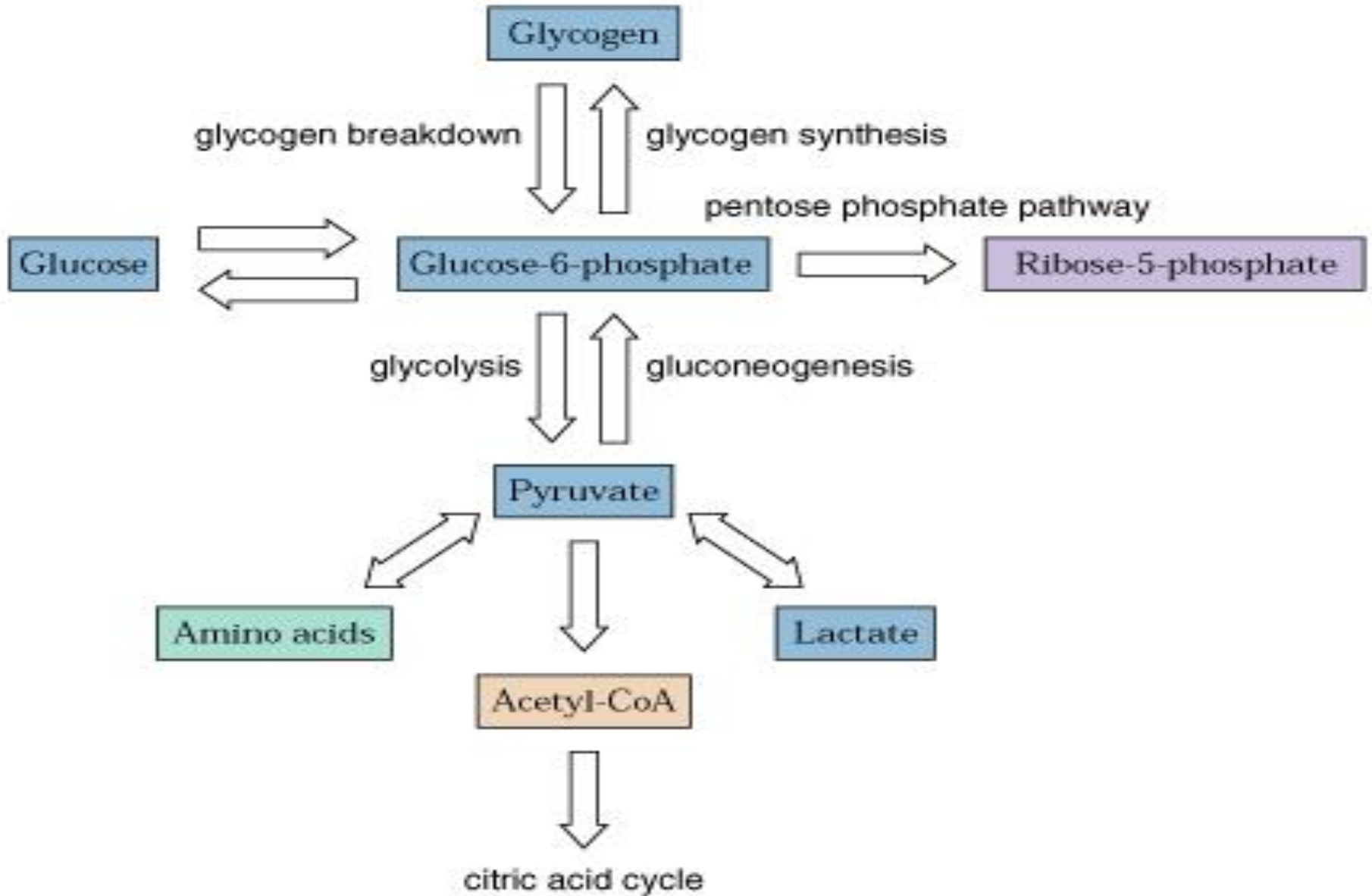


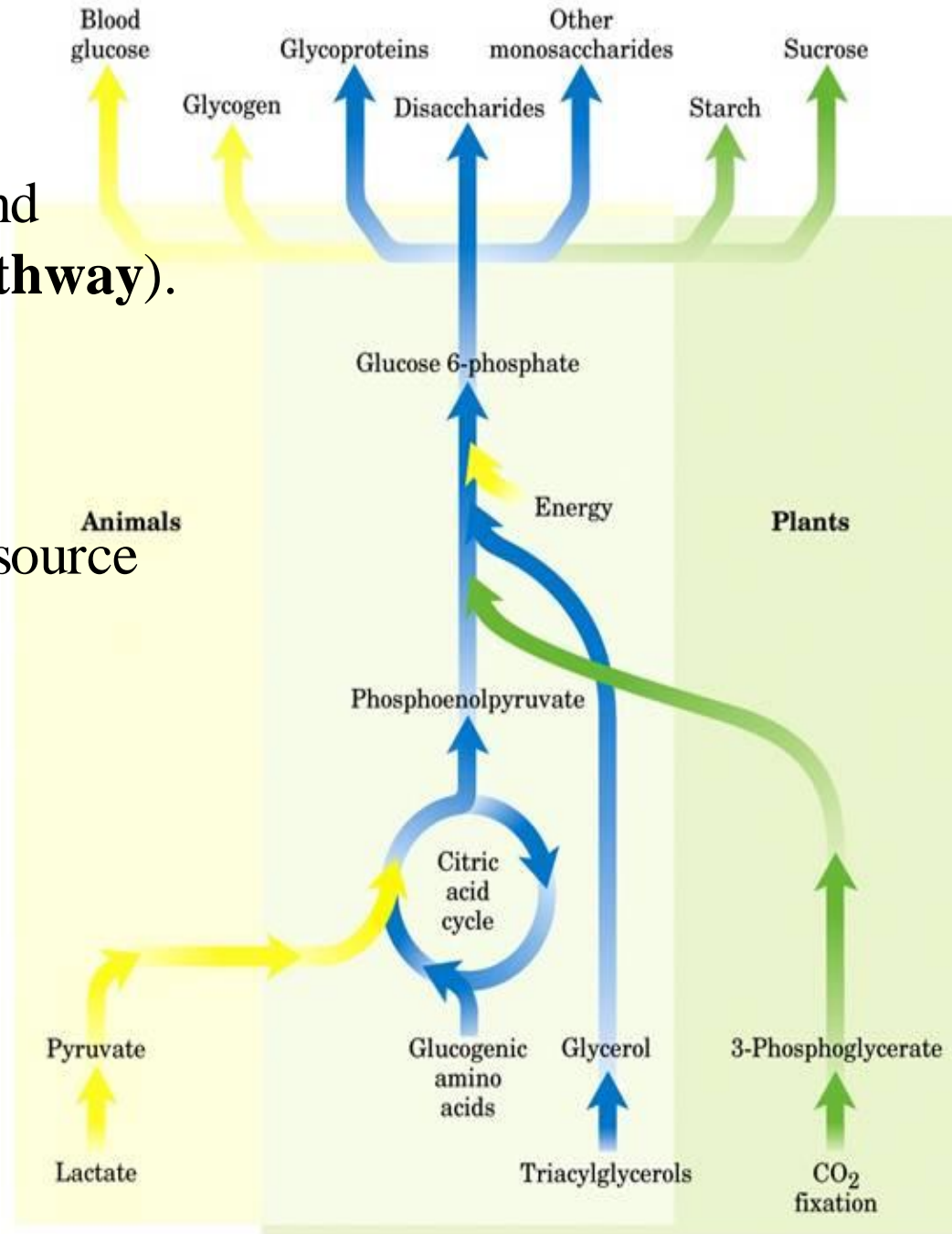
Gluconeogenesis

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
- Fasting requires all the glucose to be synthesized from these non-carbohydrate precursors.



- Gluconeogenesis occurs largely in the liver, kidney, to little extent in renal cortex and intestine under certain condition.
- It occurs mostly in the **cytosol**, some reactions in the **mitochondria** and the last step occurs within the **endoplasmic reticulum** cisternae.
- It does 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 glycolytic reactions are **reversible** and