GLUCONEOGENESIS

GLUCONEOGENESIS (PRODUCTION OF GLUCOSE FROM NON-CARBOHYDRATES PRECURSORS)

Gluconeogenesis is the reversible pathway of the glycolysis, when we talk about gluconeogenesis we are talking about fast condition for a long time which mean that we don't have a diet , glycogen is consumed , we know that brain , RBC , medulla are dependent on glucose and can't synthesis it so we need to supply them with glucose

We say that our body can store around 500 g of glycogen , one of the reasons that we store small of amount of glycogen is for the hydrophilic nature of the glycogen (then the water will go in the direction of the hydrophilic glycogen then the size of our body will be very large with excessive of fluids ,remember that glucose used in measuring the osmolarity then storage of glucose will change the osmolarity then changing the movement of water and the osmotic pressure) , other reason is the amount of energy in this case fatty acids will give us large amounts of energy so fatty acids is more suitable as a storage form of energy (1g of fatty acids will give 9kcal , 1g of glucose will give 4kcal)

- **BRAIN IS DEPENDENT ON GLUCOSE 120G/DAY**
- **BODY GLUCOSE RESERVE IS LIMITED**

≈ 20 G (EXTRA CELLULAR FLUID) (MODIFIED GLUCOSE IN THE GAGS)

≈ 75 G (LIVER GLYCOGEN); ENOUGH FOR 16 HOURS

≈ 400 G (MUSCLE GLYCOGEN); FOR MUSCLE USE ONLY

MAIN SOURCE OF ENERGY FOR RESTING MUSCLE IN POST-ABSORPTIVE STATE

- **70 KG MAN HAS ≈ 15 KG FAT**

- FATTY ACIDS CAN NOT BE CONVERTED TO GLUCOSE

- UTILIZATION OF FA IS INCREASED 4-5 X IN PROLONGED FASTING

- IN PROLONGED FASTING; FA KETONE BODIES AT HIGH RATE

The storage form of the fats in the adipose tissue must be triacylglycerol inside adipocyte fat droplets , triacylglycerol consist of

1-glycerol

2-fatty acids

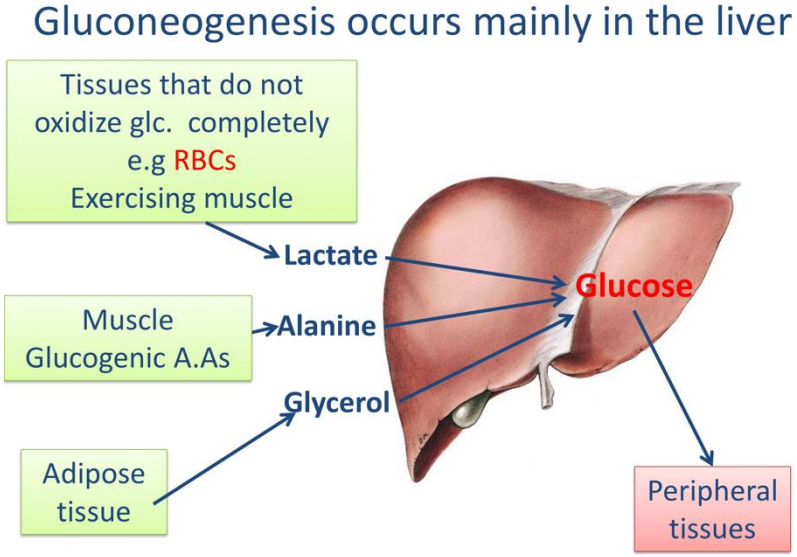
Hormones have receptors of the adipose tissue , now in the fast conditions the changes in the hormones will change the enzymes activity inside the adipose tissue , we start to degrade fatty acids from the adipose tissue and release them to blood stream , size of the fatty acids may determine the way of transporting to the blood , small and medium fatty acids chains with solubility can be released without carriers but for large chain we carry the chains by albumins until reaching the cells.

