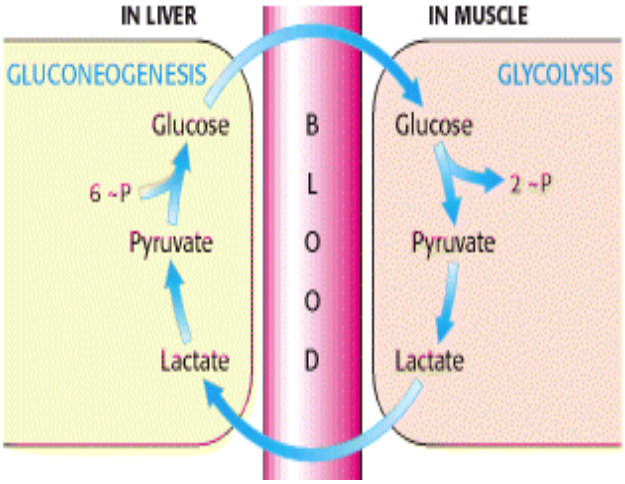
Gluconeogenesis

- When dietary intake of carbohydrate is low
- Blood glucose concentration declines
- Hormones including glucagon trigger accelerated glucose synthesis from noncarbohydrate sources in a process called ***gluconeogenesis*.**
- **Noncarbohydrate sources**
- Glycerol
- Lactate
- Amino acids
- **This occurs mainly in the liver**
- But in prolonged starvation **kidney** will also perform gluconeogenesis
- Most of the glucose formed by the liver and the kidneys is released into the blood to maintain blood glucose levels.

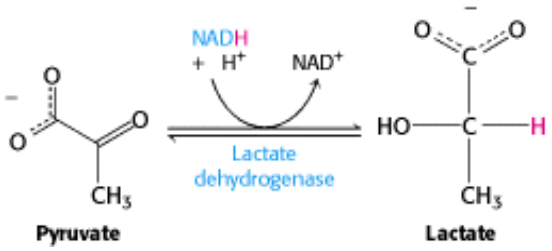
- Many steps in gluconeogenesis are the reverse of glycolysis.
- Gluconeogenesis consumes ATP and NAD+
- Most of the cytosolic enzymes involved in glycolysis catalyze their reactions reversibly and therefore provide the means for also converting pyruvate to glucose.
- When the cell is oxidizing glucose for energy, it does not need to make glucose from gluconeogenesis so **Both glycolysis and gluconeogenesis must be regulated.**
- **The nonreversible reactions are regulated**

- In mammals, some tissues depend almost completely on glucose for their metabolic energy
  - Human brain and nervous system(The brain alone requires about 120 g of glucose each day—more than half of all the glucose stored as glycogen in muscle and liver)
  - Erythrocytes
  - Testes, renal medulla, and embryonic tissues
- The supply of glucose from glycogen stores is not always sufficient; between meals and during longer fasts, or after vigorous exercise, glycogen

Cori Cycle



During muscular activity, the store of ATP needs to be constantly replenished.. Pyruvate TCA cycle
Intense workout- less O2--- lactic acid produced and NADH is reoxidized to NAD+ more glycolysis
After vigorous exercise, lactate produced by anaerobic glycolysis in skeletal muscle returns to the liver and is converted to glucose, which moves back to muscle and is converted to glycogen (if muscular activity has stopped)a circuit called the Cori cycle



- Three reactions in the glycolytic sequence are highly exergonic, highly regulated, and *not* reversible: those catalyzed by the enzymes glucokinase (hexokinase), phosphofructokinase, and pyruvate kinase (reactions 1, 3, and 10)
- These three reactions have a large negative free-energy change, delta *G*, whereas other glycolytic reactions have a delta *G* near 0= equilibrium so direction will depend on substrate or reactant availability

- The three irreversible reaction are bypassed in Gluconeogenesis

Step 1 The first of the bypass reaction

Conversion of pyruvate to phosphoenolpyruvate (PEP)

- Occurs Matrix of Mitochondria
- The bypass of the pyruvate kinase reaction involves the formation of oxaloacetate as an intermediate.
- First Pyruvate from cytosol enters mitochondria
- Mitochondrial pyruvate can be converted to oxaloacetate by **pyruvate carboxylase (found in mitochondria)**
- ***Anaplerotic* process**
  - Pyruvate- oxaloacetate
- USES an ATP to attach a CO2 to Pyruvate

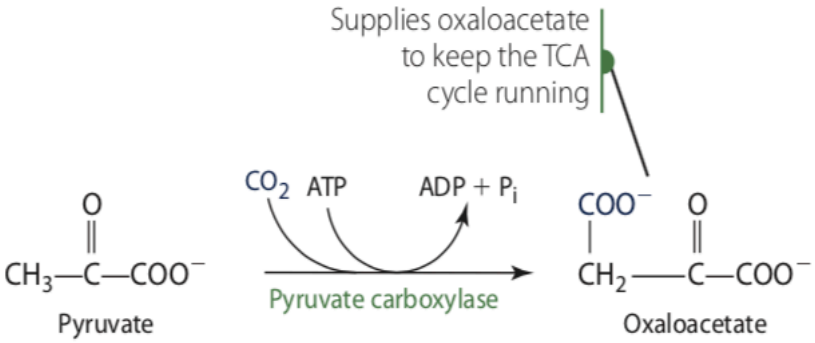


Figure 3.22 Formation of oxaloacetate from pyruvate and CO_2.