used in the gluconeogenesis but **three** of them are **irreversible** and should be bypassed by other four reactions.

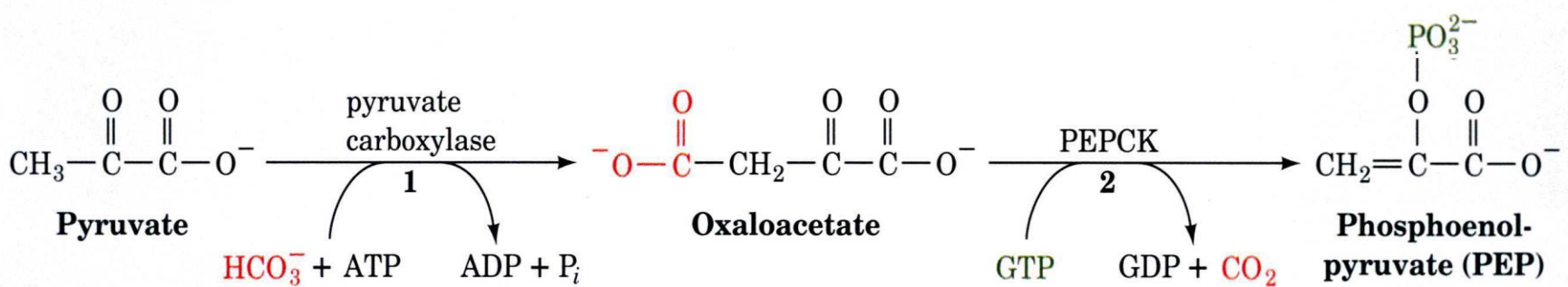
The three steps which should be bypassed in gluconeogenic pathway:

1- Pyruvate to PEP

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

3- Glucose-6-Phosphate to glucose

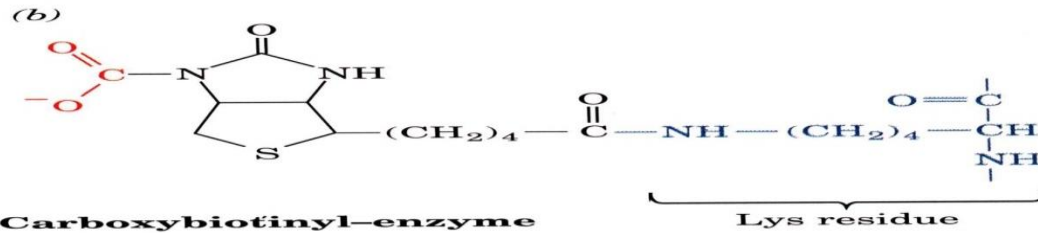
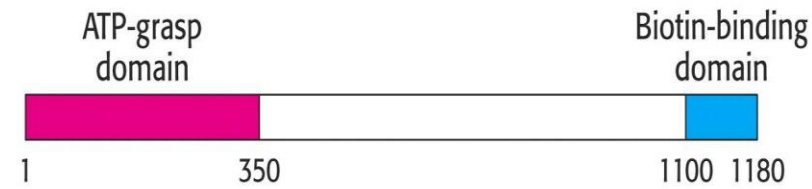
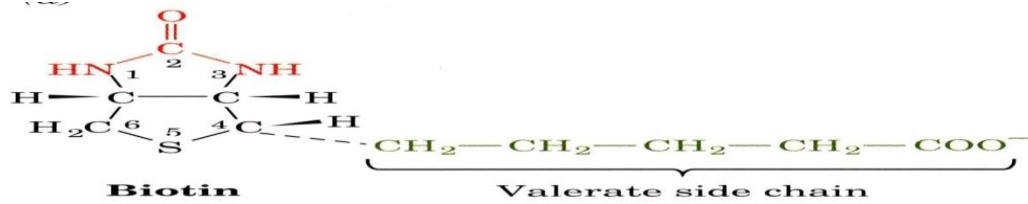
- Conversion of pyruvate to PEP requires two exergonic reactions mediated by the formation of oxaloacetate.



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.