We need to know that fatty acids is used as source of energy and we don't use them to synthesis glucose, the remain part of the triacylglycerol is the glycerol and this part is used for gluconeogenesis (glycerol is non carbohydrate residue)

Fatty acids produce huge amounts of acetyl coA because most common fatty acids contains 16-18 carbons, acetyl coA consist of 2 carbons then we can use each fatty acids to produce 8-9 acetyl coA which is larger than the amount that glucose can produce (2)

Acetyl coA needs oxaloacetate to run the Krebs cycle , but in these conditions we don't have enough amounts of oxaloacetate (OAA is consumed in these conditions) then acetyl coA accumulate and result in ketogenesis or ketone bodies formation

- **WHERE AND WHEN DOES GLUOCNEOGENSIS OCCUR?**
- **DURING AN OVERNIGHT FAST, ~ 90% OF GLUCONEOGENESIS OCCURS IN THE LIVER** (HYPATOCYTES) **AND 10% BY THE KIDNEYS**
- **DURING PROLONGED FASTING KIDNEYS BECOME MAJOR GLUCOSE-PRODUCING ORGANS (40% OF TOTAL GLUCOSE PRODUCTION)**



We will use all available source such as lactate (mostly in exercising muscles which will go through anaerobic glycolysis , and RBC), remember that gluconeogenesis take place in kidney and liver only

We can also use glucogenic amino acids (like alanine which came from muscles) , amino acids metabolism leads to alpha keto acids formation (such as pyruvate)

Amino acids can be

1-Glucogenic amino acids : amino acids which can be metabolised into Krebs cycle intermediates or pyruvate and used these can be used in gluconeogenesis

2-Ketogenic amino acids : amino acids which can be metabolised into acetyl coA or Acetoacetyl-CoA which mean that they can used in synthesis of ketobodies

3- glucogenic + ketogenic (the major amino acid used is alanine)

We said that we can degrade fats by breaking ester bonds and using glycerol for gluconeogenesis precursor , producing glucose go from the liver to the blood stream and distribute to the tissues according to the priority , small fraction of glucose remain in the bloodstream

Generally gluconeogenesis is the opposite of glycolysis

According to the substrates we use in the gluconeogenesis we have many entry points , if the substrate is glycerol (3C alcohol) then usually we will go in trioses pathway

For amino acid , amino acids can enters as oxaloacetate , pyruvate , lactate and another Krebs cycle intermediates .

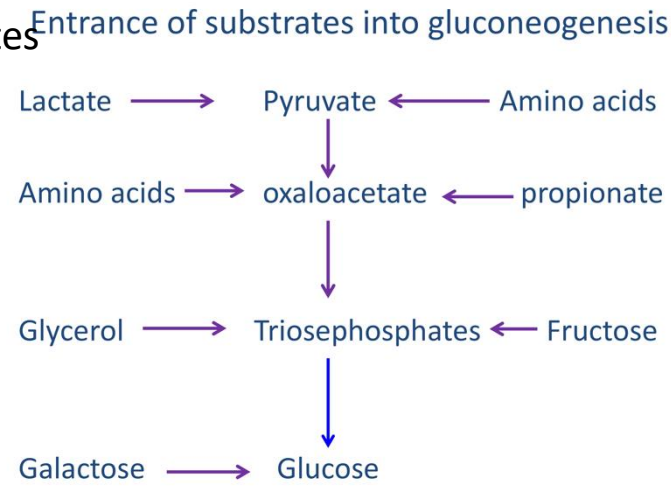
Fructose and galactose are carbohydrates

So we can't use them as precursors in

Gluconeogenesis , but we can synthesis

them by non carbohydrates precursors

Then convert them to glucose



Galactose (aldose) is more similar to glucose (aldose) than fructose(ketose).

Fructose can enter the pathway as triose and we will discuss this in the next lectures.

GLUCONEOGENESIS IS OPPOSITE TO GLYCOLYSIS BUT ?