Requires Biotin as a cofactor

Additionally, Pyruvate can be generated in mitochondria from from alanine within by transamination reaction

Step 2 The first of the bypass reaction

- Occurs in Cytoplasm
- mitochondrial membrane has no transporter for oxaloacetate, before export to the cytosol the oxaloacetate formed from pyruvate must be reduced to malate by mitochondrial **malate dehydrogenase,** at the expense of NADH:

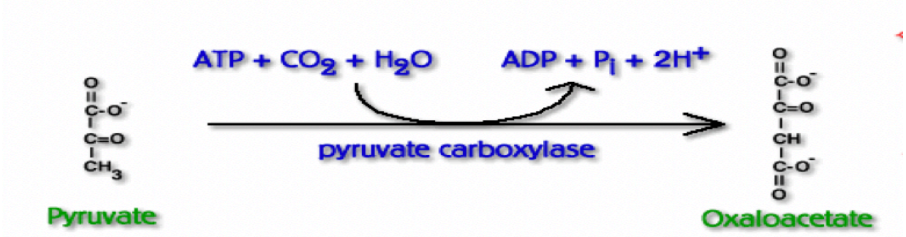
- Oxaloacetate must be reduced to malate to move to cytoplasm

Oxaloacetate +NADH + H → Malate + NAD+

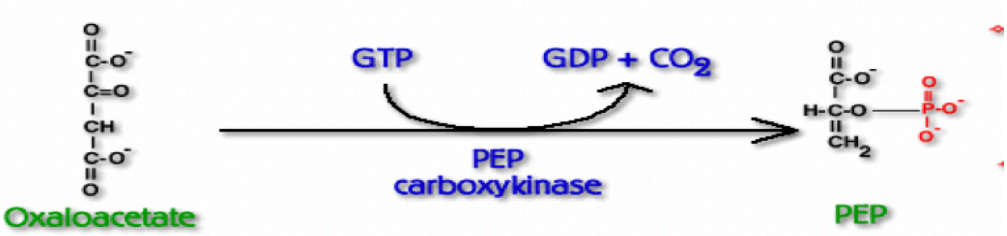
Step 2 (CONTINUED)

- In the cytosol, malate is reoxidized to oxaloacetate, with the production of cytosolic NADH:
- The oxaloacetate is then converted to PEP by **phosphoenolpyruvate carboxykinase**
- Mg+2 -dependent reaction requires GTP as the phosphoryl group donor

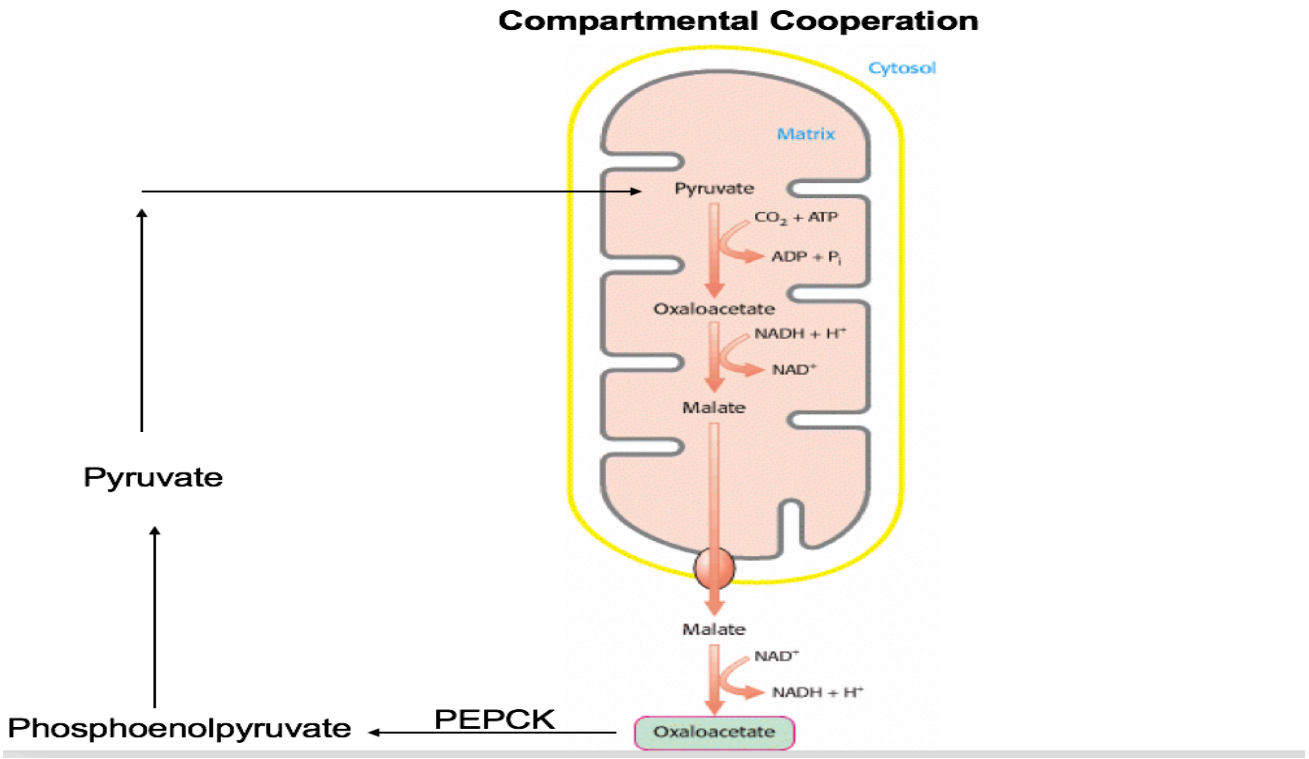
$$\text{Oxaloacetate} + \text{GTP} \rightleftharpoons \text{PEP} + CO_2 + \text{GDP}$$