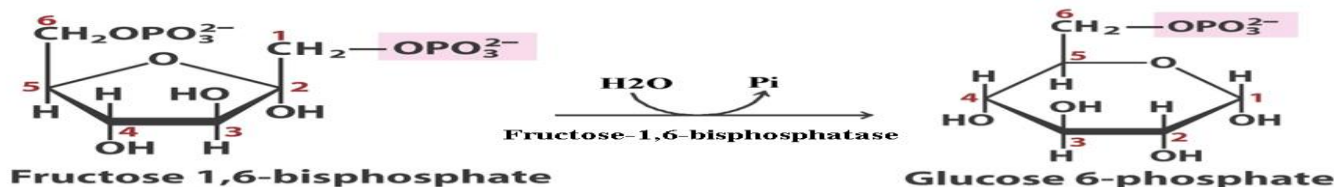


3- Hydrolysis of fructose-1,6-phosphate by fructose 1,6-bisphosphatase bypasses the irreversible PFK-1 reaction.

- This reaction is an important **regulatory step** in gluconeogenesis and it occurs only in the **liver and kidney**.

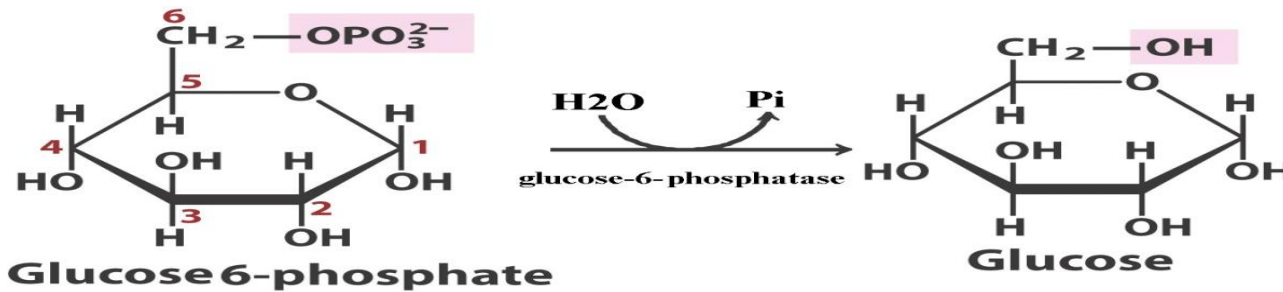
- This enzyme is inhibited by high level of AMP which is a signal of an energy-poor state in the cell, while high ATP stimulates gluconeogenesis.

- It is inhibited also by **fructose 2,6-bisphosphate** which is an **allosteric modulator**, its level is affected by the circulating **glucagon**.

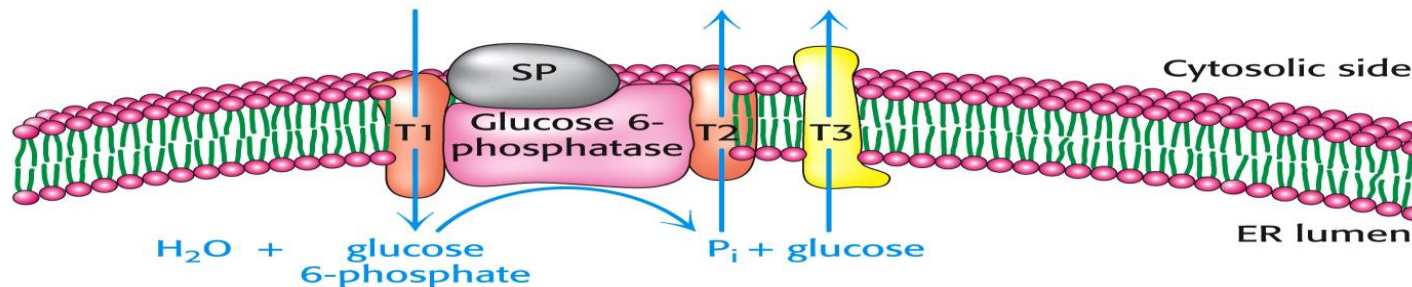


4- Hydrolysis of glucose-6-phosphate by glucose-6-phosphatase bypasses the irreversible hexokinase reaction.

