Biochemistry lec.
2nd stage

by

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Formation of pyruvic acid (P.A.) in the body:

- 1- From oxidation of glucose (glycolysis).
- 2- From lactic acid by oxidation.
- 3- Deamination of alanine.
- 4- Glucogenic amino acids-pyruvate forming.
- 5- Decarboxylation of oxaloacetic acid (OAA)

Fate of pyruvic acid (P.A.):

- 1- Form acetyl CoA by oxidative decarboxylation (in presence of O2)
- 2- Forms lactic acid by reduction (in absence of O2).
- 3- Forms alanine by amination.
- 4- Forms glucose (gluconeogenesis).
- 5- Forms malic acid → to O.A.A (oxaloacetic acid).
- 6- Forms oxaloacetic acid (O.A.A) by CO2-fixation reaction.

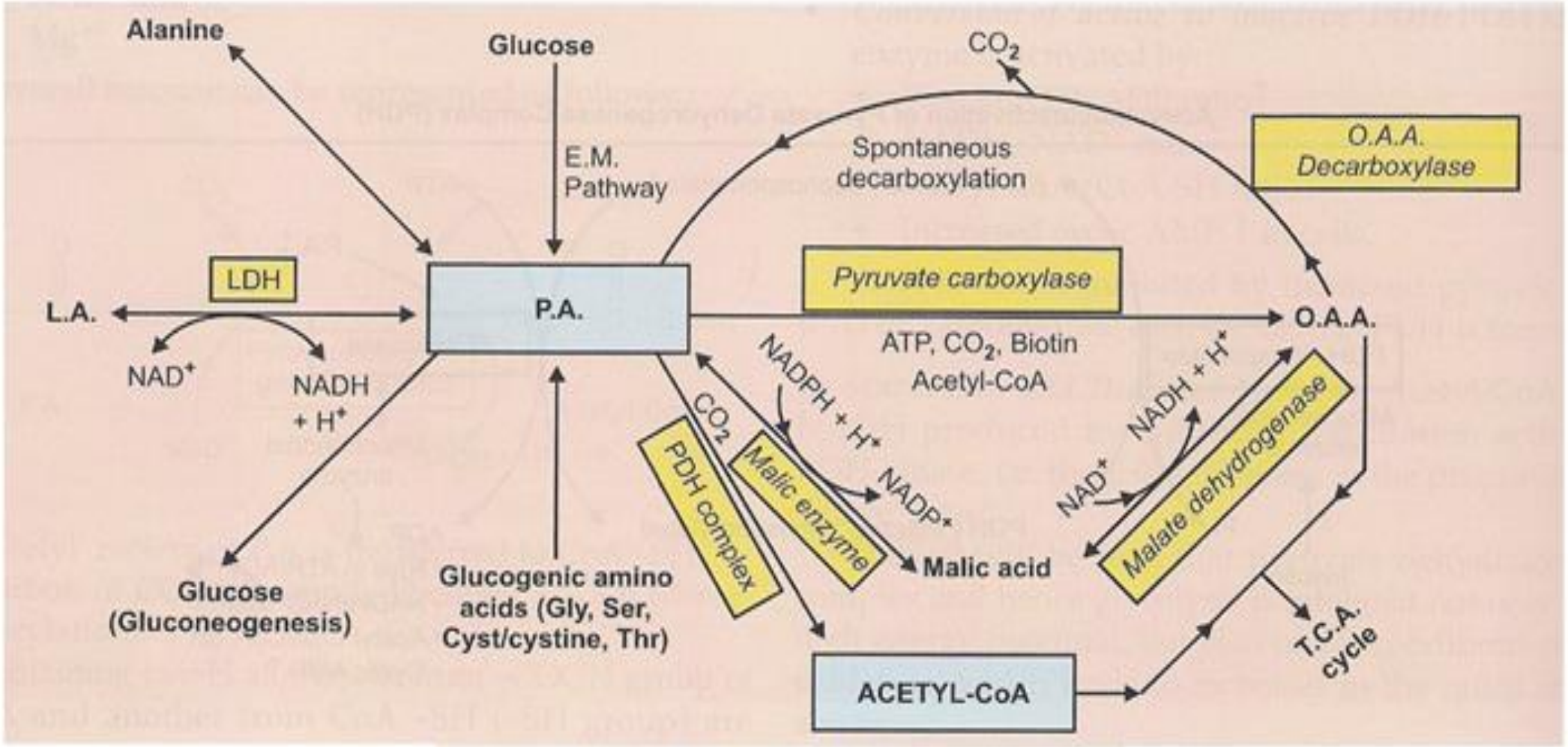


FIG. (1) FORMATION AND FATE OF PYRUVIC ACID

- **Before pyruvate can enter the TCA cycle,** it must be transported into **the mitochondria** via a special pyruvate transporter that aids its passage across the **inner mitochondrial membrane**. Within the mitochondria, pyruvate is oxidatively **decarboxylated to acetyl-CoA**; this reaction is catalyzed by sequentially **multienzyme complex (pyruvate dehydrogenase complex).**