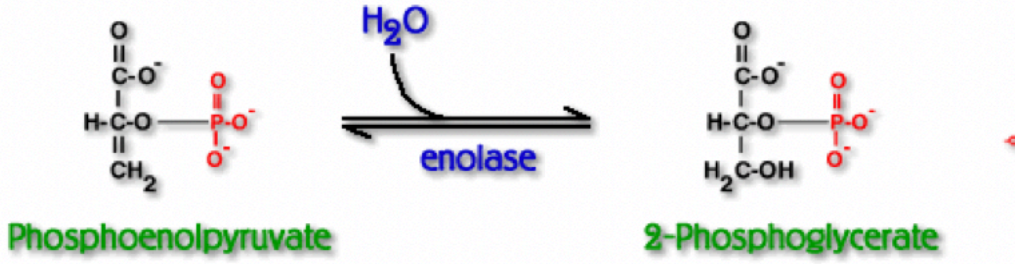
First step of Bypass I.
Pyruvate moves from the cytoplasm into the mitochondrial matrix. After conversion, the oxaloacetate moves back to the cytoplasm.



Second step of Bypass I.
Oxaloacetate is converted to Phosphoenolpyruvate (PEP).



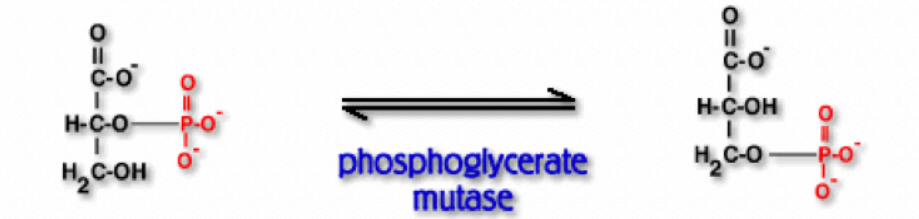
Phosphoenolpyruvate is converted to fructose 1,6-bisphosphate by reversal of the glycolytic reactions.



Phosphoenolpyruvate

2-Phosphoglycerate

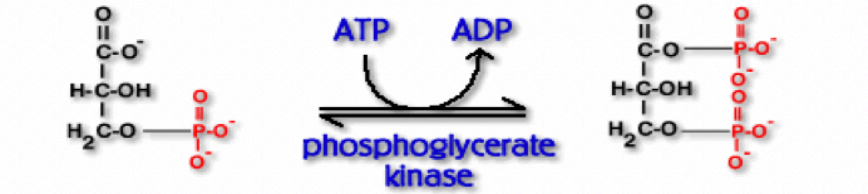
Reverse of Glycolysis reaction.



2-Phosphoglycerate

3-Phosphoglycerate

Reverse of Glycolysis reaction.

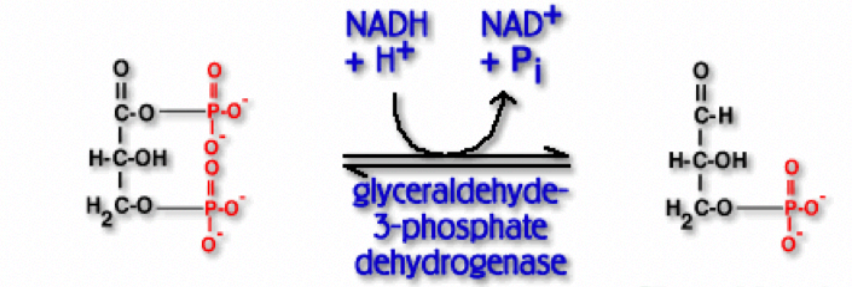


3-Phosphoglycerate

1,3-Bisphosphoglycerate

Reverse of Glycolysis reaction.