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



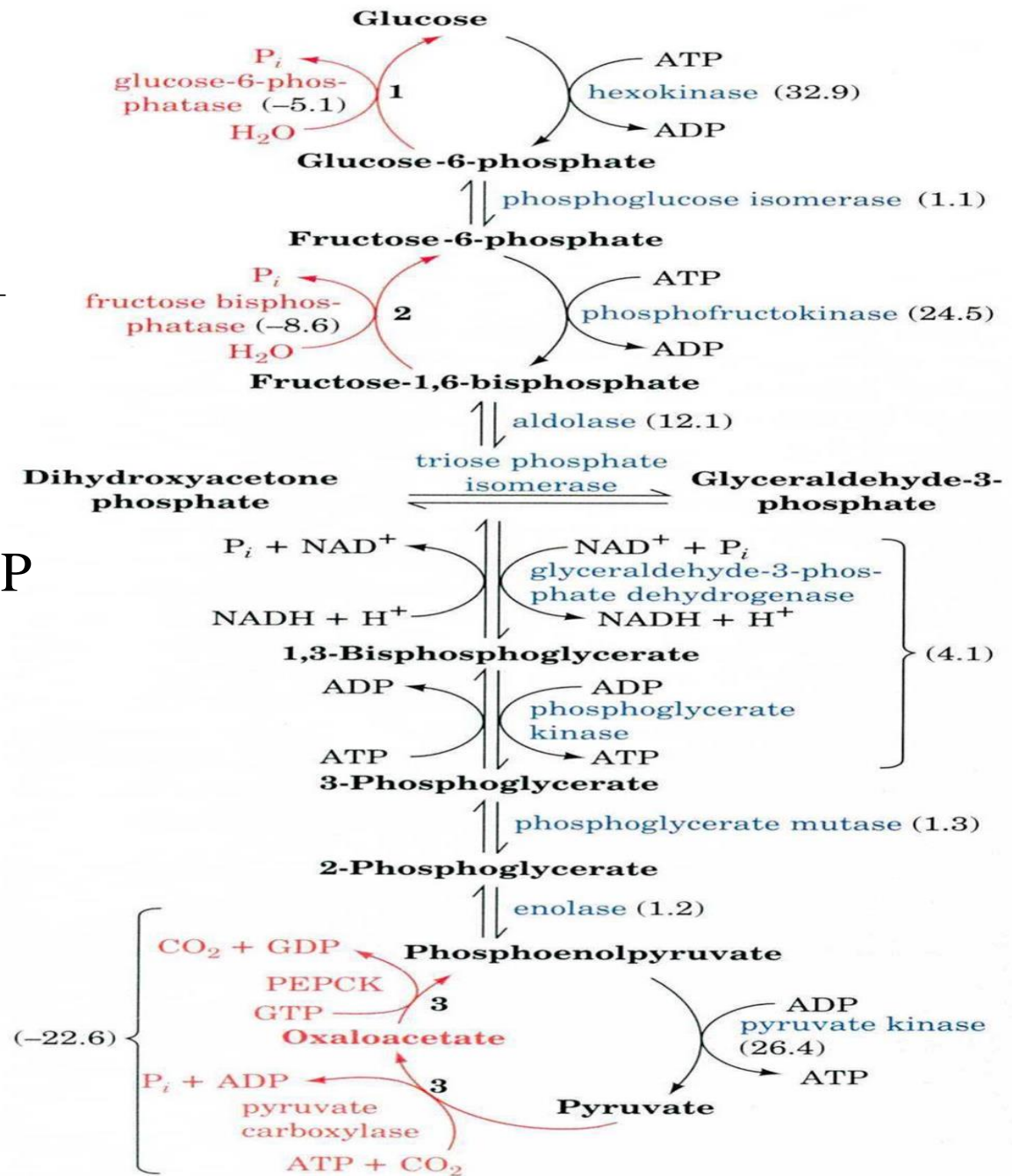
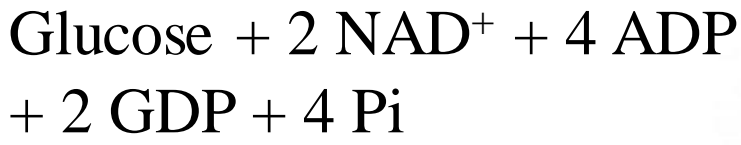
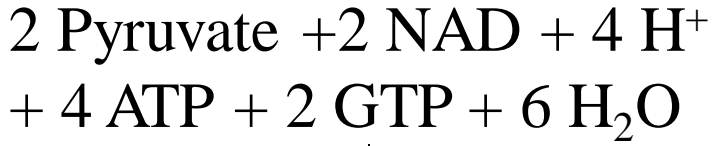
- Ca^{2+} -binding stabilizing protein is essential for phosphatase activity.
- Glucose and P_i are then shuttled back to the cytosol by transporters.



- 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 P_i and glucose, respectively back into the cytosol.