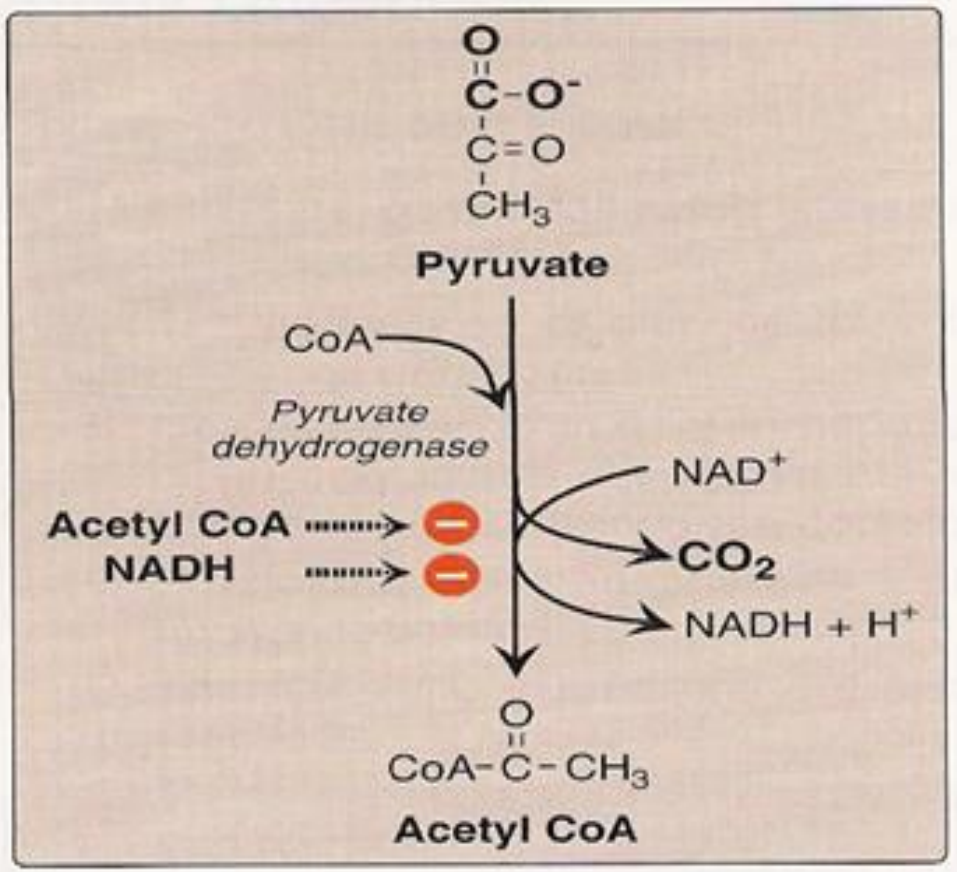


Figure (2)
Oxidative decarboxylation of pyruvate.

The citric acid cycle (TCA) cycle

- 1-TCA cycle (tricarboxylic acid cycle), also known as the citric acid cycle or the Krebs' cycle, is the major energy production pathways in the body. ***The cycle occurs in the mitochondria.**
- 2- It is a cyclic process.
- 3-The cycle involves a sequence of compounds inter-related by **oxidation-reduction** and other reactions which **finally produces [CO2 and H2O].**
- 4- It is the final common pathway of breakdown or catabolism of carbohydrates, fats and proteins.
- **5-Acetyl CoA** derived mainly from oxidation of either **glucose** or **Beta-oxidation of FA** and partly from certain amino acids combines with **oxaloacetic acid** (OAA) to **form "citrate" the first reaction of TCA cycle.**
- 6-By stepwise dehydrogenations and loss of two molecules of CO2, accompanied by internal re-arrangements, the citric acid is reconverted to OAA, which again starts the cycle by taking up another acetyl group from acetyl-CoA.
- 7-All the enzymes of the TCA cycle are in the mitochondrial matrix, except succinate dehydrogenase which is in the inner mitochondrial membrane.
- 8-Electrons are transferred by the cycle to NAD+ (Nicotinamide Adenine Dinucleotide) and FAD (Flavin Adenine Dinucleotide).

- 9-As the electrons subsequently are passed to O2 by the electron transport chain, ATP is generated by the process of oxidative phosphorylation.
- 10-ATP is also generated from GTP (Guanosine Triphosphate), produced in one reaction of the cycle by substrate level phosphorylation.
- 11-The whole process is aerobic, requiring O2 as the final oxidant of the reducing equivalents. Absence of O2 (anoxia) or partial deficiency of O2 (hypoxia) causes total or partial inhibition of the cycle.
- 12-The H atoms removed in the successive dehydrogenations are accepted by corresponding coenzymes. Reduced coenzymes transfer the reducing equivalents to electron-transport system, where oxidative phosphorylation product ATP molecules.

There are three key enzymes in TCA cycle:

- 1-Citrate synthase (1)
- 2- Isocitrate dehydrogenase (I.C.D) (3)
- 3- α-ketoglutarate dehydrogenase (4) or named α-oxoglutarate dehydrogenase