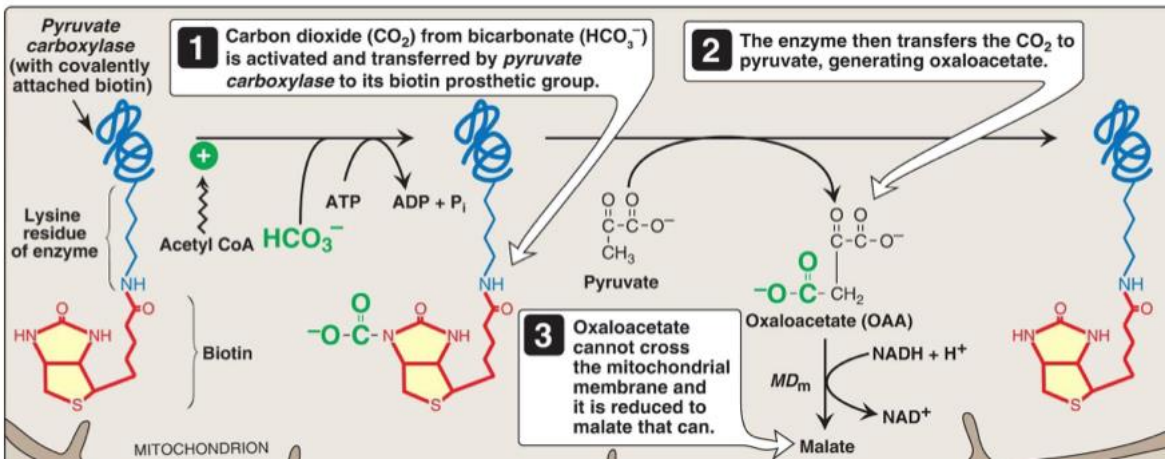
But we have 3 irreversible reactions , irreversible reactions in biochemistry means that reaction can't occur in the opposite way by the same enzyme then we need to use different enzymes , we won't discuss the 7 reversible steps