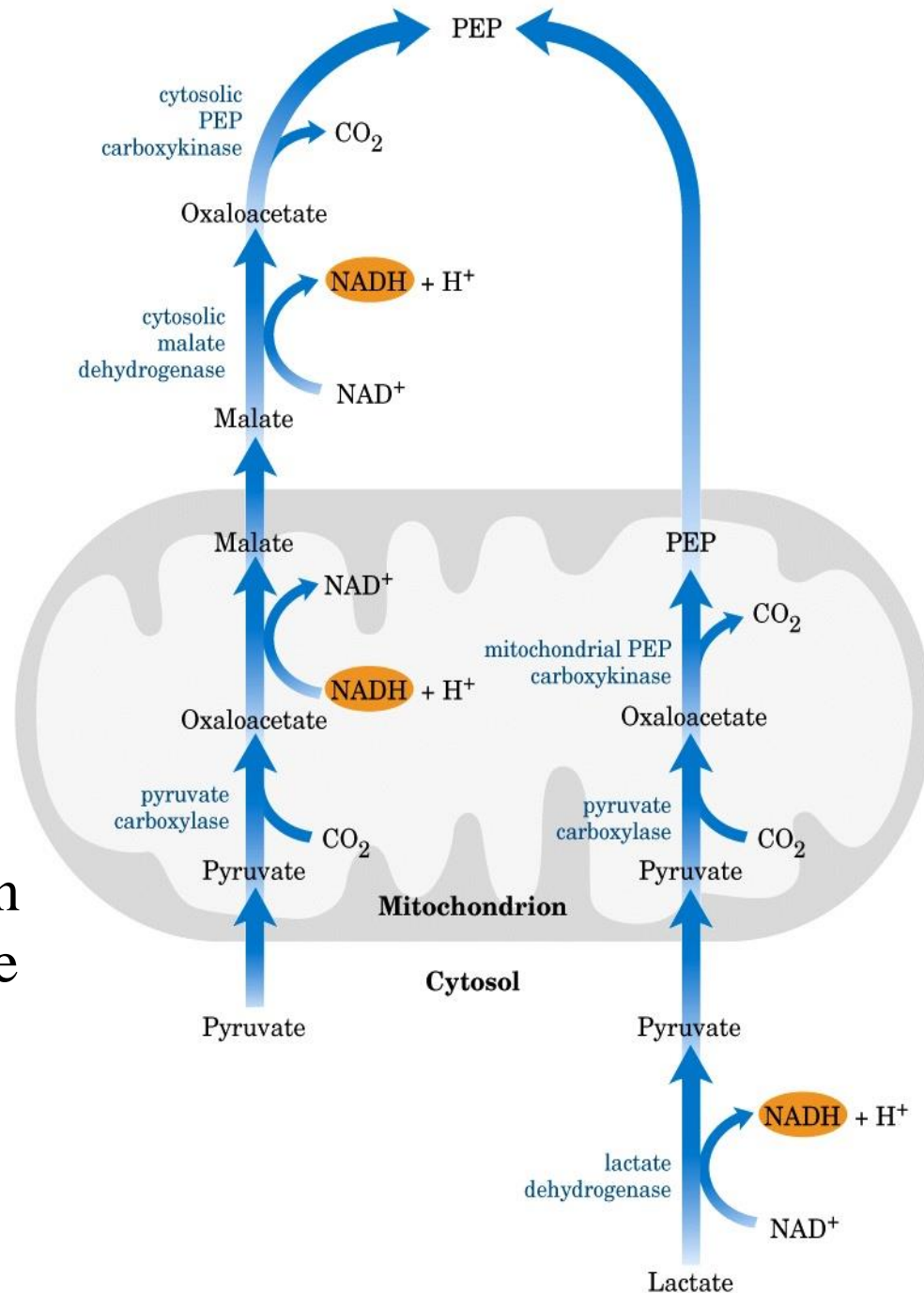
Gluconeogenesis

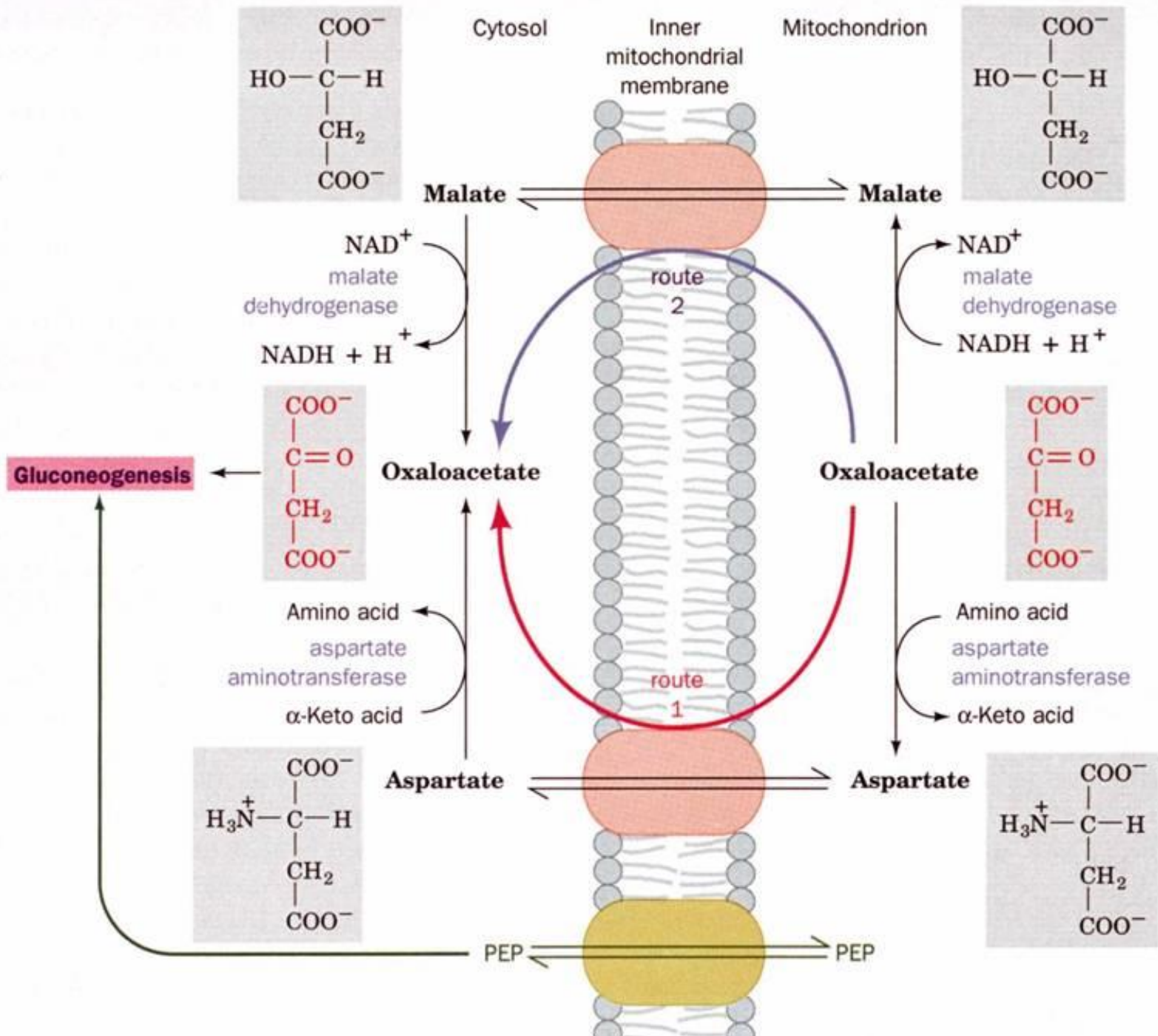
-The overall reactions of gluconeogenesis are:



Transport between the mitochondria and the cytosol

- Generation of oxaloacetate occurs in the mitochondria only, but, gluconeogenesis occurs in the cytosol.
- PEPC 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 be transported out of the mitochondria in form of Malate





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-bisphosphatase
 - 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:

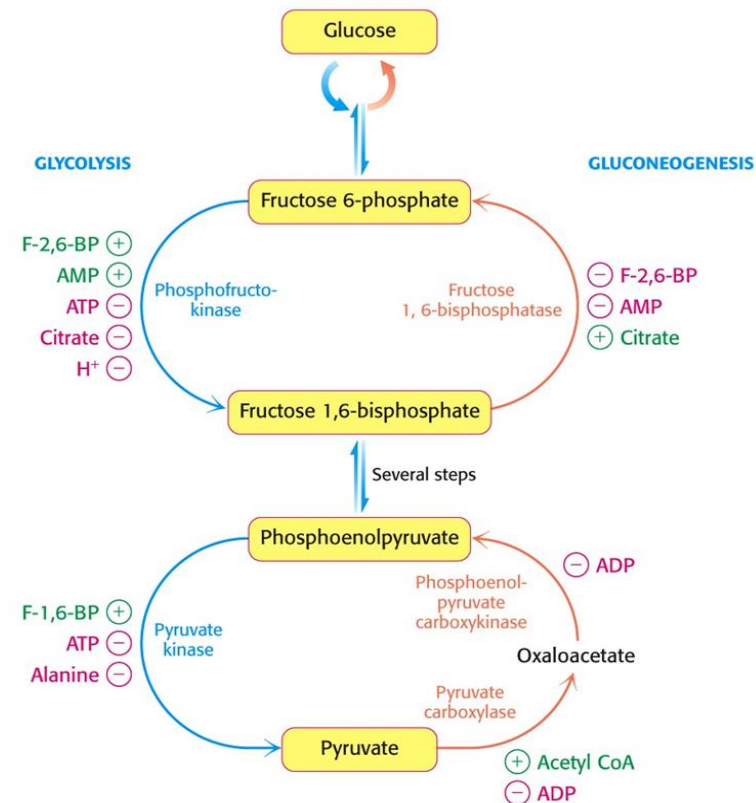
- Fructose-2,6-biphosphate stimulates glycolysis by activating PFK-1 and inhibits gluconeogenesis through the inhibition of fructose-1,6-biphosphatase.
- During starvation, gluconeogenesis predominates because the level of F-2,6-BP is very low.