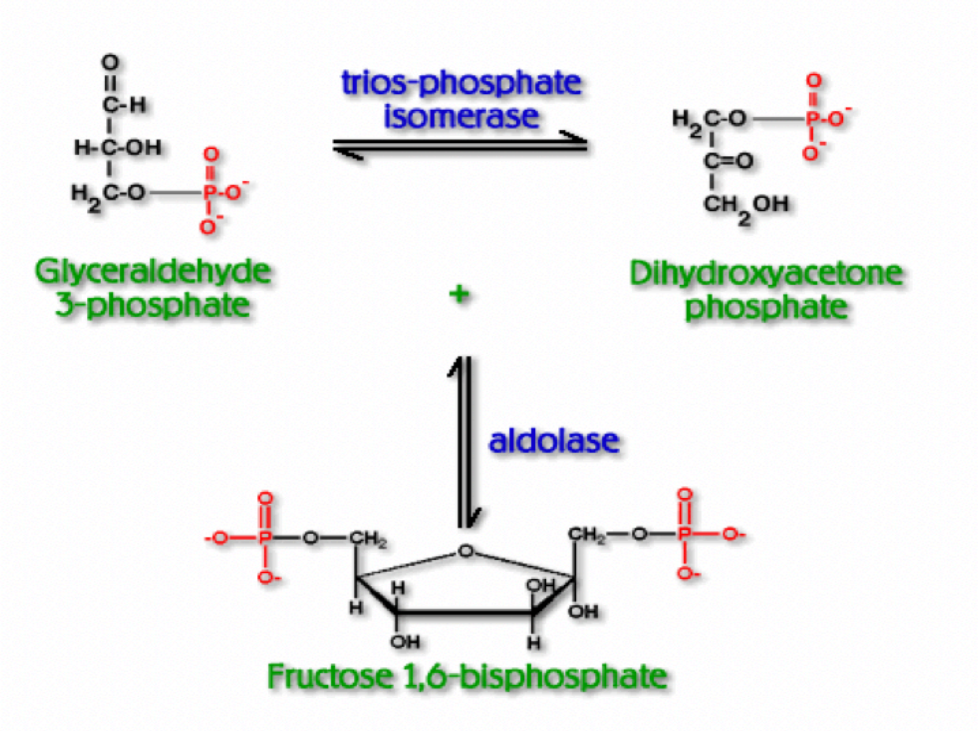
Energy is required (as ATP) for this step.



1,3-Bisphosphoglycerate

Glyceraldehyde
3-phosphate

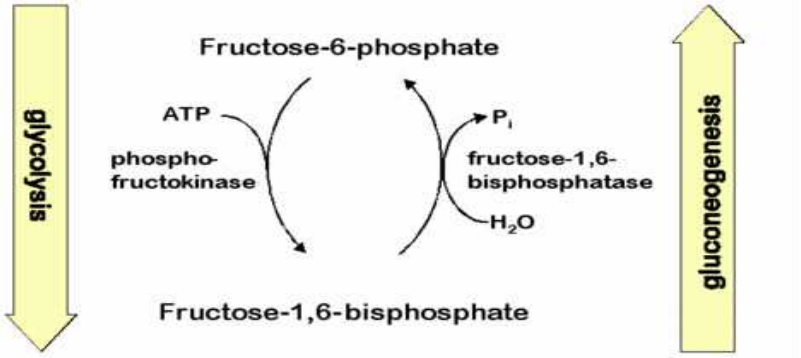
Reverse of Glycolysis reaction.