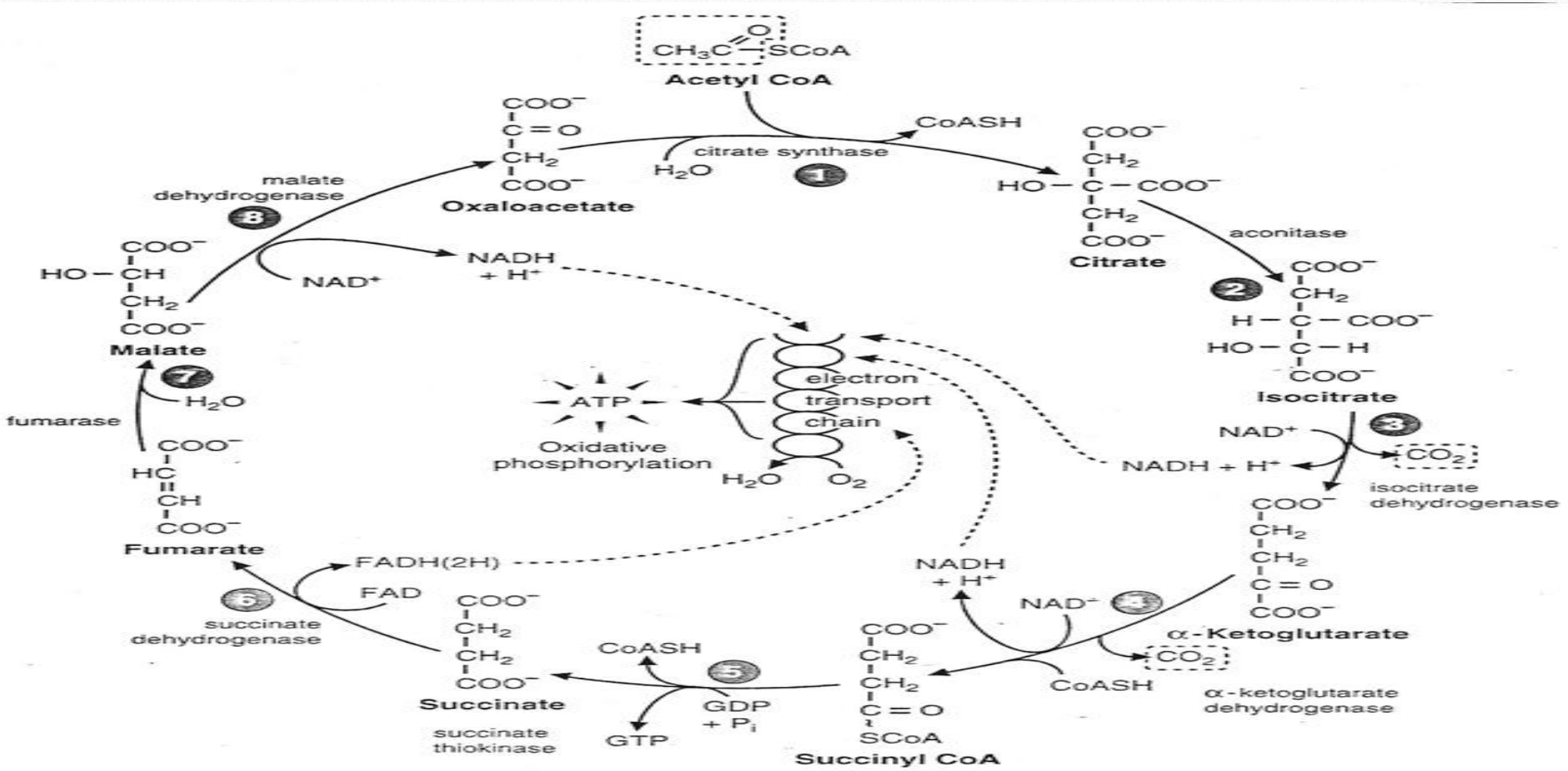
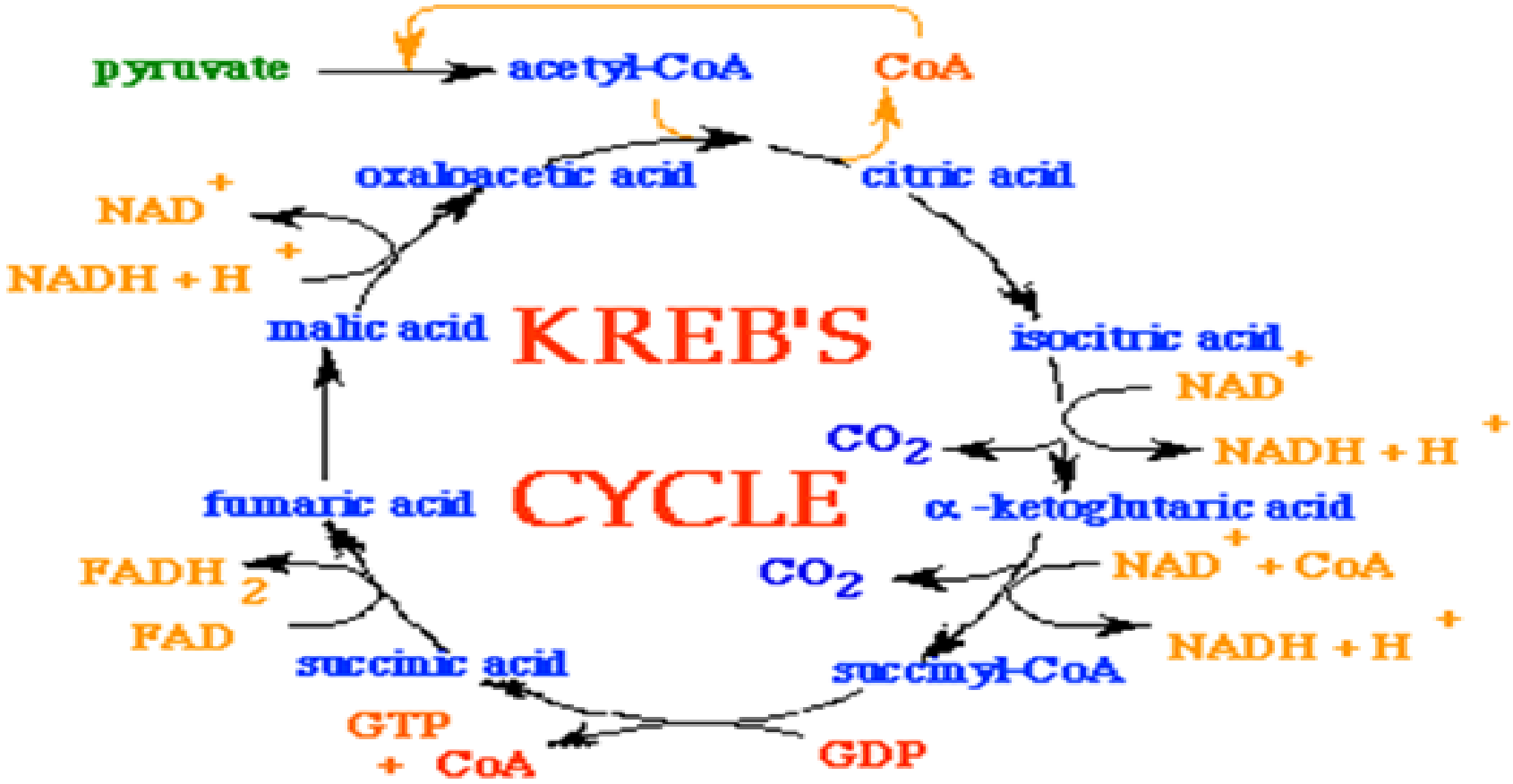


Figure (4) . The tricarboxylic acid (TCA) cycle.


pyruvate
acetyl-CoA
CoA
oxaloacetic acid
citric acid
NAD+
NADH + H+
malic acid
KREB'S
CYCLE
isocitric acid
NAD+
CO2
NADH + H+
fumaric acid
α -ketoglutaric acid
FADH2
FAD
CO2
NAD+ + CoA
NADH + H+
succinic acid
succinyl-CoA
GTP
+ CoA
GDP

Biomedical importance of TCA cycle:

- Final common pathway for carbohydrates, proteins and fats, through formation of 2 – carbon unit acetyl-CoA.
- Acetyl-CoA is oxidized to CO2 and H2O giving out energy (Catabolic role).
- Intermediates of TCA cycle play a major role in synthesis also like heme formation, formation of non essential amino acids, FA synthesis, cholesterol and steroid synthesis (anabolic role).