Reciprocal regulation by hormones

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

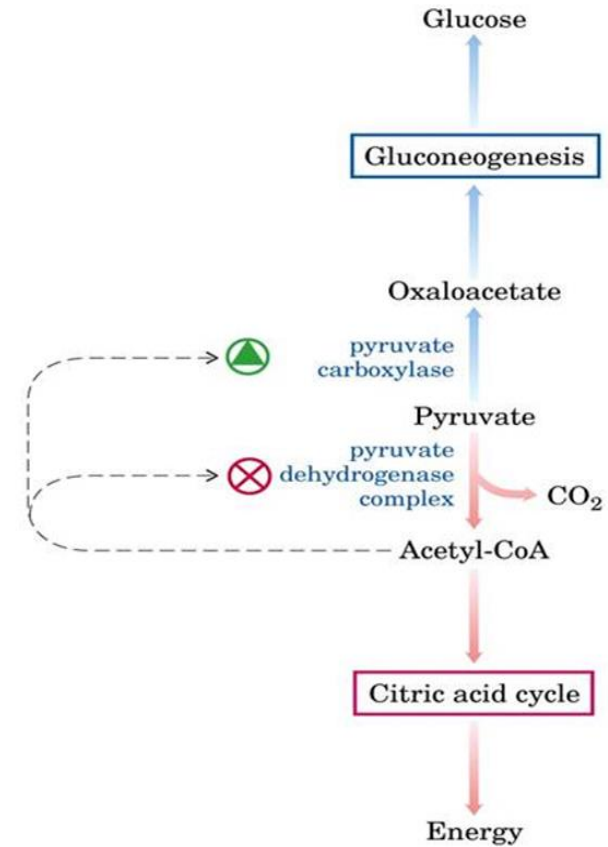
Fructose-1,6-bisphosphatase: repressed in feeding by insulin and induced in starvation by glucagon

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.

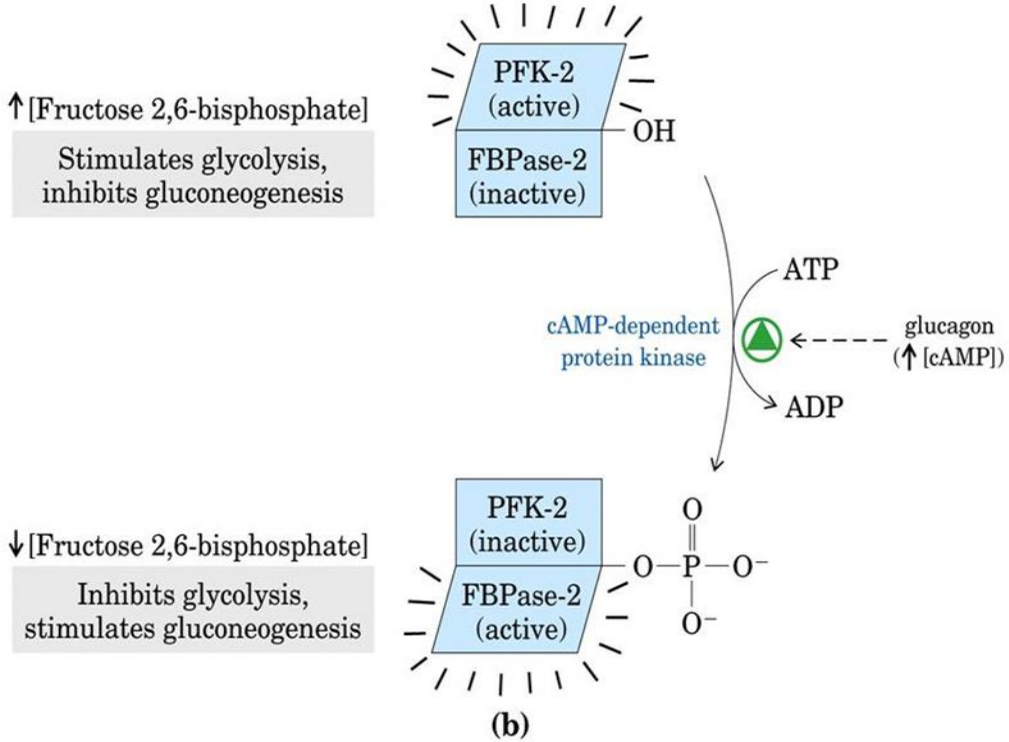
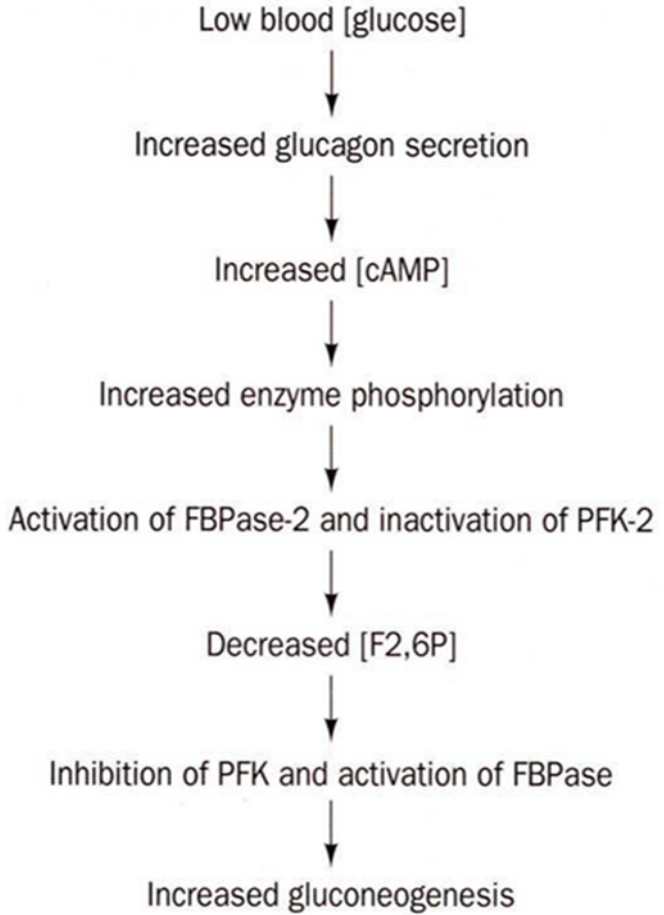
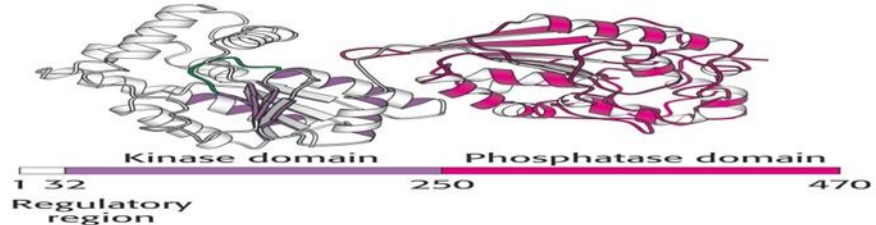