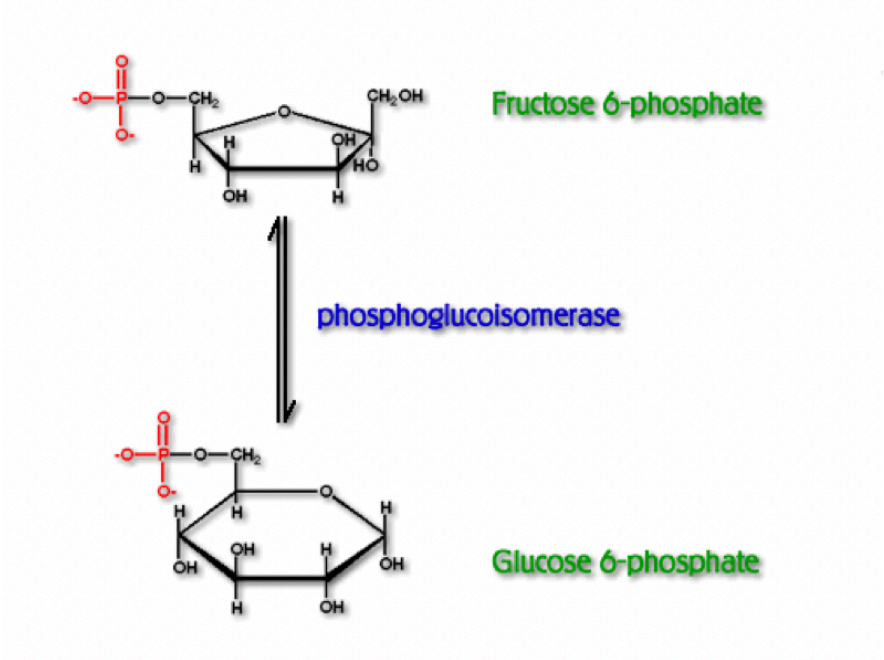


Conversion of Fructose 1,6-Bisphosphate to Fructose 6-Phosphate Is the Second Bypass

- phosphorylation of fructose 6- phosphate by PFK-1 to fructose 1,6-bisphosphate is a highly exergonic and therefore irreversible in intact cells
- The generation of fructose 6-phosphate from fructose 1,6-bisphosphate is catalyzed by a different Mg+2-dependent enzyme called **fructose 1,6-bisphosphatase (FBPase-1)**
- Fructose 1,6-bisphosphate +H2O---------> fructose 6-phosphate +Pi
- NO ATP produced instead hydrolysis of C-1 phosphate