First step in glycolysis will be the last step in gluconeogenesis

These irreversible steps

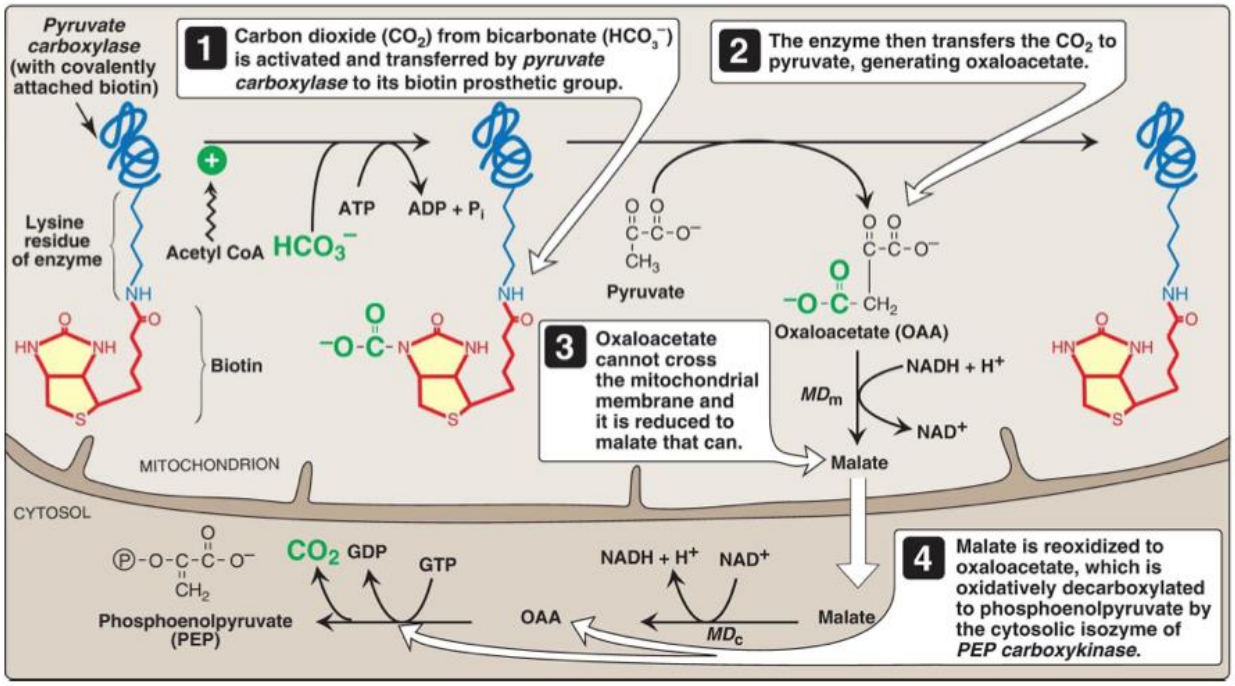
1-FROM PYRUVATE TO PEP

Pyruvate exist in the cytosol where glycolysis occur , now we convert pyruvate to acetyl coA to run the Krebs cycle (in the well fed state) , so pyruvate entered the mitochondria then the first step is converting pyruvate to oxaloacetate but pyruvate contain 3C and oxaloacetate contains 4C so it's a carboxylation reaction by pyruvate carboxylate (carboxylases need ATP) , so we convert oxaloacetate then we need to convert oxaloacetate to PEP and this reaction occur outside the mitochondria then we need to transport oxaloacetate outside it , but we know that can't cross the mitochondrial membrane and don't have any transporter so we need to convert oxaloacetate to another molecule which has a transporter like malate , so we use Krebs cycle enzymes to reverse this reaction , in Krebs cycle we oxidized malate to oxaloacetate but now we reduce oxaloacetate into malate (by oxidation of NADH)



After reducing oxaloacetate to malate , malate cross the mitochondrial membrane now we don't ever need malate so we use malate dehydrogenase and end up with oxaloacetate (by reduce NAD+), after that we want to reach the form of PEP (3C and 1 phosphate) we need to decarboxylase + phosphorylation the oxalacetate , we use GTP but we are using 2 molecules of GTP because we want to end up by 2PEP so we are in synthetic pathway and using energy during fast conditions.

Acetyl coA is an activator of Pyruvate carboxylase, in this case large acetyl coA which won't be used then this pathway occurs to produce oxaloacetate



- **BY PYRUVATE CARBOXYLASE**
- **IN MITOCHONDRIA**
- **ALLOSTERICALLY ACTIVATED BY COA**
- **ENZYME IS FOUND IN BOTH CYTOSOL AND MITOCHONDRIA**
- **THE GENERATED PEP IN THE MITOCHONDRIA IS TRANSPORTED TO THE CYTOSOL BY A SPECIFIC TRANSPORTER**
- **THE PEP THAT IS GENERATED IN THE CYTOSOL REQUIRES THE TRANSPORT OF OAA FROM THE MITOCHONDRIA TO THE CYTOSOL**

Remember that we have additional step relative to conversion of pyruvate to oxaloacetate , two steps to reverse the last step of glycolysis (first step in gluconeogenesis)

(If you are asking why we need to enter the mitochondria to convert pyruvate into oxaloacetate , it's related to the enzymes we need to use in each step)

2-FROM FRUCTOSE 1,6 BISOPHOSPHATE TO FRUCTOSE 6-PHOSPHATE

• DEPHOSPHORYLATION OF FRUCTOSE 1,6-BISPHOSPHATE

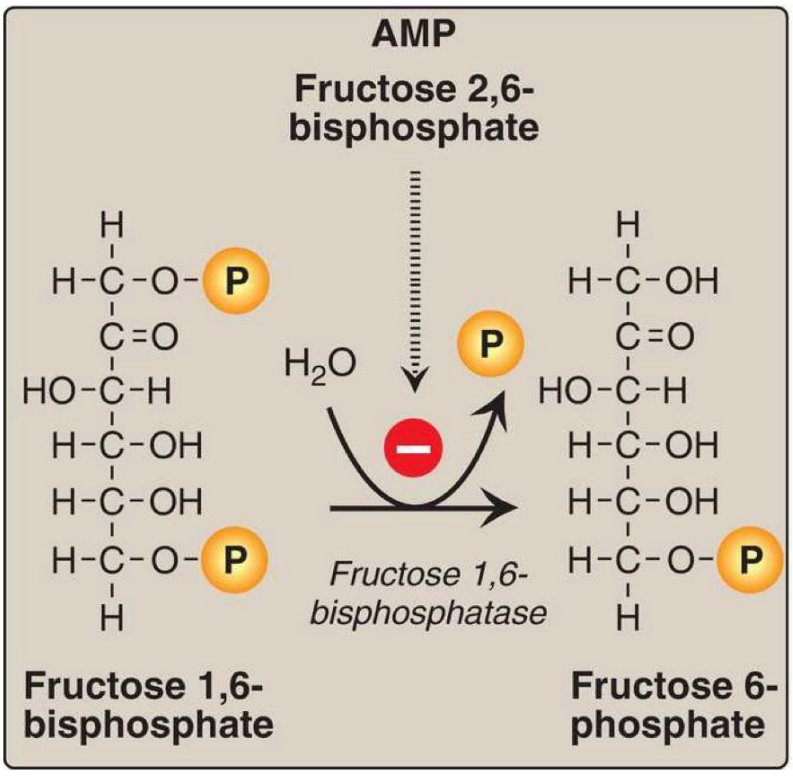
• THIS REACTION BYPASSES THE IRREVISIBLE PHOSPHOFRUCTOKINSE -1

In glycolysis we add phosphate by phosphofructokinase-1 so now we need to use phosphatase (one step), phosphatase can be inhibited by

