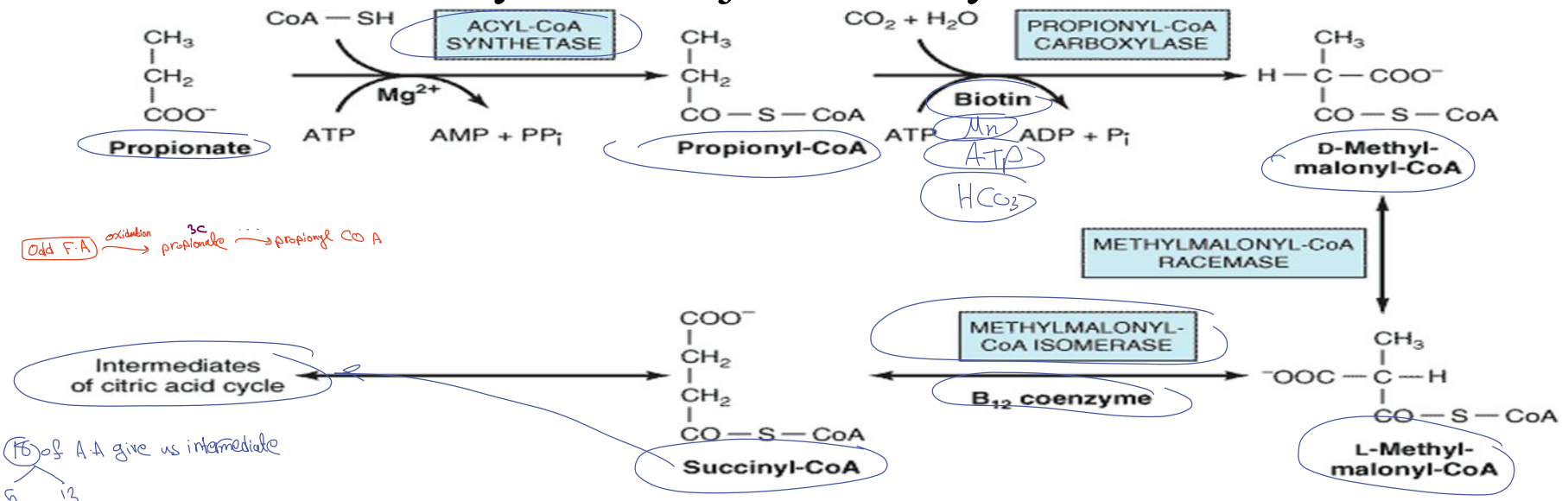


Lactate that produc from Glycolysis need for lactat dehydrogenase enzyme but it will not accumulat so it will go outside to blood then to liver.

organs do Glycolysis → under anaerobic condit: 1. Adrenal cortex & Cornea 2. muscle 3. RBC

Cycle
Lactate → Glucose

- **Odd chain fatty acids**: upon oxidation → propionyl CoA to be converted into succinyl CoA to join TCA cycle.



- **α-keto acids**: like pyruvate and α-ketoglutarate derived from amino acids alanine and glutamate. These substances enter TCA cycle to provide the oxaloacetate.
- All amino acids can feed into gluconeogenesis except leucine and lysine.

Glucogenic Amino Acids, Grouped by Site of Entry*

Pyruvate	**Succinyl-CoA**
Alanine	Isoleucine
Cysteine	Methionine
Glycine	Threonine
Serine	Valine
Tryptophan	
α-Ketoglutarate	**Fumarate**
Arginine	Phenylalanine
Glutamate	Tyrosine
Glutamine	
Histidine	**Oxaloacetate**
Proline	Asparagine
	Aspartate

- Acetyl CoA cannot give rise to a net synthesis of glucose because of the irreversible nature of PDH that converts pyruvate to acetyl CoA.

The Alanine cycle

- The liver can also use the amino acid alanine similarly to lactate

- Following transamination to pyruvate, gluconeogenesis allows the liver to convert it to glucose for secretion into the blood