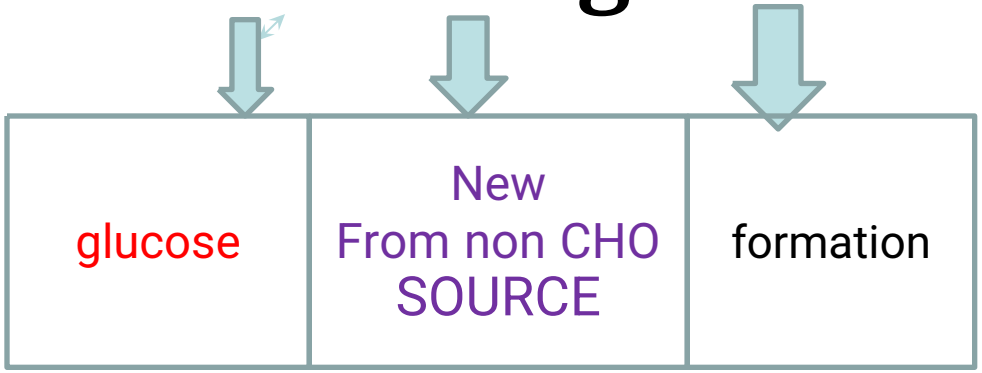
Gluconeogenesis

(formation of glucose from non CHO source)

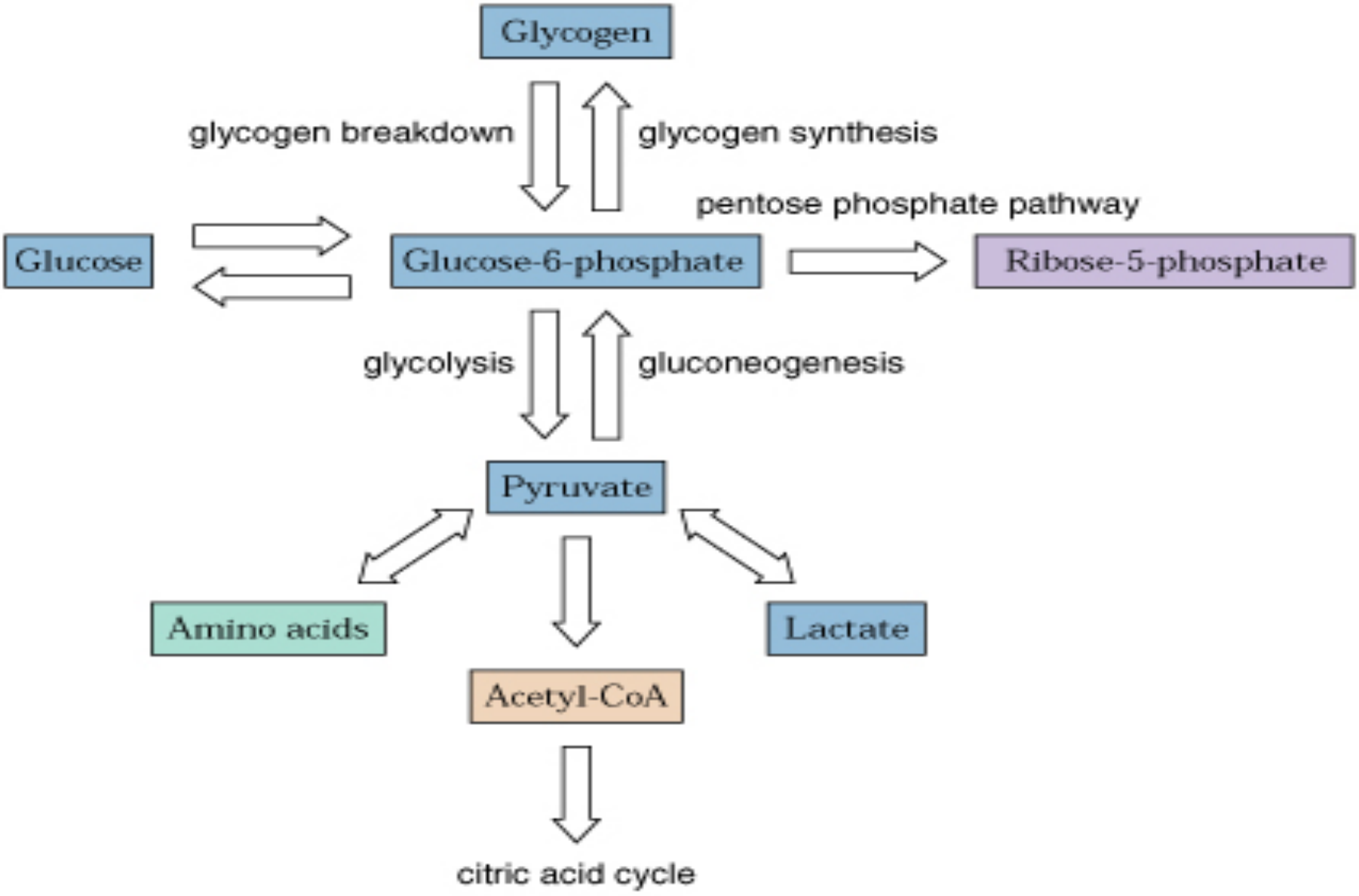
(depend on glycolysis)