- **TCA cycle is called Amphibolic in nature because TCA cycle has dual role catabolic and anabolic.**

Energy produced by the TCA cycle:

Energy – producing reaction	*Number of ATP produced*
$3NADH \rightarrow 3NAD^{+}$	**3 x 3 = 9**
$FADH_2 \rightarrow FAD$	**2 x 1 = 2**
$GDP + P_i \rightarrow GTP$	**1 x 1 = 1**
Net gain :	**12 ATP**

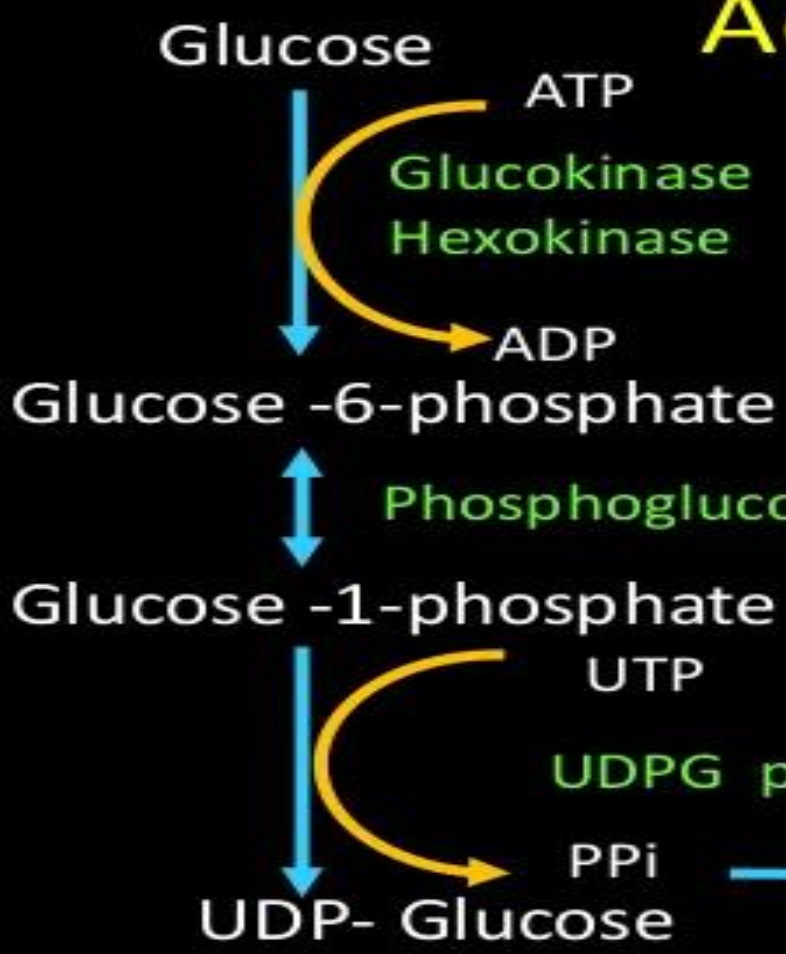
What is Glycogen

- Large, highly branched polymer of glucose
- Mainly contain α - 1—4 glycosidic bonds in straight chain and α - 1 –6 glycosidic bonds at branch points
- Readily mobilized form of glucose
- Broken down when energy is needed.
- Stored in liver and muscle , Liver store more glycogen than muscle

Glycogenesis

- Synthesis of glycogen from glucose
- Occurs in muscle and liver
- Steps:
- 1. Activation of glucose
- 2. Initiation
- 3. Elongation
- 4. Branching of glycogen

Activation of Glucose



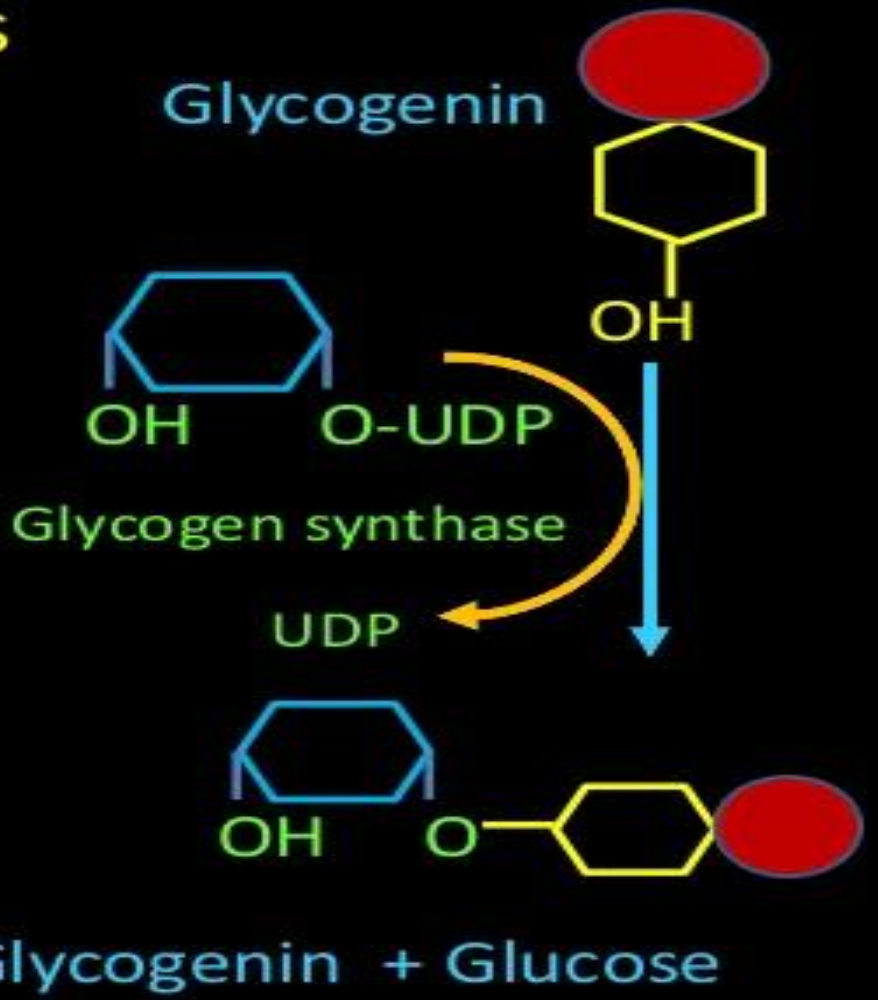
Spontaneous hydrolysis of phosphate bond Provides reaction energy