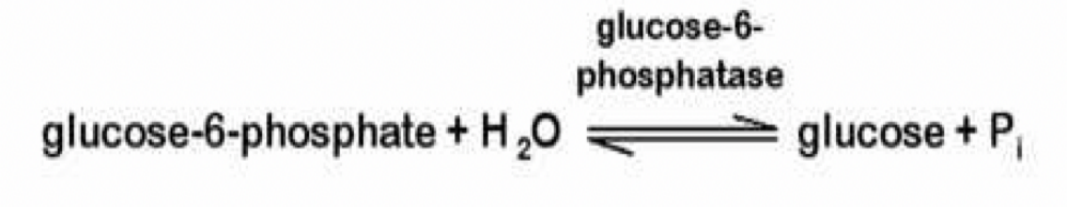
Gluconeogenesis

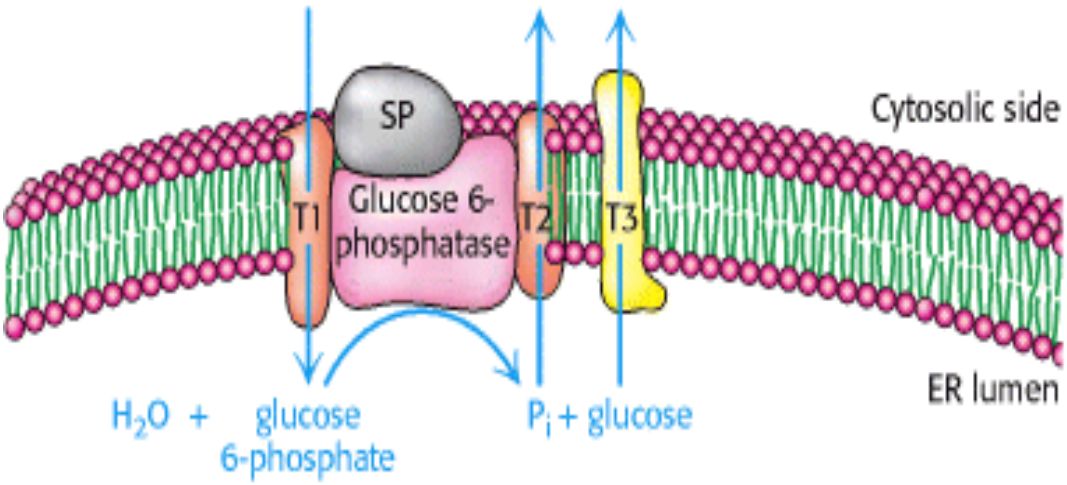


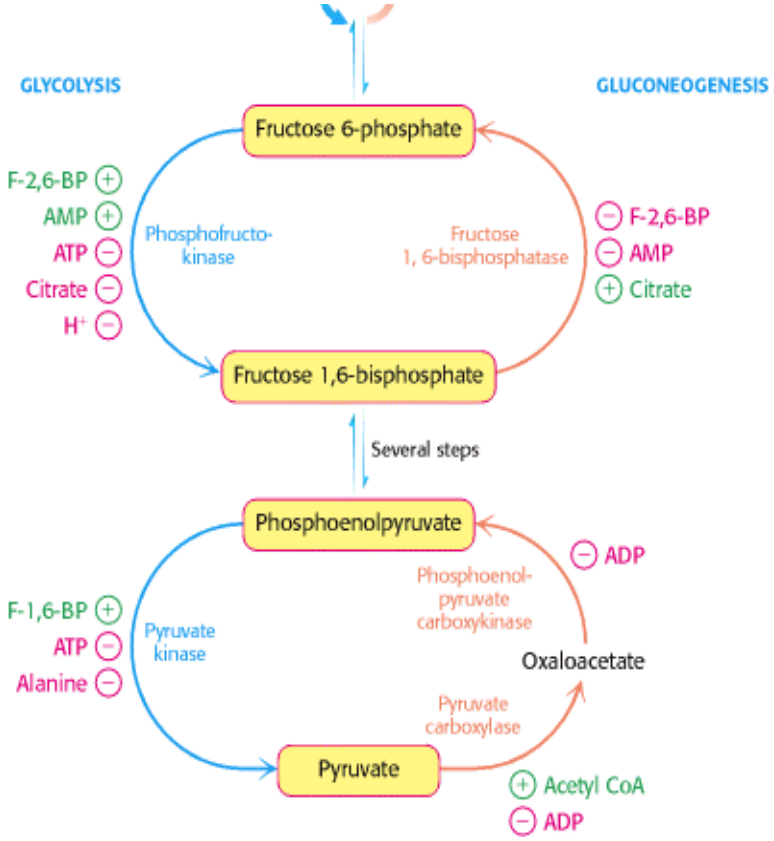


Third Bypass: conversion of Glucose 6-Phosphate to Glucose

- Dephosphorylation of glucose 6-phosphate to yield glucose
- phosphoryl group transfer from glucose 6-phosphate to ADP, forming ATP, an energetically unfavorable reaction
- Catalyzed by **glucose 6-phosphatase** does not require synthesis of ATP; it is a simple hydrolysis of a phosphate
- Mg+2-activated enzyme found on the lumenal side of the endoplasmic reticulum of hepatocytes and renal cells
- ınot carry





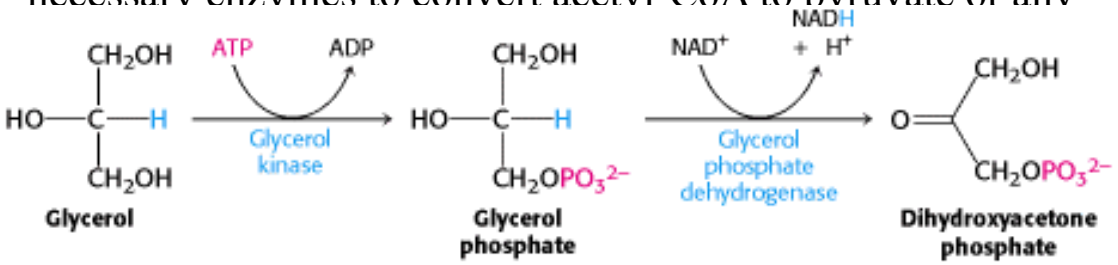


Lactate Utilization

- Lactate is produced by red blood cells continuously and by skeletal muscle during strenuous physical exertion. The majority of lactate produced is released into the blood, where it travels to the liver for conversion to glucose via gluconeogenesis
- The newly made glucose can, in turn, be released into the blood.
- muscle cells lack glucose-6-phosphatase and cannot produce free glucose from noncarbohydrate sources.
- The liver is able to prevent the accumulation of lactate while replenishing blood glucose. This is an important relationship between muscle and liver, especially during strenuous (anaerobic) physical activity when blood glucose is being used

Glycerol Utilization

- The hydrolysis of triacylglycerols stored in adipose tissue produces fatty acids and glycerol
- Fatty acids catabolized to acetyl-CoA
- The remaining glycerol molecule is released from adipose tissue into the blood, where it travels to the liver for conversion to glucose via gluconeogenesis.
- Fatty acids cannot be used to make glucose because humans lack the necessary enzymes to convert acetyl-CoA to pyruvate or any



To prevent the waste of a futile cycle, Glycolysis & Gluconeogenesis are **reciprocally regulated**.

Local Control includes reciprocal allosteric regulation by **adenine nucleotides**.

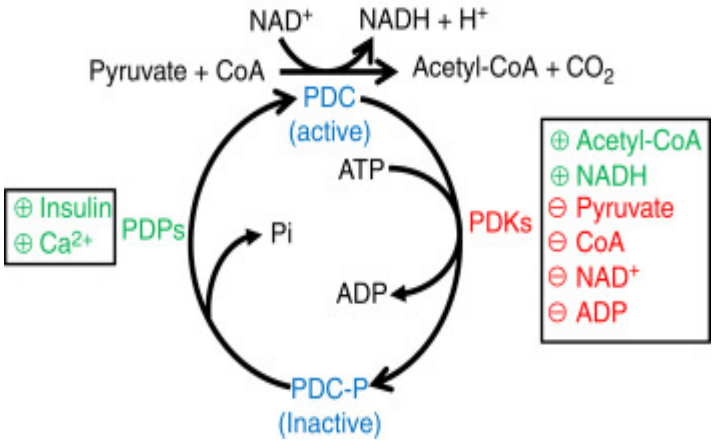
- **Phosphofructokinase** (Glycolysis) is inhibited by ATP and stimulated by AMP.
- **Fructose-1,6-bisphosphatase** (Gluconeogenesis) is inhibited by AMP.