Gluconeogenesis



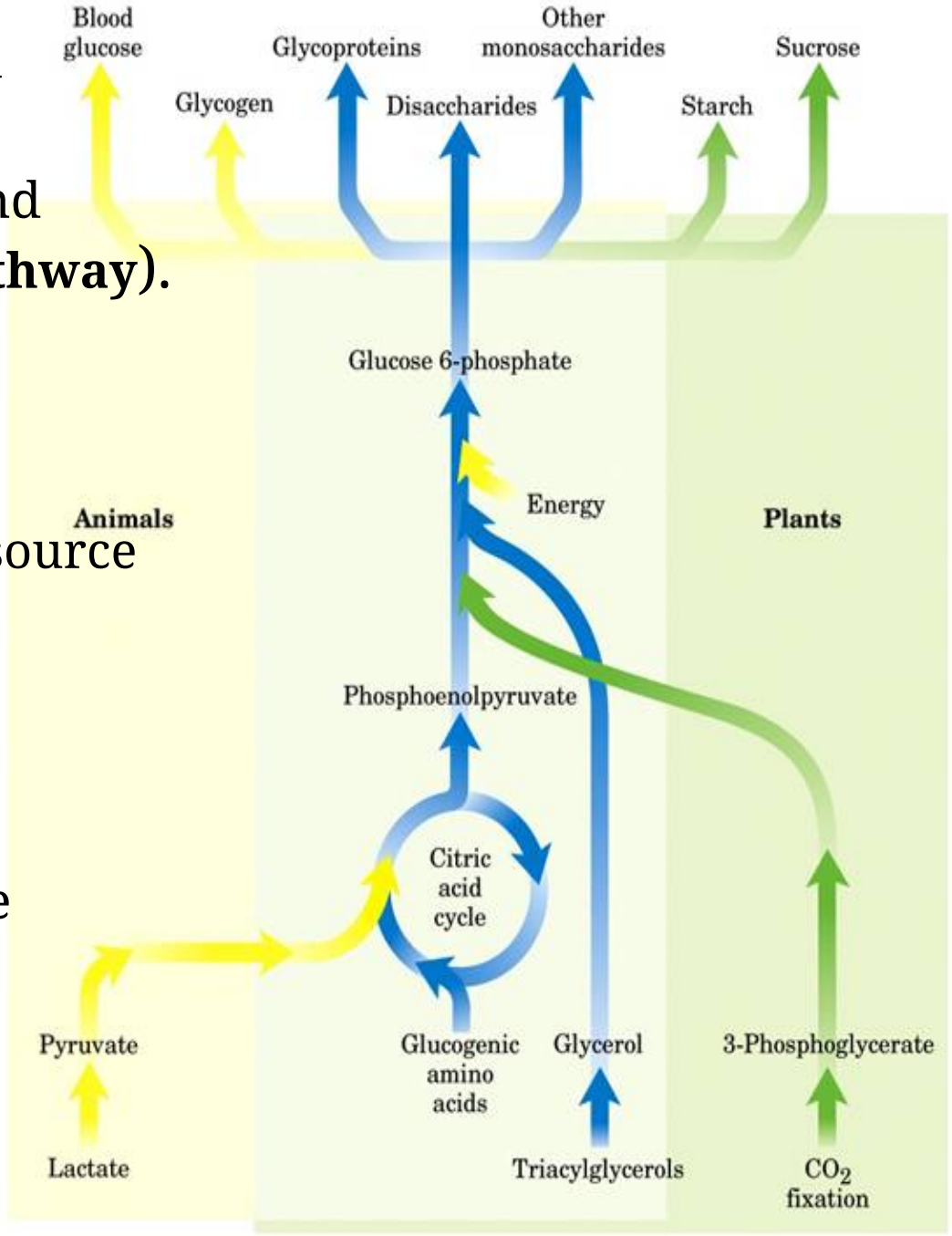
Formation of glucose from galactose(or fructose) is not cosider as gluconeogenesis

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 dose not occur by simple reversal of glycolysis. **(because here we have 3 different reaction are catalyze by irreversible enzyme)**
- **Reaction 1 :which is catalyse by hexokinase or glucokinase**
- **Reaction 3:which is catalyse by phosphofructo kinase 1**
- **Reaction 10:which is catalyse by pyruvate kinase**

- 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.

The three steps which should be bypassed in gluconeogenic pathway:

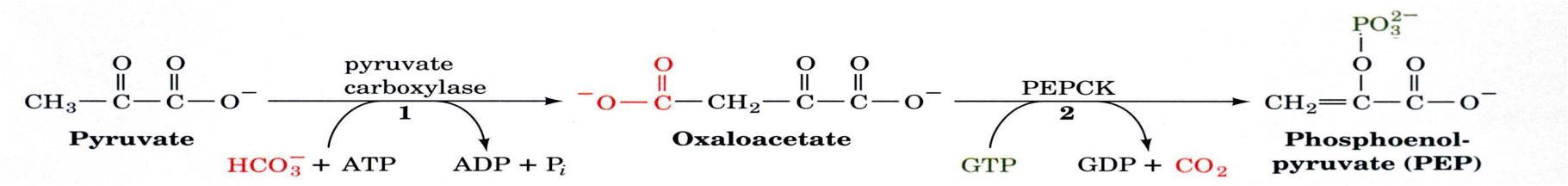
1- Pyruvate to PEP(because in glycolysis is not reversable here need 2 reaction to occure)

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.

Pyruvate carboxylase in the mitochondria of liver and kidney

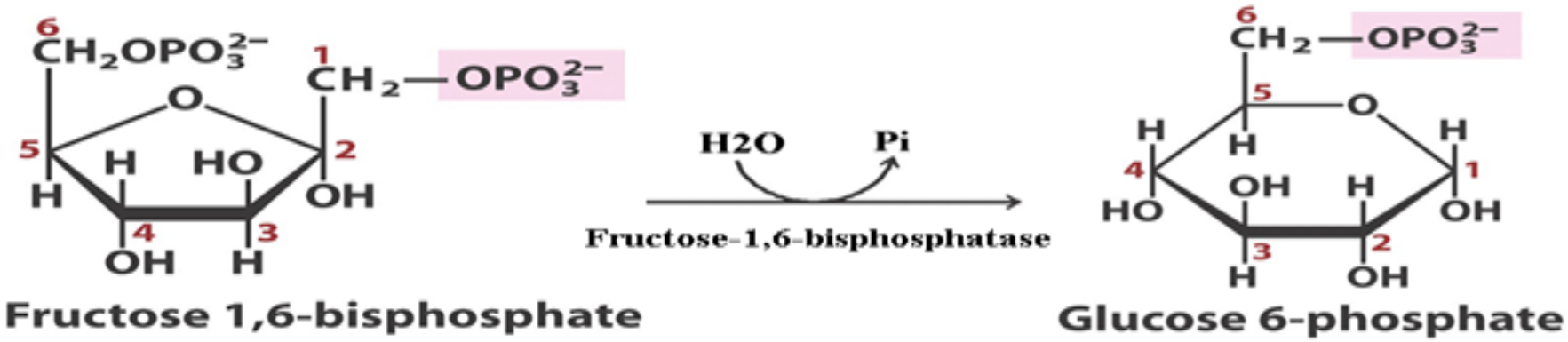


1- Pyruvate carboxylase requiring **biotin** as a cofactor, catalyses the irreversible **ATP-driven** formation of oxaloacetate from pyruvate and CO_2.