Allosteric activation by acetyl CoA

- 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.



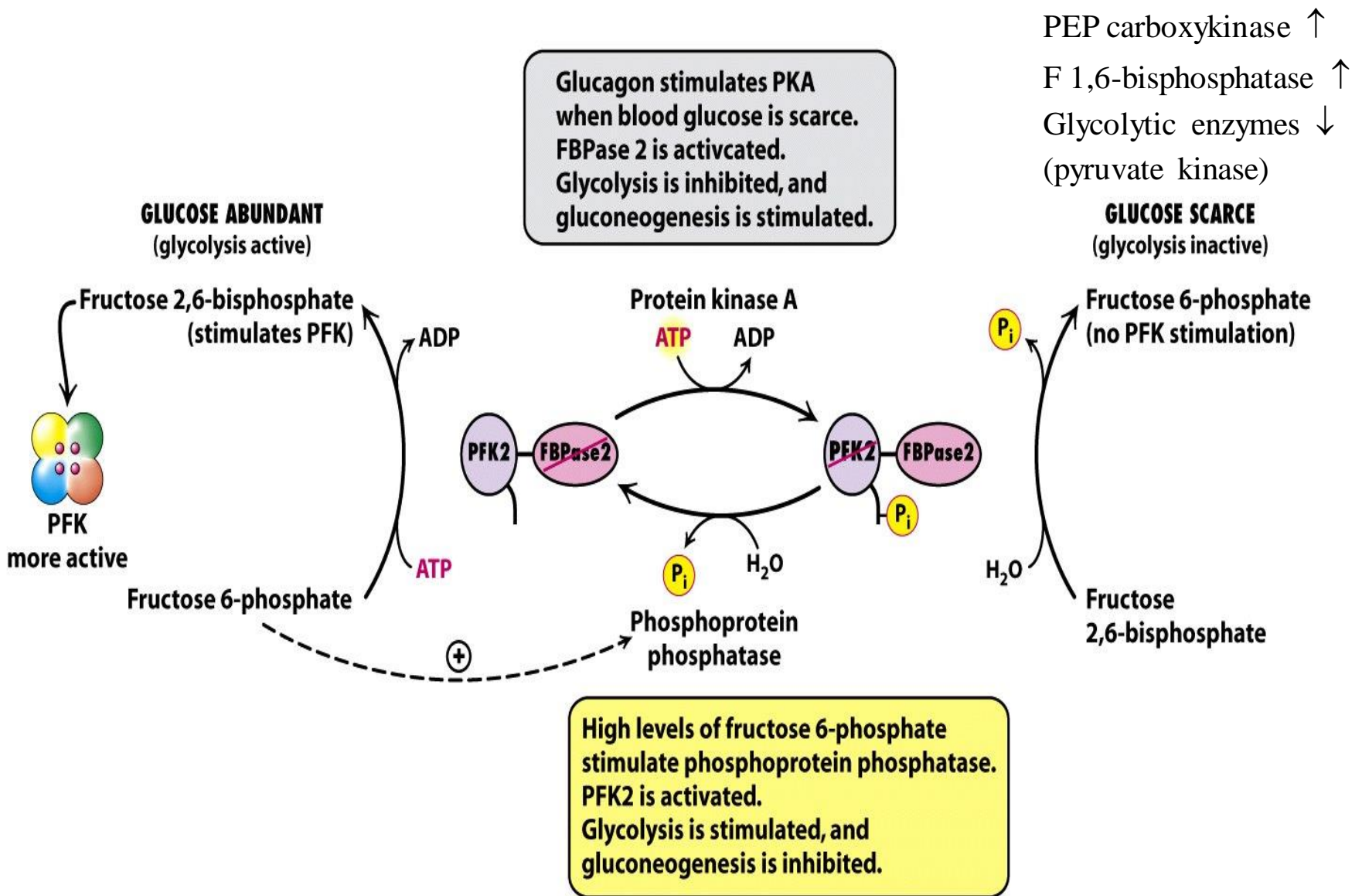
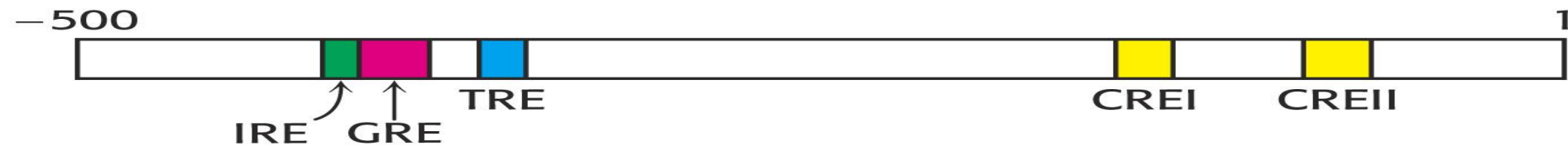