This insures that when cellular ATP is high (AMP would then be low), glucose is not degraded to make ATP.

When ATP is low (AMP would then be high), the cell does not expend energy in synthesizing glucose

does not expend energy in synthesizing glucose

- When fatty acids are readily available as fuels, their breakdown in liver mitochondria yields acetyl-CoA, a signal that further oxidation of glucose for fuel is not necessary.
- Acetyl-CoA is a positive allosteric modulator of pyruvate carboxylase and a negative modulator of pyruvate dehydrogenase, through stimulation of a protein kinase that inactivates the dehydrogenase



- When the cell's energetic needs are being met, oxidative phosphorylation slows, NADH rises relative to NAD+ and inhibits the citric acid cycle, and acetyl-CoA accumulates.
- Increased concentration of acetyl-CoA inhibits the pyruvate dehydrogenase complex, slowing the formation of acetyl-CoA from pyruvate, and stimulates gluconeogenesis by activating pyruvate carboxylase, allowing excess pyruvate to be converted to glucose.

- The second control point in gluconeogenesis is the reaction catalyzed by FBPase-1
- fBPase-1 strongly inhibited by AMP. when sufficient concentrations of acetyl-CoA or citrate (the product of acetyl- CoA condensation with oxaloacetate) are present, or when a high proportion of the cell's adenylate is in the form of ATP, gluconeogenesis is favored.

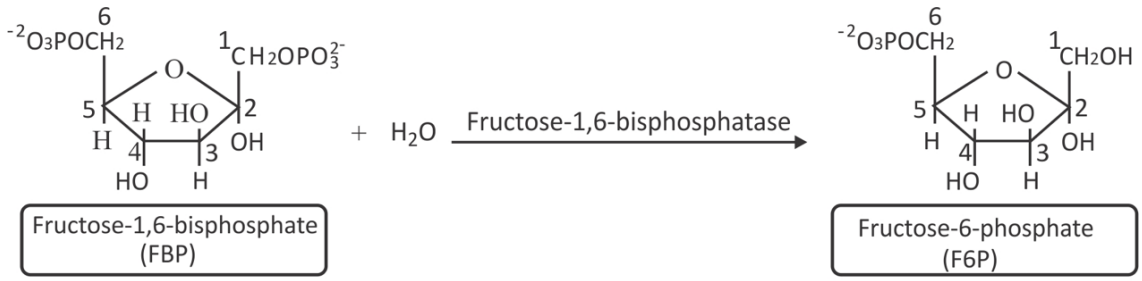
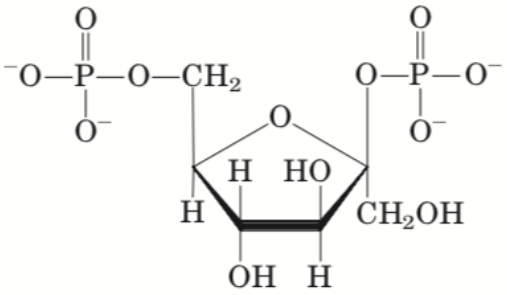


Fig. : Conversion of fructose-1,6-bisphosphate to fructose-6-phosphate.