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(source of phosphate group)** as a phosphorylating agent.in cytoplasm or cytosol

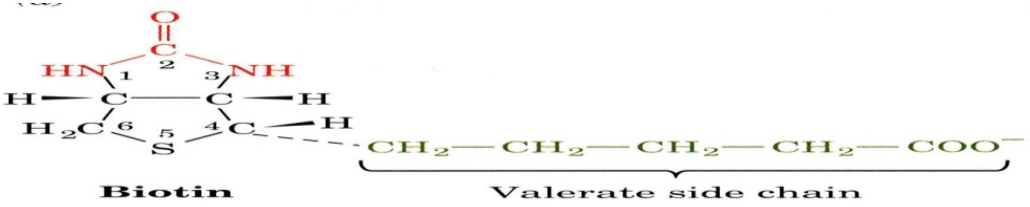
Biotin :vit.B complex water soluble vit

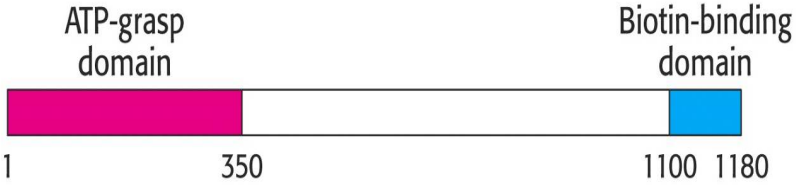


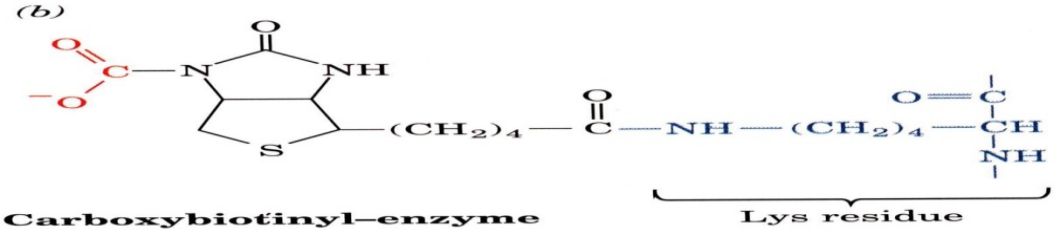
fructose 1,6-bisphosphate: increase under effect of hormone (insulin)
And inhibited under effect of glucagon

Any carboxylation reaction occur in our body need:

- **Biotin**
- **Bicarbonate as a source of carbondioxide (HCO3)**
- **Manganese**
- **ATP : for fixation of co2**







The enzyme have 2 domain:

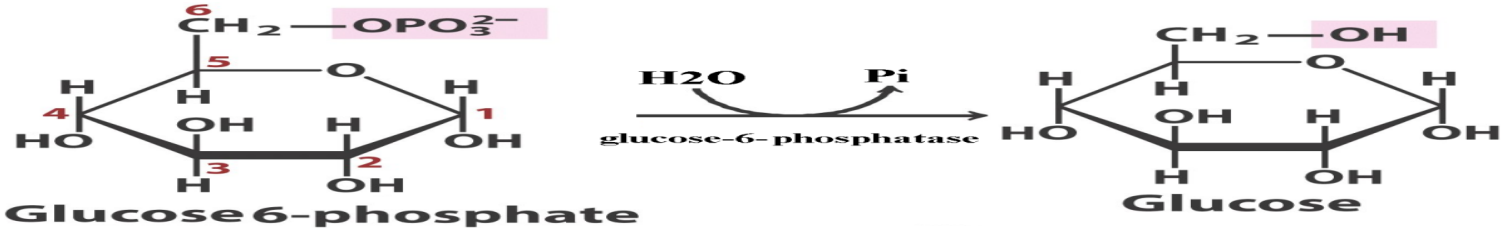
3- Hydrolysis of fructose-1,6-phosphate by fructose1,6-bisphosphatase bypasses the irreversible PFK-1 reaction.

- This reaction is an important **regulatory step**(by PFK-1) 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(IF the cell contain huge amount of ATP, no need of oxidizine phosphate
- It is inhibited also by **fructose 2,6-bisphosphate**(act as activator in glycolysis) 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.**



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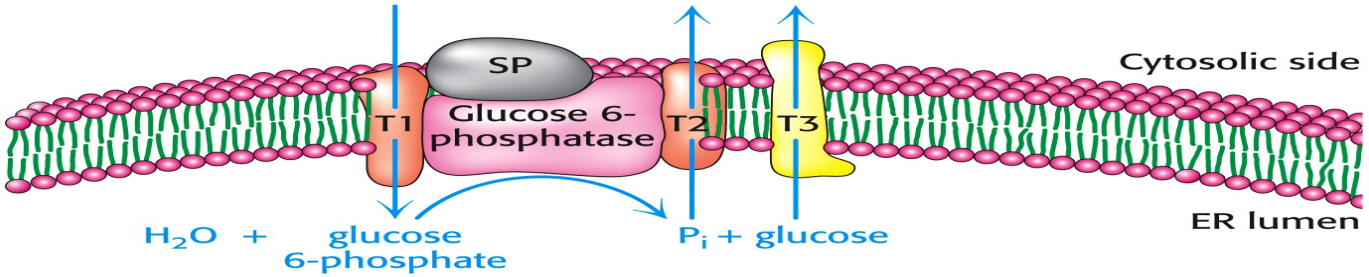
Ca^{2+}-binding stabilizing protein is essential for phosphatase activity(in ER).

- Glucose and P_i are then shuttled back to the cytosol by transporters.



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- 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(FROM cytosol)

T2 and T3 transport Pi and glucose, respectively back into the cytosol.(from ER)