Only energy cost in glycogen synthesis

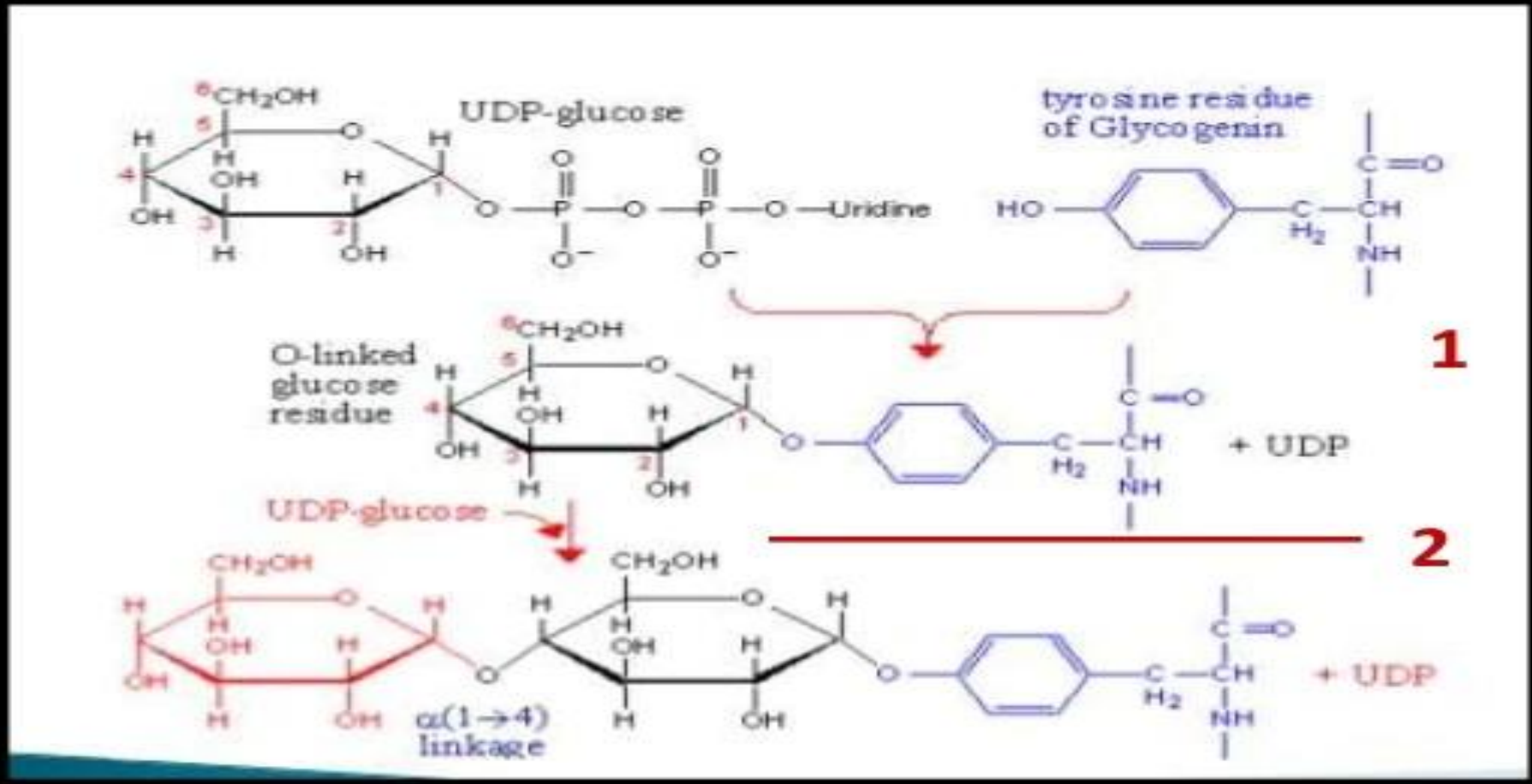
Initiation of Glycogenesis

- Glucose unit is transferred from UDP-Glucose to glycogen primer called glycogenin

- Glycogenin –
- dimeric protein, 37kd
- Protein-carbohydrate complex
- Glycosidic bond is formed between C1 of glucose and hydroxyl group of tyrosine
- UDP is released



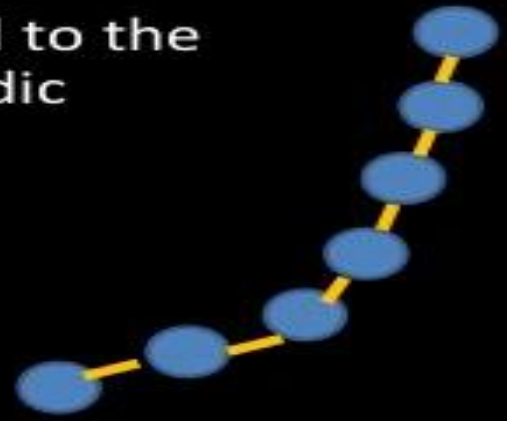
Addition of new glucose on non reducing end