1 -AMP

2-fructose 2,6-bisphosphate

(remember that both of them activate phosphofructokinase-1) , so in the same molecule we have opposite pathways



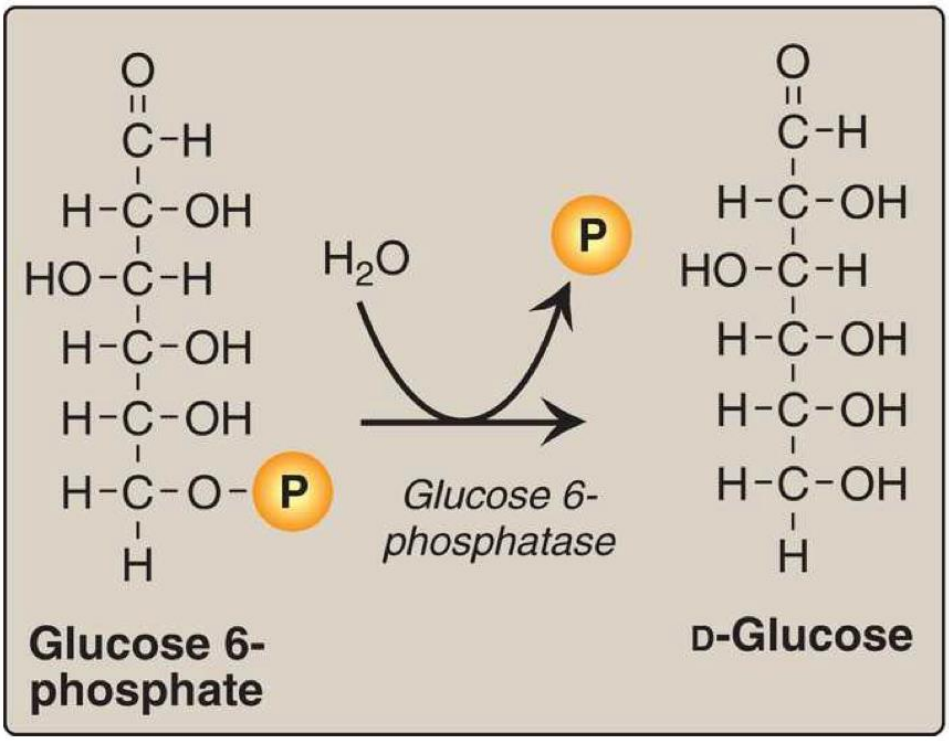
3-FROM GLUCOSE 6-PHOSPHATE TO GLUCOSE

- **DEPHOSPHORYLATION OF GLUCOSE 6-PHOSPHATE**
- **BYPASSES THE IRREVERSIBLE HEXOKINASE REACTION**
- **ONLY IN LIVER AND KIDNEY**
- **GLUCOSE 6-PHOSPHATE**
- **TRANSLOCASE IS NEEDED TO TRANSPORT G-6-P ACROSS THE ER MEMBRANE**
- **GLUCOSE 6-PHOSPHATASE IN ENDOPLASMIC RETICULUM (ER)**

HINT: MUSCLE LACKS GLUCOSE 6-PHOSPHATASE, AND THEREFORE MUSCLE GLYCOGEN CAN NOT BE USED TO MAINTAIN BLOOD GLUCOSE LEVELS.

Also we need to use phosphatase, this phosphatase called glucose 6-phosphatase this enzyme found only in liver and kidney (not in muscles)

Glucose 6-phosphate exist in the cytosol and glucose 6-phosphatase exist in the ER so we need to move glucose 6-phosphate by using translocase (glut only transport glucose) , so translocase let the glucose 6-p enter the ER and become glucose then glucose leave by glut-7 (transporter not translocase) then glucose reach the cytosol and then on the cell membrane other gluts (glut 2) can transport glucose , then glucose is released to the blood stream



Dephosphorylation of glucose 6-phosphate to glucose, delta G of the reaction in negative so we can run the reaction