Gluconeogenesis

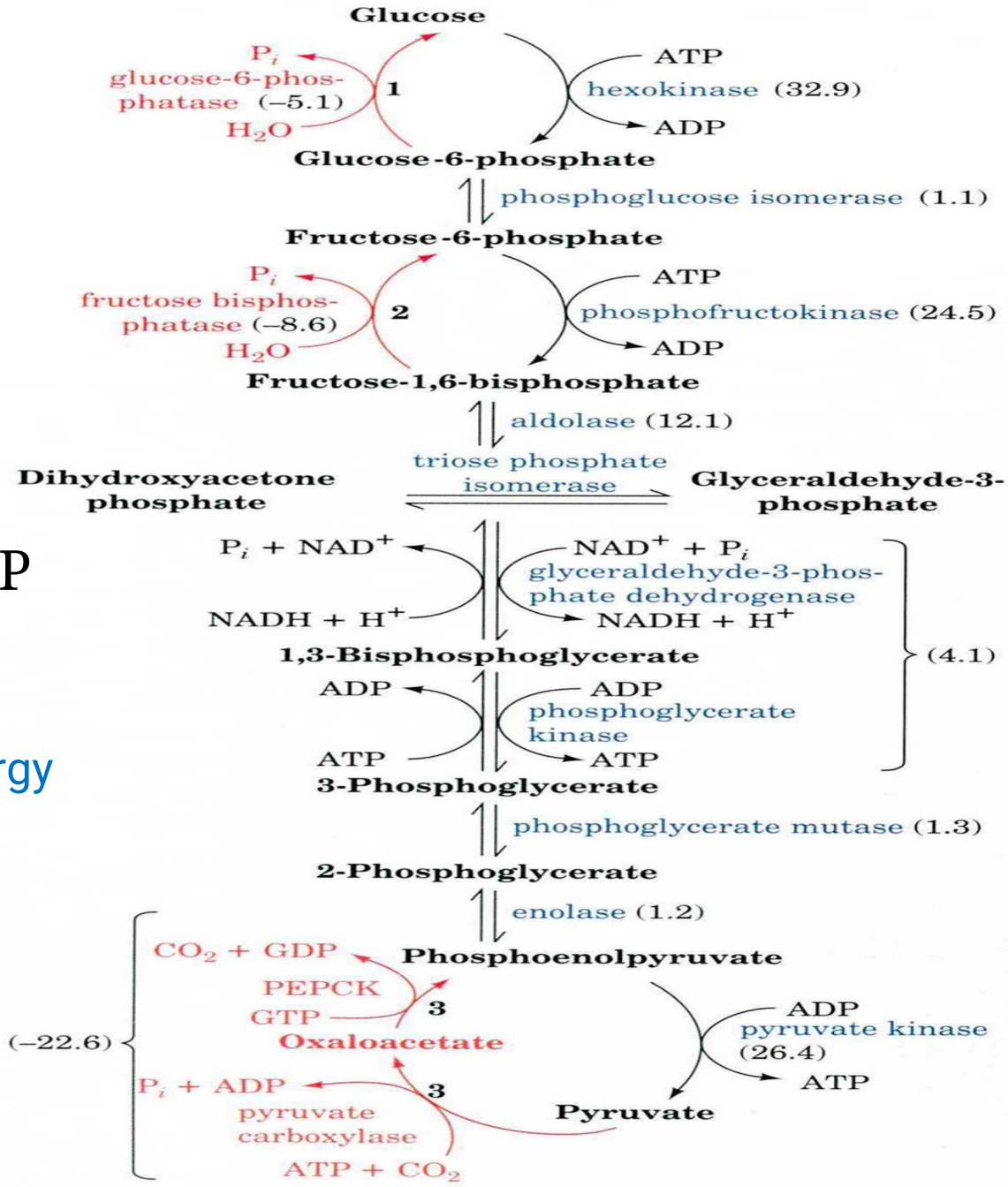
- The overall reactions of gluconeogenesis are:

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

↓

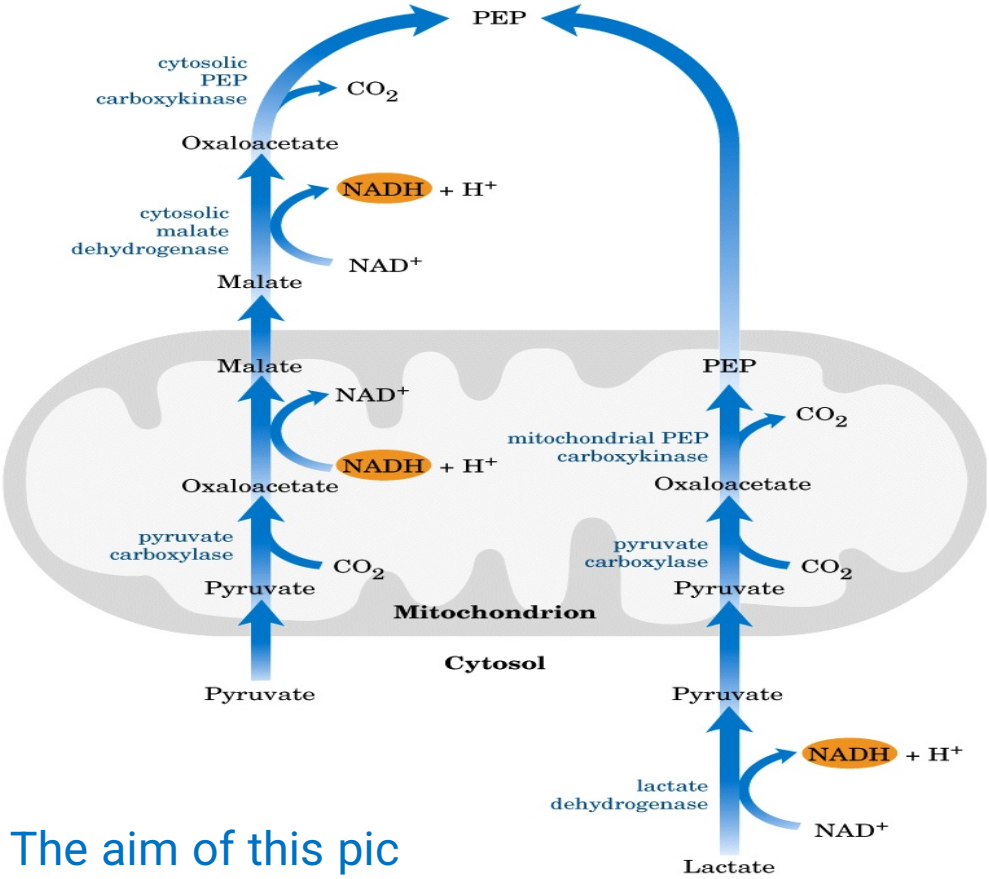
Glucose + 2 NAD^+ + 4 ADP + 2 GDP + 4 Pi