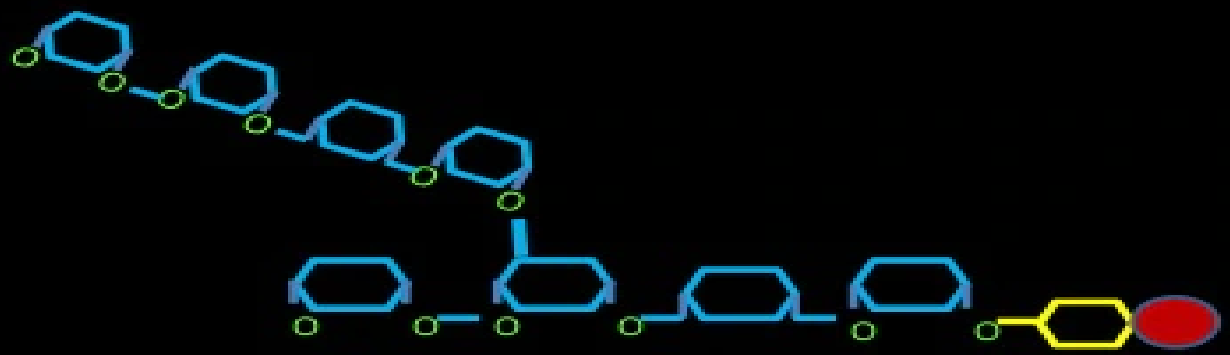


Elongation

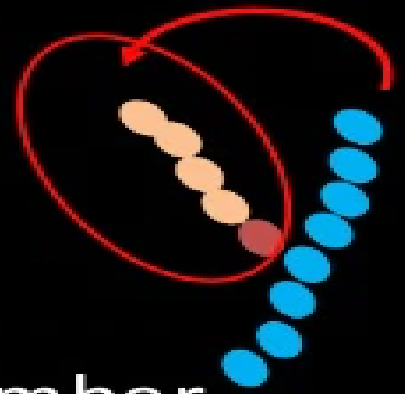


- New glucosyl units are added to the Non-reducing terminal of glycogen
- Active Glucosyl unit UDP-Glucose is transferred to the hydoxyl group of C—4 forming α - 1—4 glycosidic link
- Reaction is catalyzed by glycogen synthase
- Glycogen synthase is Key regulatory enzyme in glycogen synthesis.