Phosphorylation of glucose to glucose 6-phosphate, the delta G of this reaction is positive (without ATP hydrolysis) so we had to couple the reactions, ATP hydrolysis and phosphorylation of glucose then when we add two reactions together then the over all delta G is negative and that's why we can run the reaction

Formation vs. Hydrolysis of Glucose 6-phosphate

- Formation

$$\text{Glc.} + \text{Pi} \longrightarrow \text{Glc. 6-phosphate} + H_2O \quad \Delta G = +ve$$

$$\text{ATP} + H_2O \longrightarrow \text{ADP} + P_i \quad \Delta G = -ve$$

Hexokinase

$$\text{Glc.} + \text{ATP} \longrightarrow \text{Glc. 6-phosphate} + \text{ADP} \quad \Delta G = -ve$$

- Hydrolysis

Phosphatase

$$\text{Glc. 6-phosphate} + H_2O \longrightarrow \text{Glc.} + P_i \quad \Delta G = -ve$$

•ENERGY RECUIREMENTS OF GLUCONEOGENESIS

Up to down (glycolysis) (10 steps)

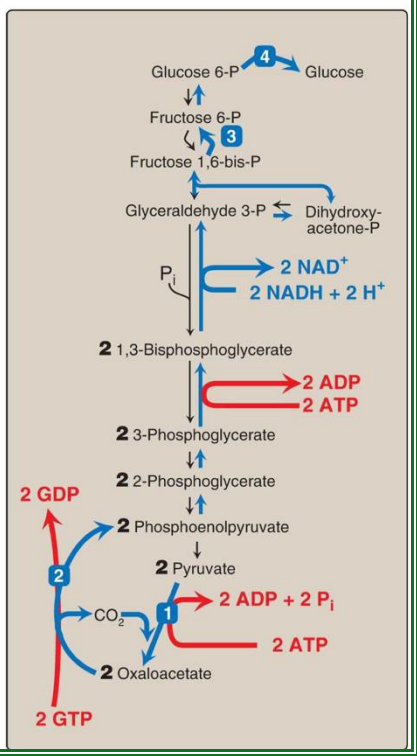
Down to up (gluconeogenesis) (11 steps)

Blue for NADH

Red for ATP

In glycolysis we use 1ATP in (glucose to glucose 6-p)

1ATP in (fructose 6-p to fructose 1,6-biosp), But in Gluconeogenesis we produce inorganic phosphate rather than ATP , and consumed 4 ATP and 2GTP (We can say 6 ATP) and 2NADH , 2ATP (pyruvate to oaa)



2GTP (oaa to pep) , 2ATP (3-phosphoglycerate to 1,3-bisphosphoglycerate)

so the net consumed are 4ATP , 2GTP , 2NADH (remember that we use 2 pyruvate molecules to produce 1 glucose , that's why we need to consume 2ATP&2GTP&2NADH)

we are in fasting state, and we consumed energy (we use fatty acids as a source of energy) , but we produce glucose for the tissues that depend on it as a source of energy (like brain) and to balance the osmotic pressure

(أَلَمْ تَرَ أَنَّ اللَّهَ أَنْزَلَ مِنَ السَّمَاءِ مَاءً فَأَخْرَجْنَا بِهِ ثَمَرَاتٍ مُخْتَلِفًا أَلْوَانُهَا ۚ وَمِنَ الْجِبَالِ جُدَدٌ بِيضٌ وَحُمْرٌ مُخْتَلِفٌ أَلْوَانُهَا وَغَرَابِيبُ سُودٌ ﴿27﴾ وَمِنَ النَّاسِ وَالدَّوَابِّ وَالْأَنْعَامِ مُخْتَلِفٌ أَلْوَانُهُ كَذَٰلِكَ ۗ إِنَّمَا يَخْشَى اللَّهَ مِنْ عِبَادِهِ الْعُلَمَاءُ ۗ إِنَّ اللَّهَ عَزِيزٌ غَفُورٌ ﴿28﴾) صدق الله العظيم

In the previous lecture:

Gluconeogenesis is synthesis of glucose molecules from non-carbohydrate precursors (lactate, glycerol, glucogenic amino acids " amino acids when degraded it produce pyruvate or kreps cycle intermediate").