Each glucose nead 6 high energy compound
2GTP
4ATP



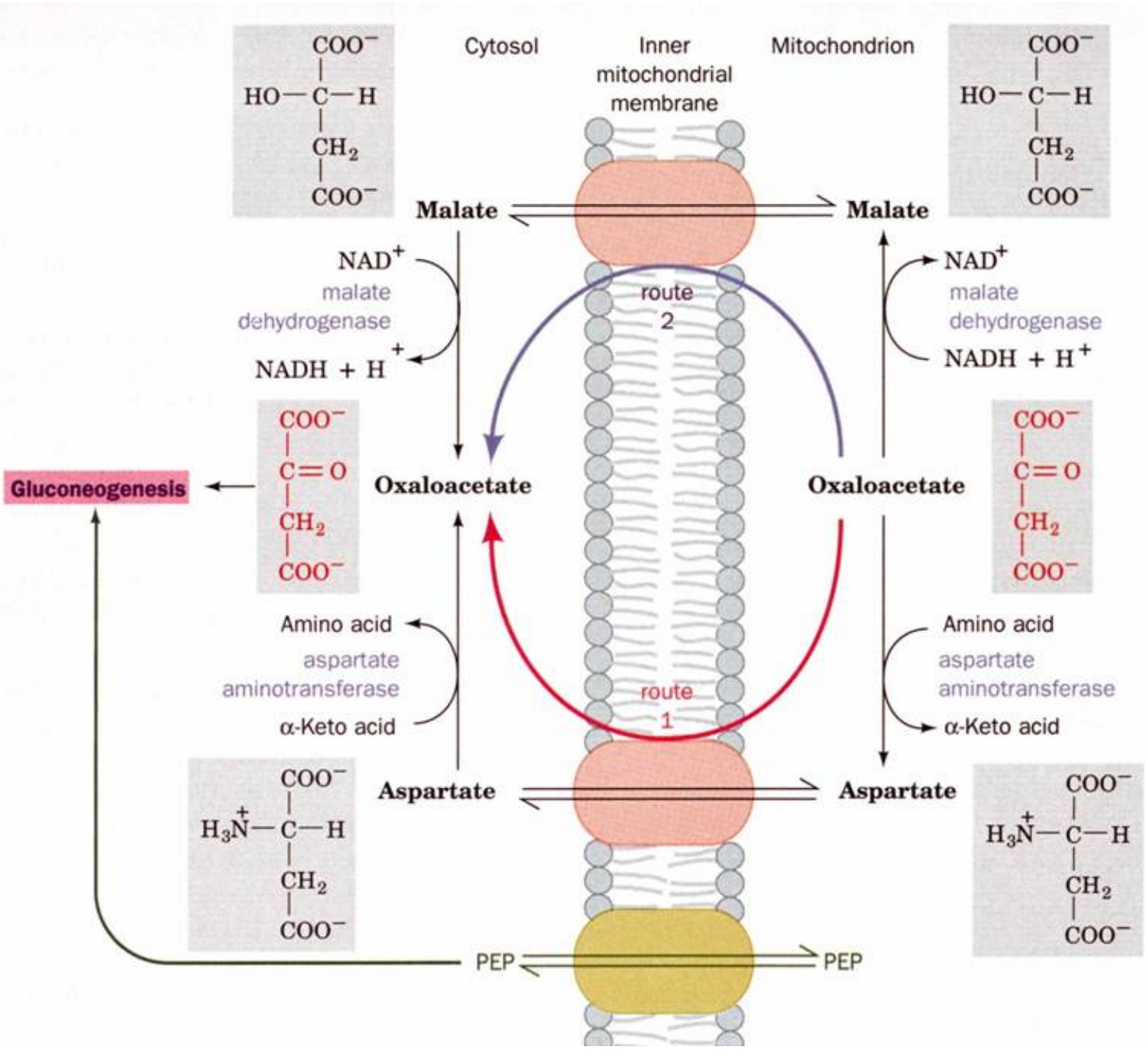
Transport between the mitochondria and the cytosol

- 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



The aim of this pic
*How PED can be translocated from inside mitochondria to Out side (cytosol)

Oxaloacitate react with A.A by transamination Reaction which include transfore amino group From A.A (become a-ketoacid) to oxaloacetate To become Aspartic acid


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

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. against PFK-1

Reciprocal regulation by ATP/AMP

- AMP inhibits fructose-1,6-bisphosphatase **(the main enzyme in gluconeogenesis**
(but activates PFK-1 **(main enzym in glycolysis(**
- ATP and citrate inhibit PFK-1 but activate fructose-1,6-biphosphatase
  - In high ATP/AMP ratio: stimulate gluconeogenesis W
  - In low ATP/AMP ratio: stimulate glycolysis**which meann there are high energy in cell**
- 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:

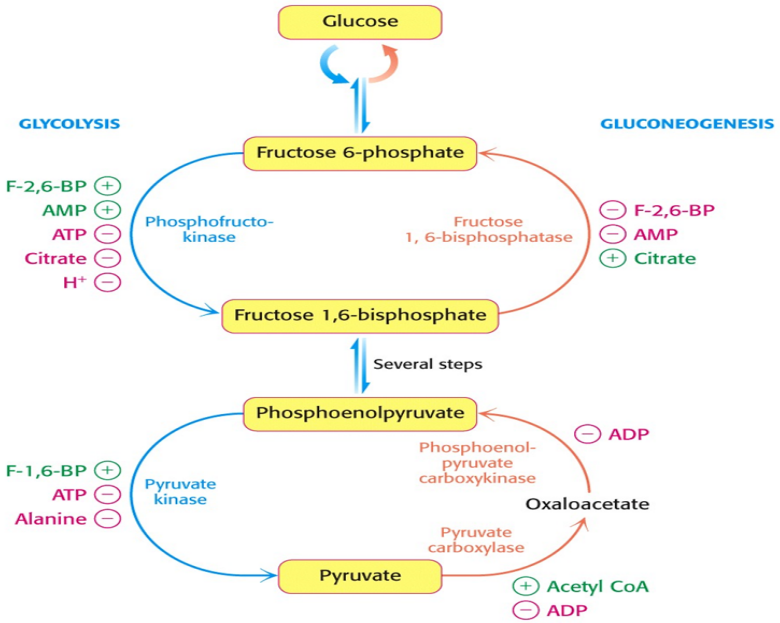
- Fructose-2,6-biphosphate**(produce by PFK-2(** stimulates glycolysis by activating PFK-1 and inhibits gluconeogenesis through the inhibition of fructose-1,6-biphosphatase.
- During starvation, gluconeogenesis predominates because the level of F-2,6-BP is very low.

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

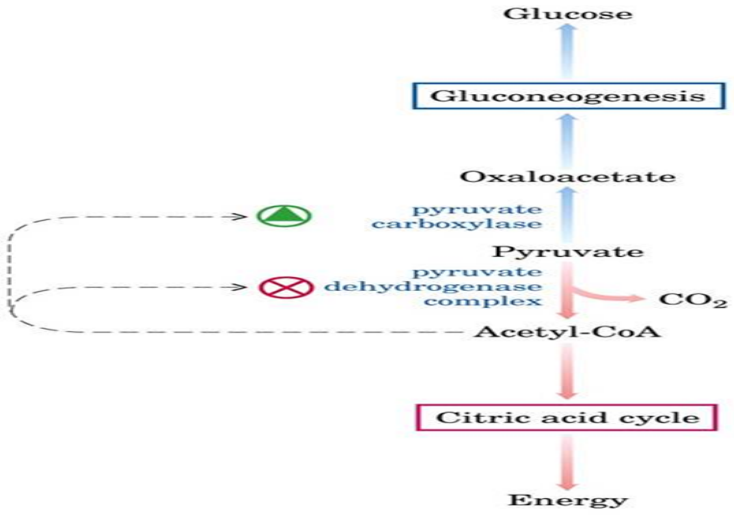


.insulin increase PFK-1Producing

gLycolysis inhibitide by Glucagon ,gluconeogenesıs

Activated by glucagon

Acetyl-CoA regulates pyruvate carboxylase(activator(



- The increase in oxaloacetate concentratior
  → 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 are high
- , TCA cycle is
  increased, oxaloacetate goes to glucose.