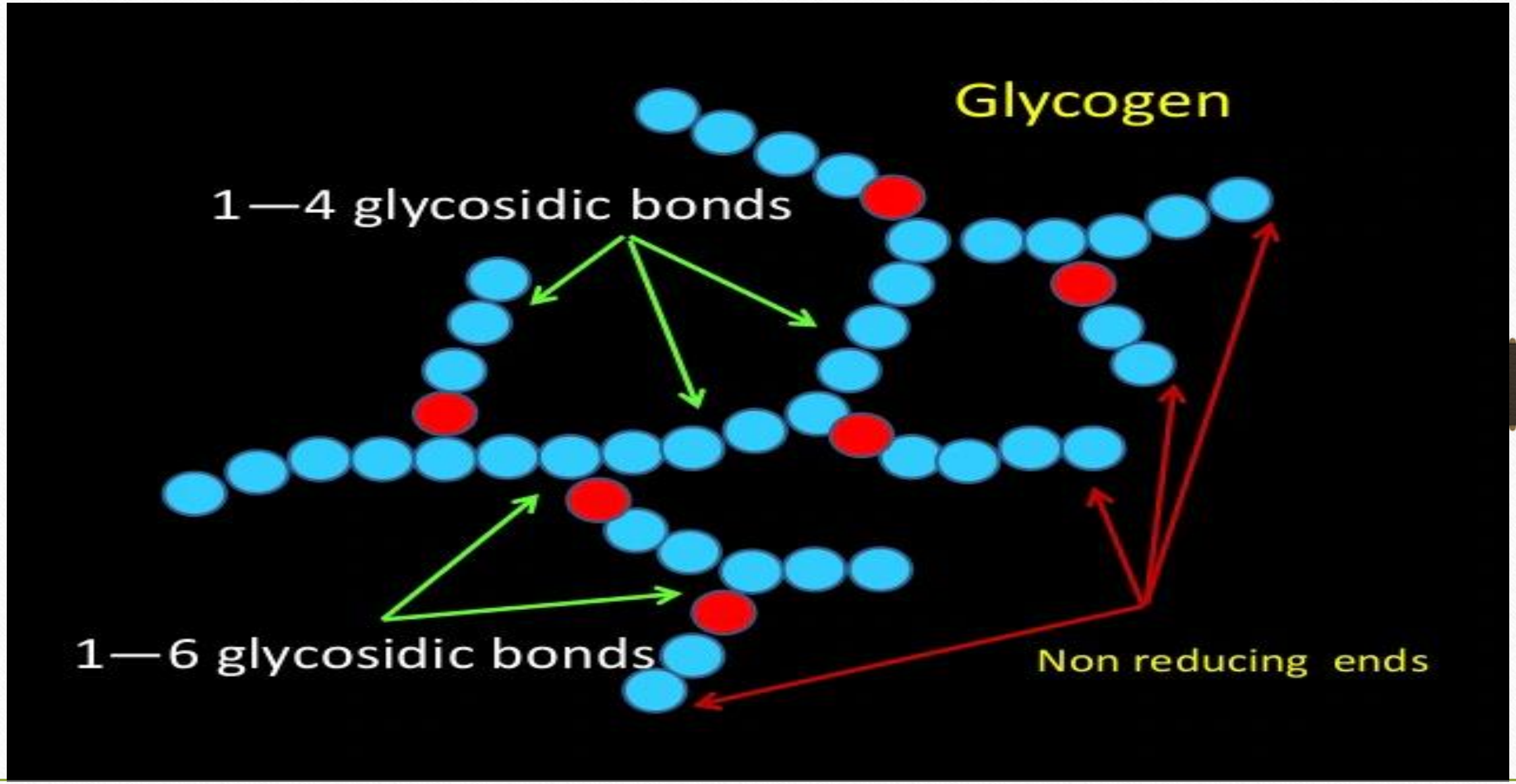


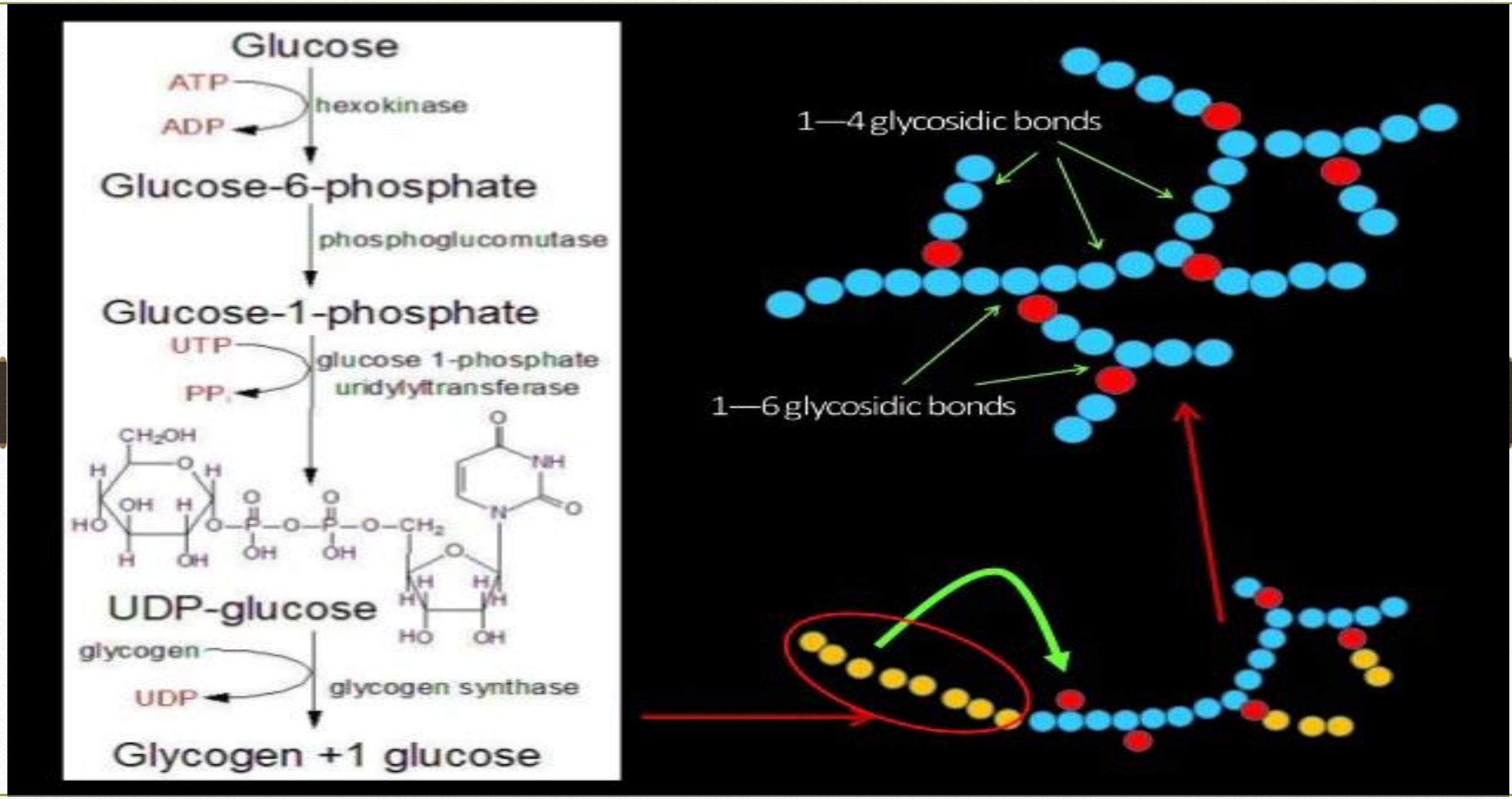
Branching



Branching is important as more number of glucose units Can be added simultaneously

Similarly Glycogen phosphorylation can release more glucose


Glycogen
1—4 glycosidic bonds
1—6 glycosidic bonds
Non reducing ends


Glucose
ATP
ADP
hexokinase
Glucose-6-phosphate
phosphoglucomutase
Glucose-1-phosphate
UTP
PP
glucose 1-phosphate uridylyltransferase
UDP-glucose
glycogen
UDP
glycogen synthase
Glycogen +1 glucose
1—4 glycosidic bonds
1—6 glycosidic bonds