It is the opposite of the glycolysis except in the three irreversible steps, it needs different enzymes:

Glucokinase or hexokinase ------ glucose 6- phosphatase (in single step)

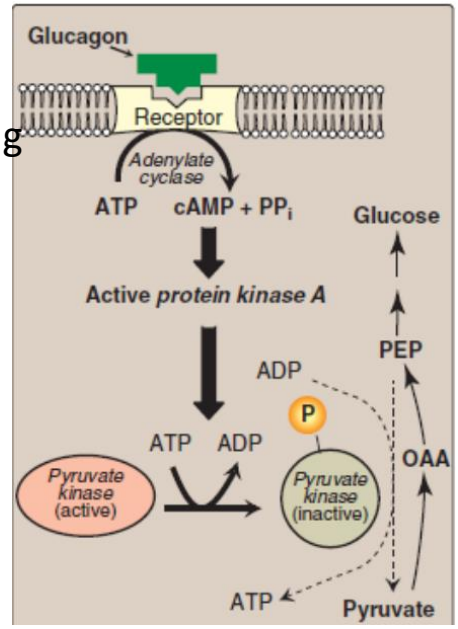
Phosphofructokinase ----- phosphatase (in single step)

Pyruvate kinase ----- pyruvate carboxylase / PEP carboxykinase (two steps)

This process is active under fasting condition which means that **GLUCAGON** is in high levels.

Sooo, let's start our lecture…

REGULATION OF GLUCONEOGENESIS



Glucagon binds to its GPCR, and this activates

Adenylyl cyclase converting ATP to cAMP→activating

Protein kinase A.

STOP, lets remember the targets of this enzyme.

1. Bifunctional enzyme (glycolysis)
2. Glycogen phosphorylase kinase
3. Glycogen synthase
4. Pyruvate kinase

Phosphorylation of pyruvate kinase inhibits it, means inhibition of glycolysis in case of gluconeogenesis, we want to build glucose not to degrade it in fasting condition.

- **regulation Mainly by:**

1. **The circulating level of glucagon**

   - **Glucagon lowers the level of fructose 2,6-bisphosphate** (which is activator of glycolysis)**, resulting in activation of fructose 1,6-bisphosphatase and inhibition of phosphofructokinase 1 (PFK1)**

- **Inhibition of pyruvate kinase**
- **Glucagon increases the transcription of the gene for PEP-carboxykinase** (converts oxaloacetate to phosphoenolpyruvate "PEP")

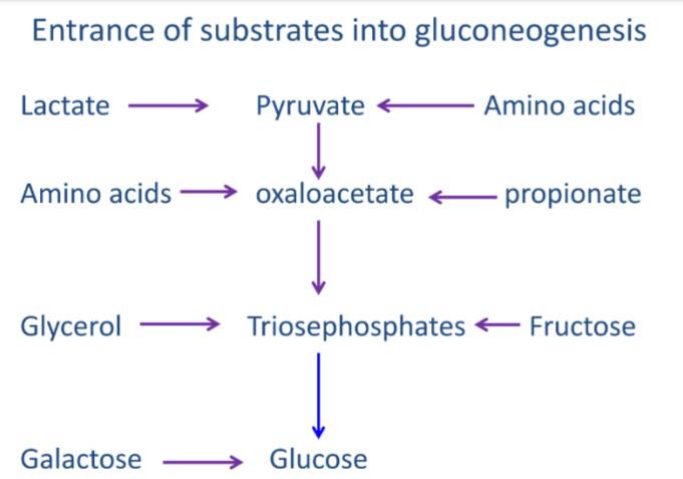
2. The availability of gluconeogenic substrates (the amount of raw materials)

3. Slow adaptive changes in enzyme activity due to an alteration in the rate of enzyme synthesis or degradation, or both

The concentration of enzyme, determined by the rate of expression to its gene, the stability of the mRNA (degraded or not), the rate of synthesis of the enzyme of this mRNA and the rate of degradation of the enzyme. This can be applied in all pathways.

Different precursors enter the Gluconeogenesis process at different stages.

remember this? →



if pyruvate produced of alanine metabolism it will start from the first step.

An amino acid produce oxa. It will start from the second step.

If an amino acid produces succinyl coA, it will proceed in krep's cycle to produce Oxaloacetate and continue from the second step.

Glycerol oxidized to dihydroxyacetone-p and this converts to glyceraldehyde 3-p and continue in the process 😊

that's it for GLUCONEOGENESIS