Allosteric activation by acetyl CoA

- During starvation **(under effect of glucagon(** → excessive lipolysis → excessive oxidation of
  fatty acid into acetyl CoA → accumulation of acetyl CoA → activation of pyruvate carboxylase → activation of gluconeogenesis.

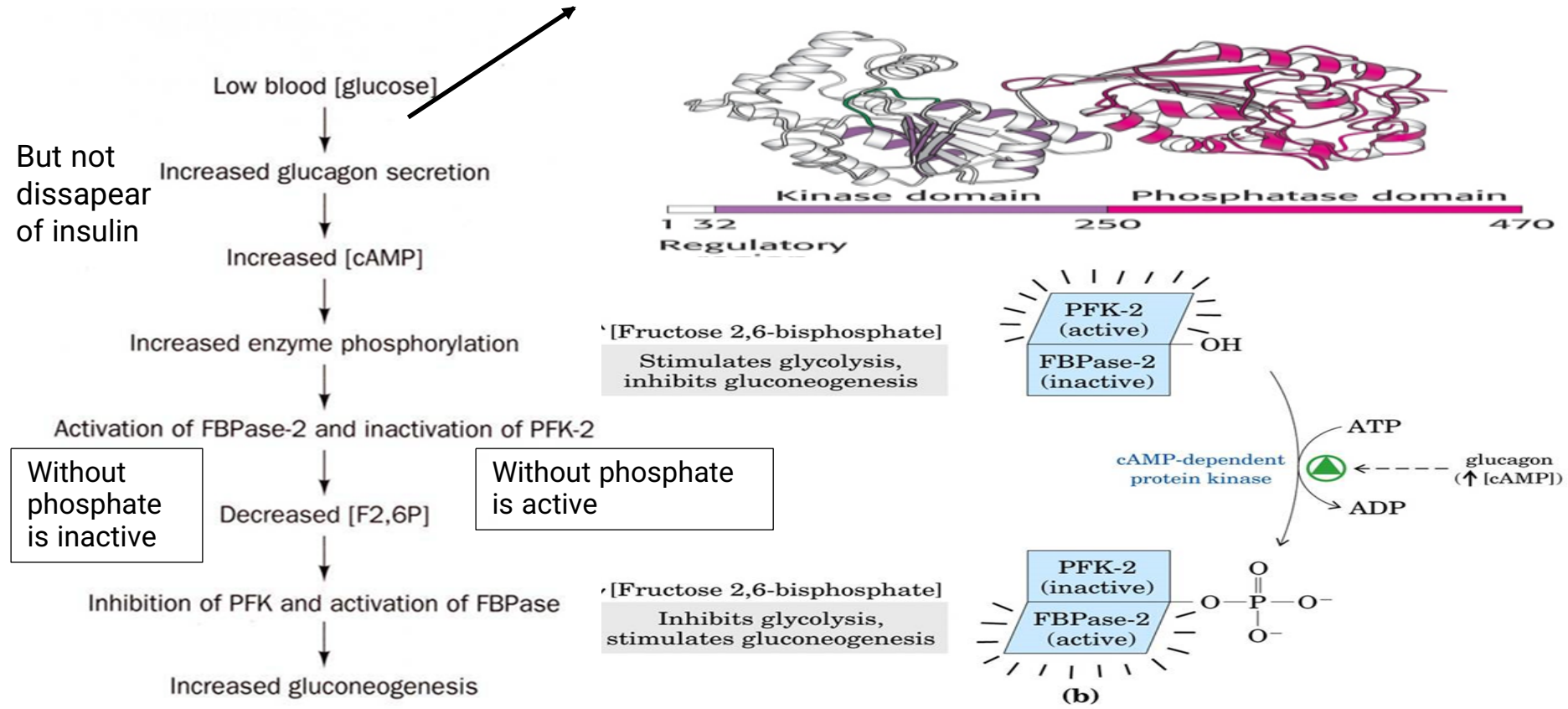
Citric acide cycle produce energy if energy is Exceed The Need of cell,Extra amount of Actyl coA available ,activate gluconeogenic

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.

PFK-2 →produce 2,6-biphosphate

FBPase-2 → responsible for degradation
Fructose-2,6 biphosphate doesnt activate glycolysis, but activate glucogenesis



PEP carboxykinase ↑

F 1,6-bisphosphatase ↑

Glycolytic enzymes ↓
(pyruvate kinase)