Figure 16-30
 Biochemistry, Sixth Edition
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Hormones

- 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;

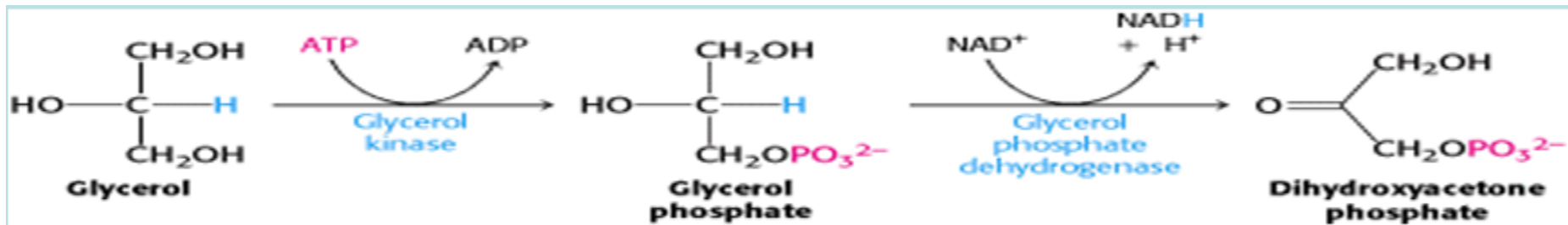
GRE: glucocorticoid response element

TRE: thyroid response element

CREI and II: cAMP response elements

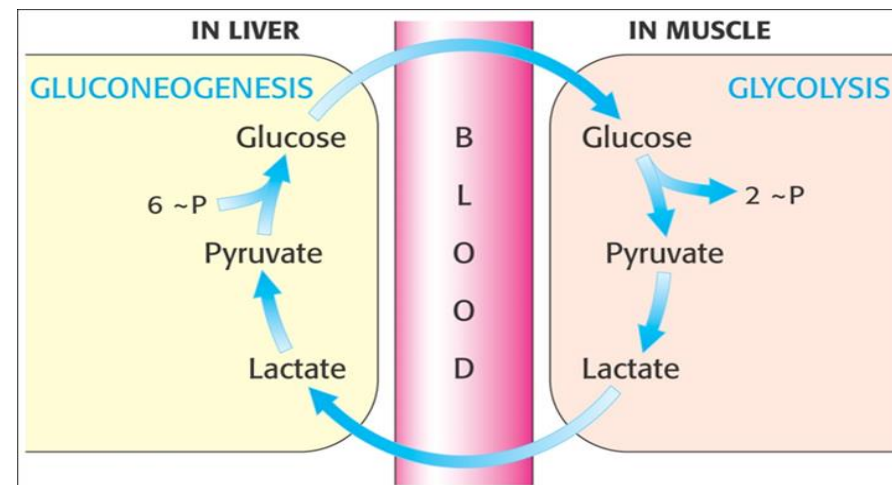
Substrates for gluconeogenesis

- 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.

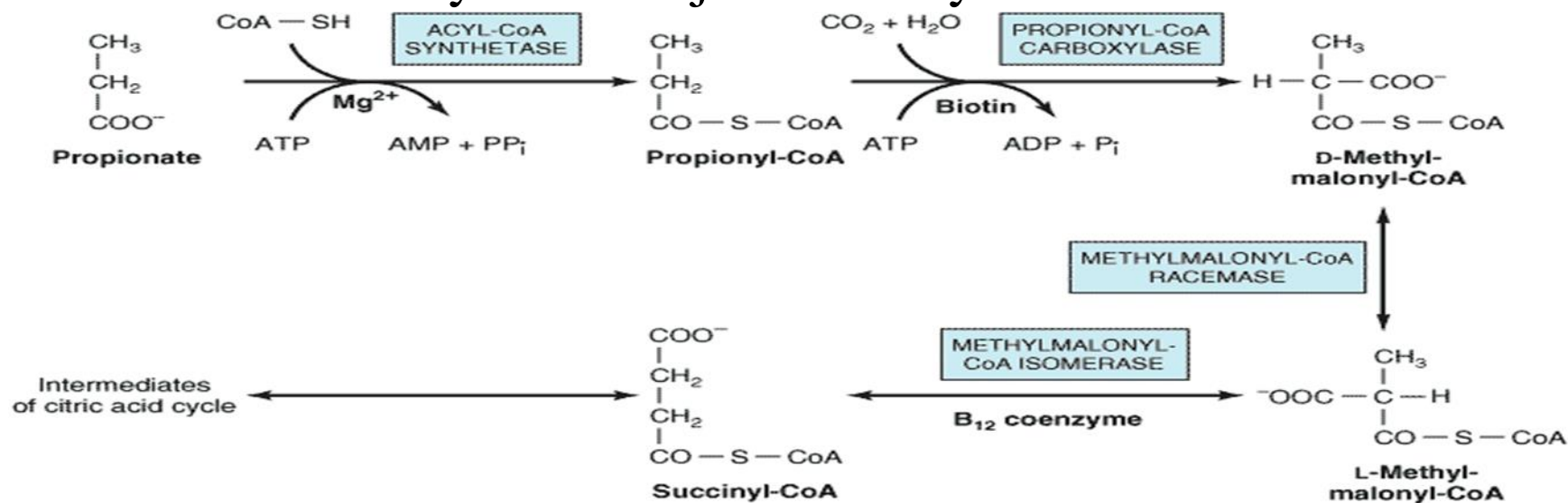


- DHAP is converted into glyceraldehyde 3-P by triose isomerase.

- **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.



- **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