Role of liver in maintaining a constant blood glucose level

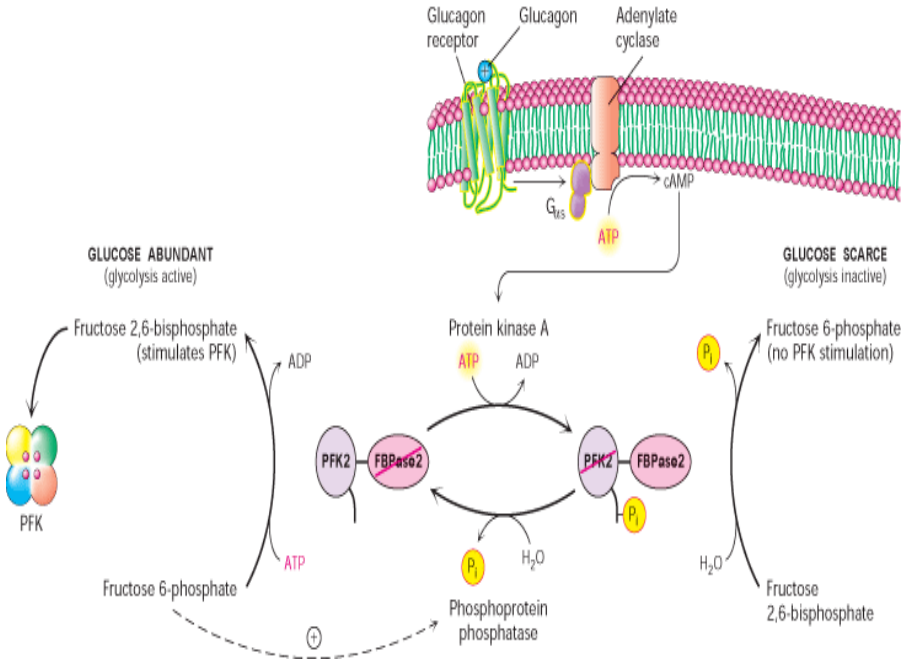
- When the blood glucose level decreases, the hormone **glucagon** signals the liver to produce and release more glucose and to stop consuming it for its own needs.
- One source of glucose is glycogen stored in the liver; another source is gluconeogenesis.
- fructose 2,6-bisphosphate binds to its allosteric site on PFK-1, it increases that enzyme's affinity for its substrate **(Glycolysis)**
- PFK-1 is virtually inactive in the absence of fructose 2,6-bisphosphate. Fructose 2,6-bisphosphate *activates* PFK-1 stimulating glycolysis in liver and,
- At the same time, *inhibits* FBPase-1, thereby slowing gluconeogenesis.

- Fructose 2,6-bisphosphate is not an intermediate in gluconeogenesis or glycolysis; it is a *regulator*



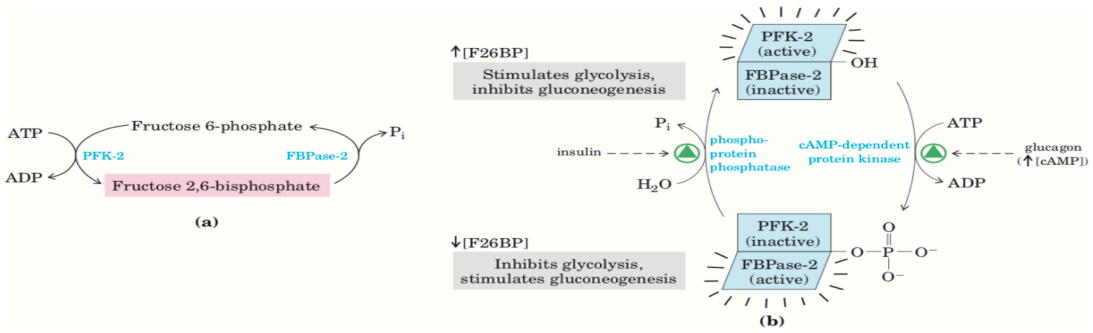
Fructose 2,6-bisphosphate

- Glucagon stimulates the adenylyl cyclase of liver to synthesize 3',5'-cyclic AMP (cAMP) from ATP. Then cAMP activates cAMP-dependent protein kinase, which transfers a phosphoryl group from ATP to the bifunctional protein PFK-2/FBPase-2. Phosphorylation of this protein enhances its FBPase-2 activity and inhibits its PFK-2 activity. Glucagon thereby lowers the cellular level of fructose 2,6-bisphosphate,



inhibiting glycolysis and stimulating gluconeogenesis.

• Insulin has the opposite effect, stimulating the activity of a phosphoprotein phosphatase that catalyzes removal of the phosphoryl group from the bifunctional protein PFK-2/FBPase-2, activating its PFK-2 activity, increasing the level of fructose 2,6-bisphosphate, stimulating glycolysis, and inhibiting gluconeogenesis.



Key Concepts in regulation of CHO metabolism

• ***AMP, ADP, and ATP as Allosteric Modulators***

Positive modulation by AMP: An increase in AMP (or ADP) concentration signifies a depletion of ATP and the need to produce more of this energy source- positively modulate energy releasing pathways

Examples:

1. positive regulator of phosphorylase (an enzyme used in glycogenolysis)

2. Activation of phosphofructokinase

- **ATP—negative regulator of**
  - Pyruvate dehydrogenase complex
  - Citrate synthase
  - Isocitrate dehydrogenase.
- ***NADH/NAD+ Ratio***
- NADH is a product of glycolysis. Its buildup would indicate the pathway is not needed to produce additional ATP. In the fasted state, the liver typically has more NAD+ because doing gluconeogenesis, muscle will be utilizing glucose—less NAD+/NADH ration
- **Covalent Regulation**
- **Directional Shifts in Reversible Reactions** (The concentration of the reactants and products in the cell)
- **Genetic Regulation**
  - The abundance of an enzyme can be either induced or suppressed(Gene expression may be influenced by hormones, dietary intake, environment)