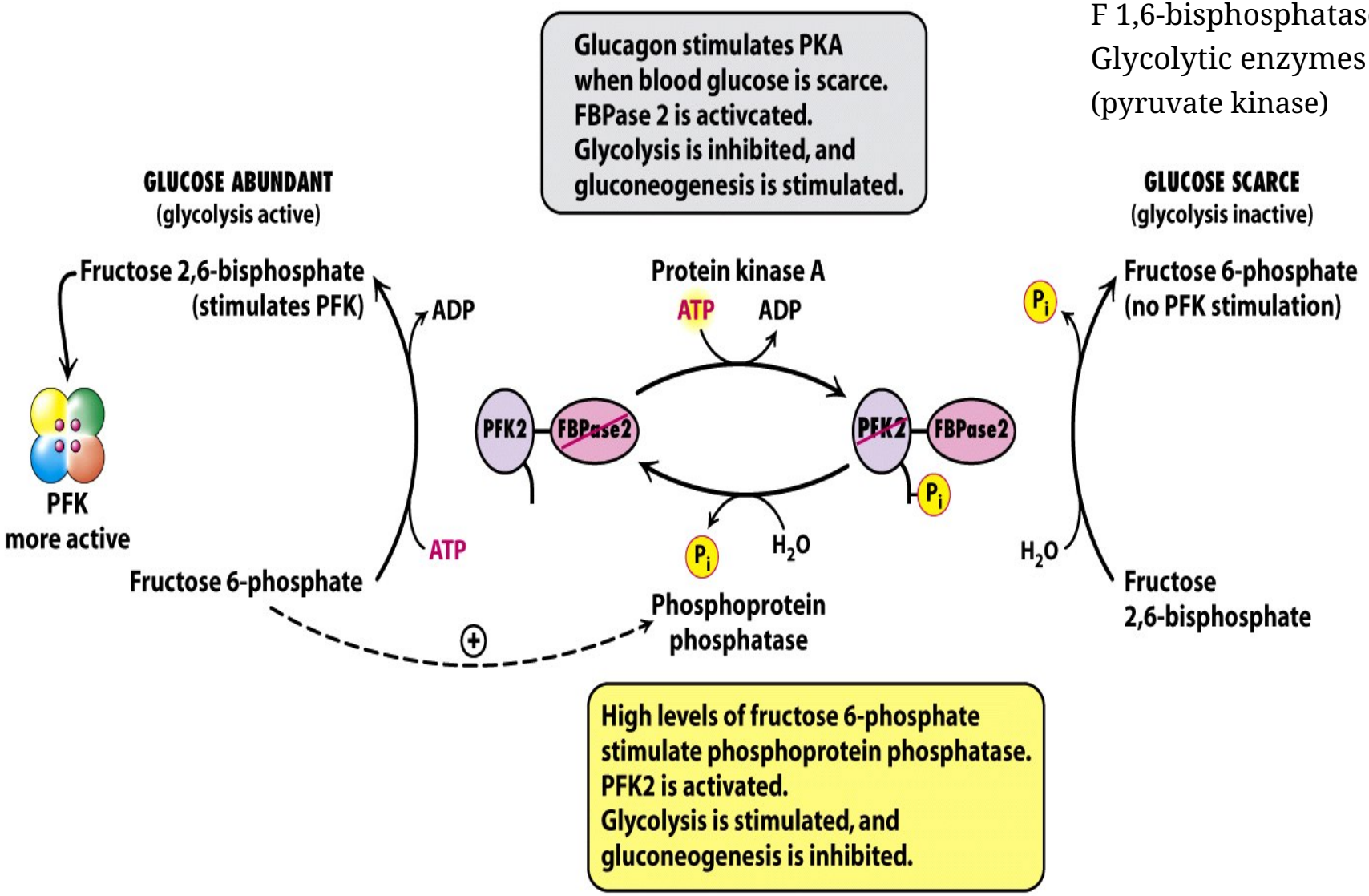
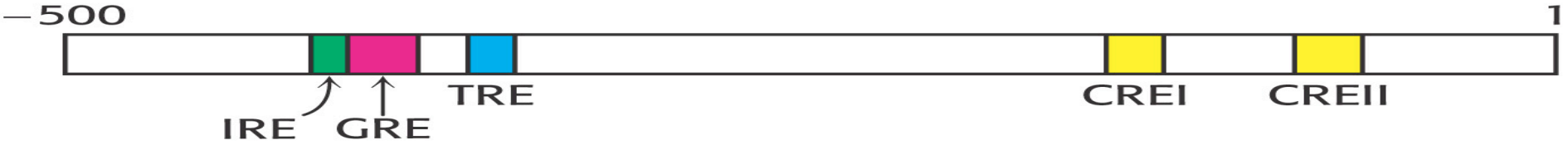


Figure 16-30
Biochemistry, Sixth Edition

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)

Genetic level regulation



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

IRE: insulin response element;

GRE: glucocorticoid response element

TRE: thyroid response element

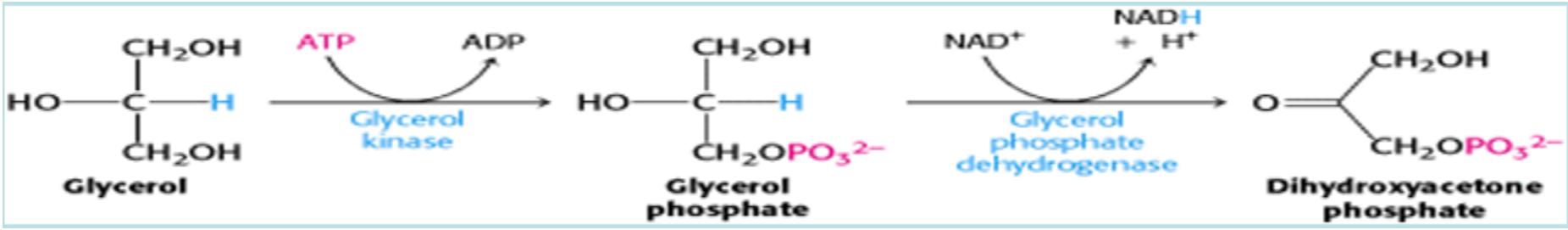
CREI and II: cAMP response elements

There are the response element of these enzyme

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. By libolysis

By gycerol kinase → This enzyme is not found in muscle and adipose tissue



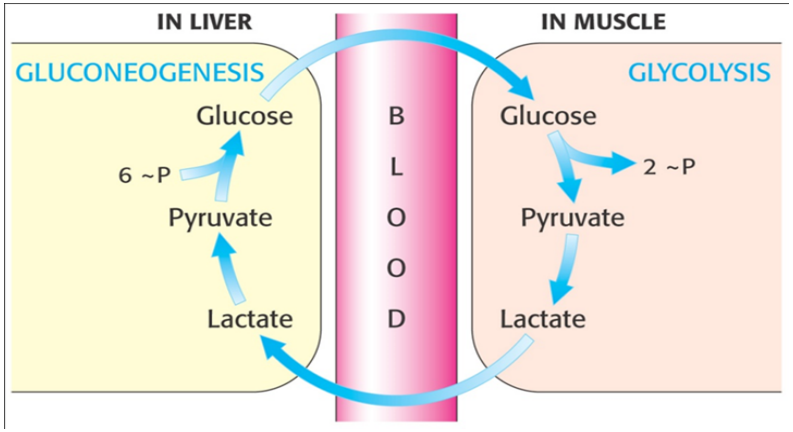
DHAP is converted into glyceraldehyde 3-P by triose isomerase.

Part of it stay DHAP

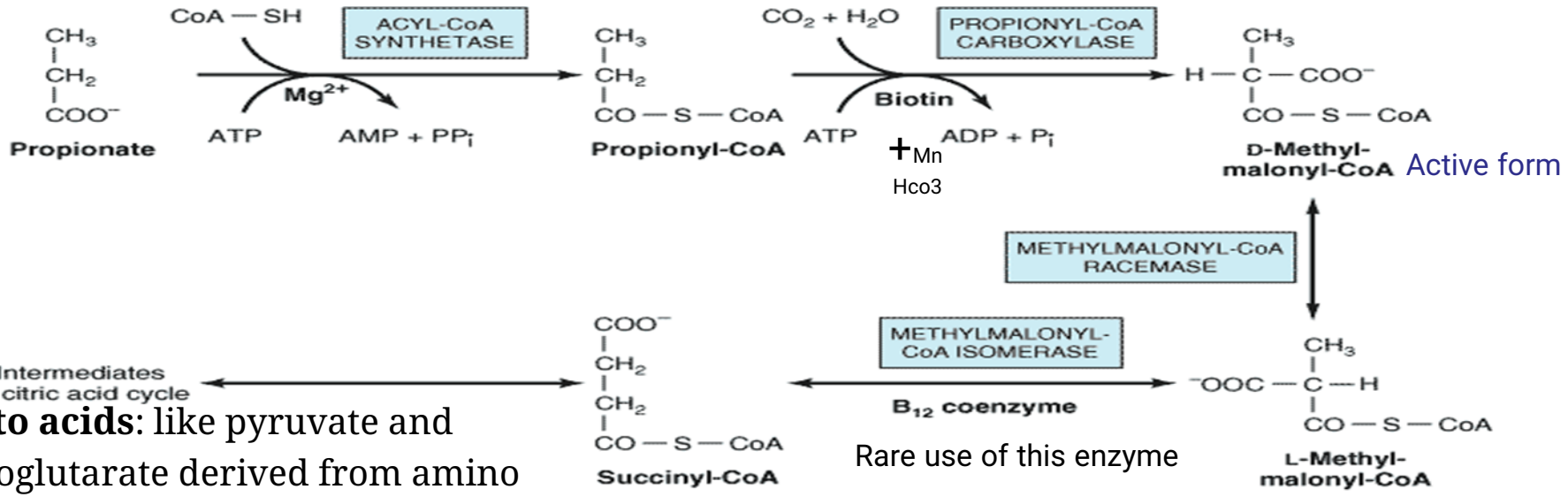
- **Lactate**: under anerobic condition
- released from the RBC and exercising muscle, carried to the liver by the blood and converted to glucose and released again to blood through Cori cycle. By lactate dehydrogrnase

Becouse it Catalytic Reversable enzyme

Pyrouvate



Odd chain fatty acids(الاحماض الدهنية عدد كربوناتها فردي): 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. (which are ketogenic)

Because during their catabolism, they give intermediate to CAC as long as→it gives oxaloactate → glucose

Glucogenic Amino Acids, Grouped by Site of Entry*

Pyruvate
Alanine
Cysteine
Glycine
Serine
Tryptophan

They are not outer CAC

α-Ketoglutarate
Arginine
Glutamate
Glutamine
Histidine
Proline

Succinyl-CoA
Isoleucine
Methionine
Threonine
Valine

Fumarate
Phenylalanine
Tyrosine

Oxaloacetate
Asparagine
Aspartate

F.A in each cycle to be oxidized in B-oxidation will loss 2 carbon atoms
so if we have F.A (30 C) , it will loss 2C in each time

Propionic acid →

اما اذا كان عدد الكربونات فردي رح ينتج مركب نهائي بتكون من3 كربونات

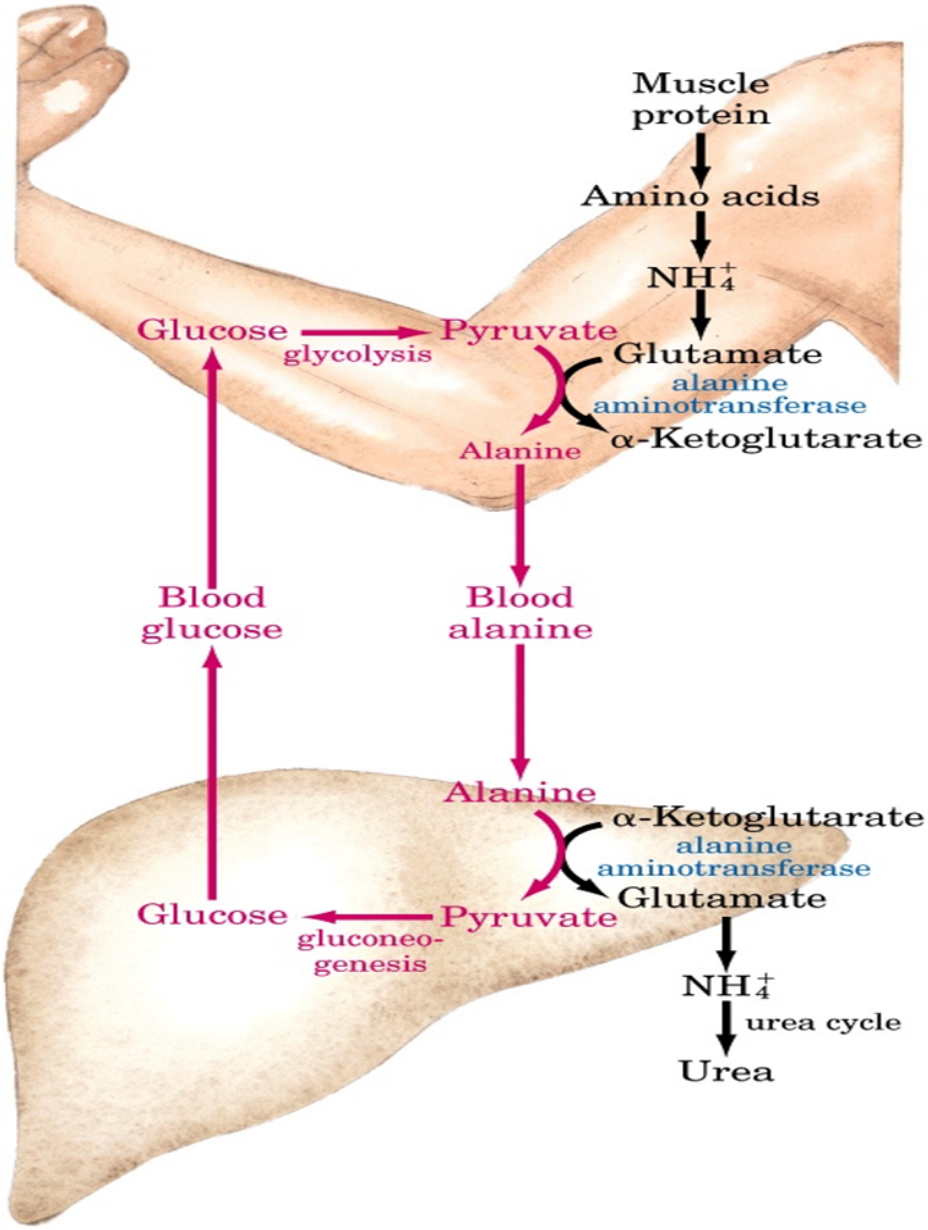
- Acetyl CoA cannot give rise to a net synthesis of glucose because of the irreversible nature of PDH that converts pyruvate to acetyl CoA.

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

Glucose → pyruvate → alanine → liver
يحدث العكس



* Acetyl Co-A → is not Glucogenic compound? Why?
because Acetyl Co-A → is can formed from 20 compound

* The main enzyme which are produced Acetyl Co-A is PDH complex which is catalyze irreversible reaction and our cell can't contain an enzyme reverse it

---

* There are 2 cycle to resupply the muscle with glucose

(1) Cori cycle (muscle - liver) for conversion lactate to glucose

(2) Alanine (muscle - live) for conversion alanine to pyruvate → glucose