Glycogenolysis

- Takes place in Liver and muscle cytosol
- Divided in to three phase

– 1. Release of G-1-P from glycogen

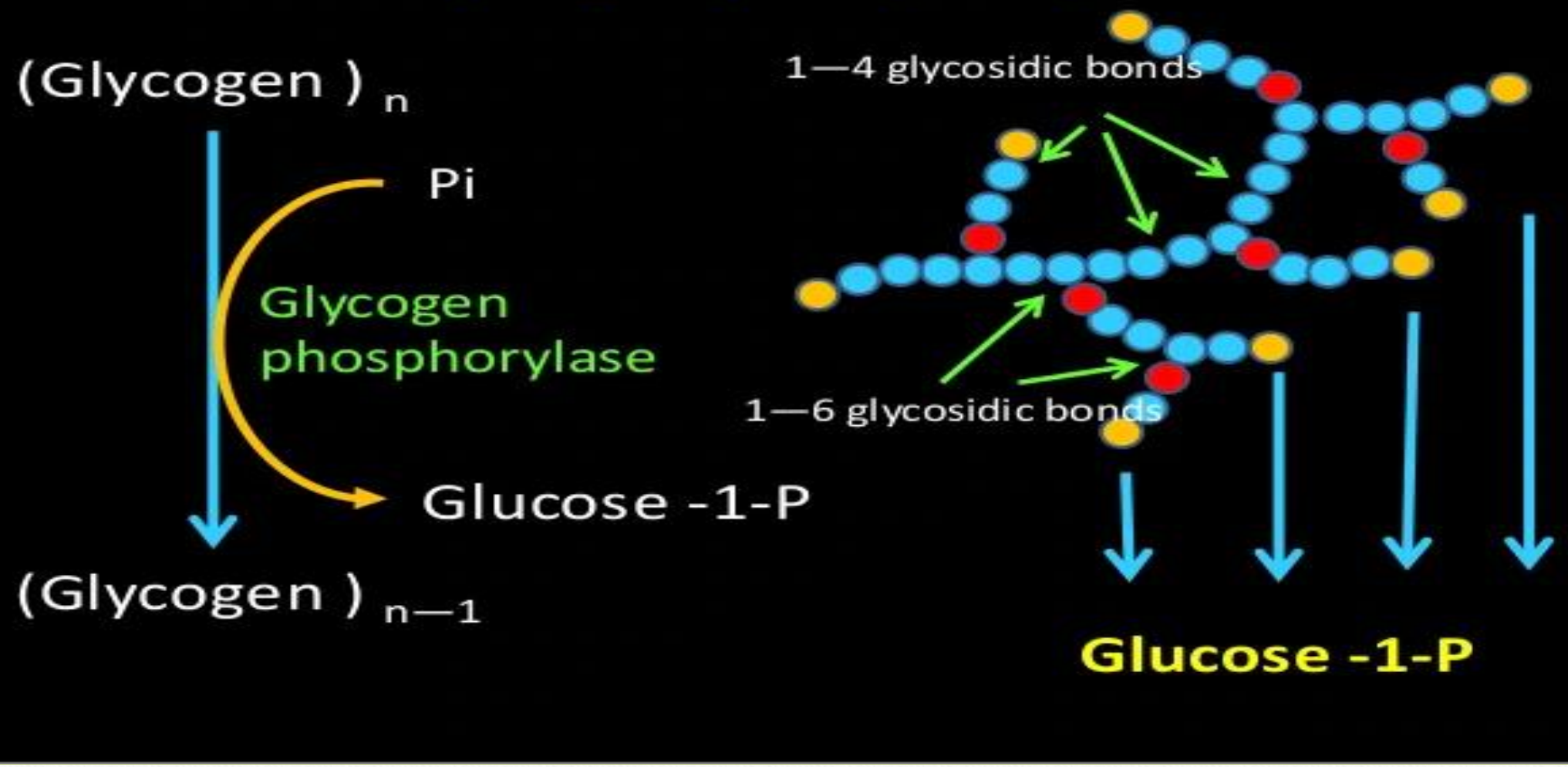
– 2. Remodeling of limit dextrin

– 3. Conversion of G-1-P to Glucose -6 – Phosphate

❑Two major enzymes required

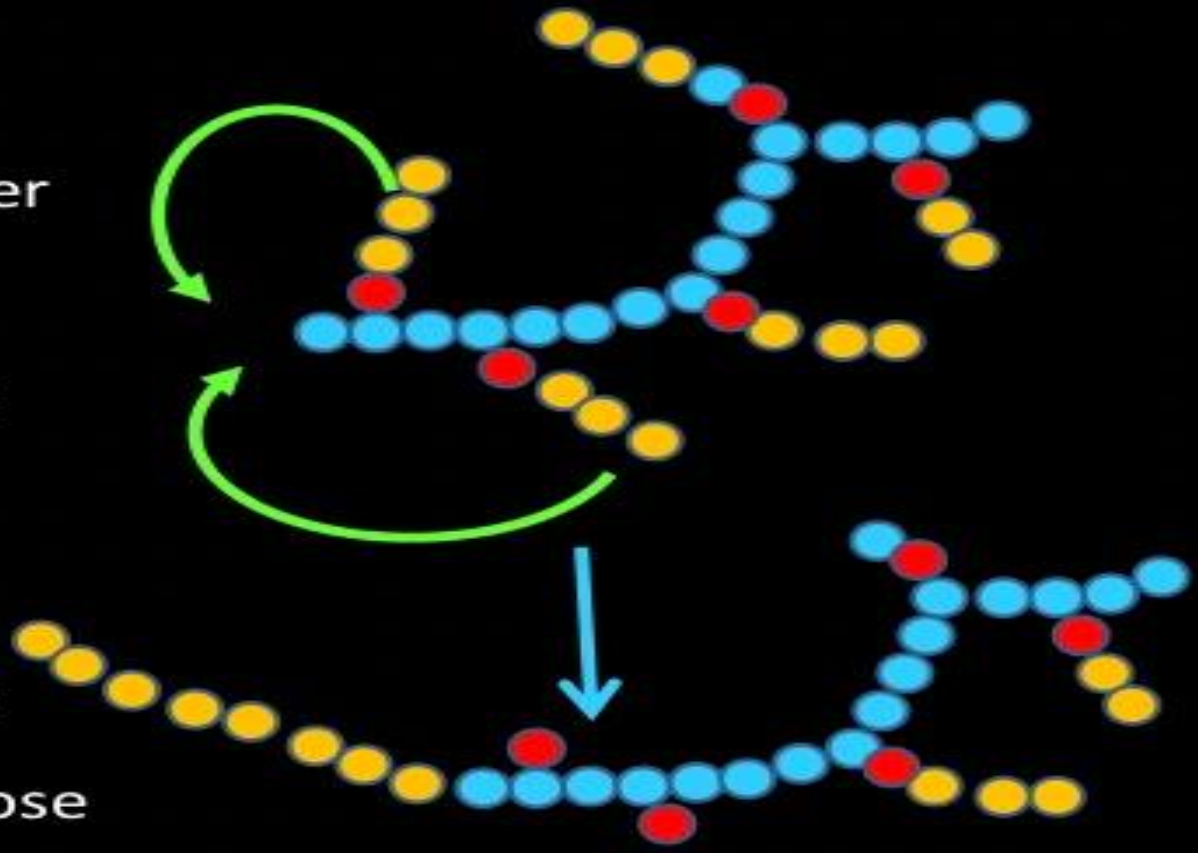
- 1. Glycogen phosphorylase
- 2. Glycogen debranching enzyme

Action of Glycogen phosphorylase



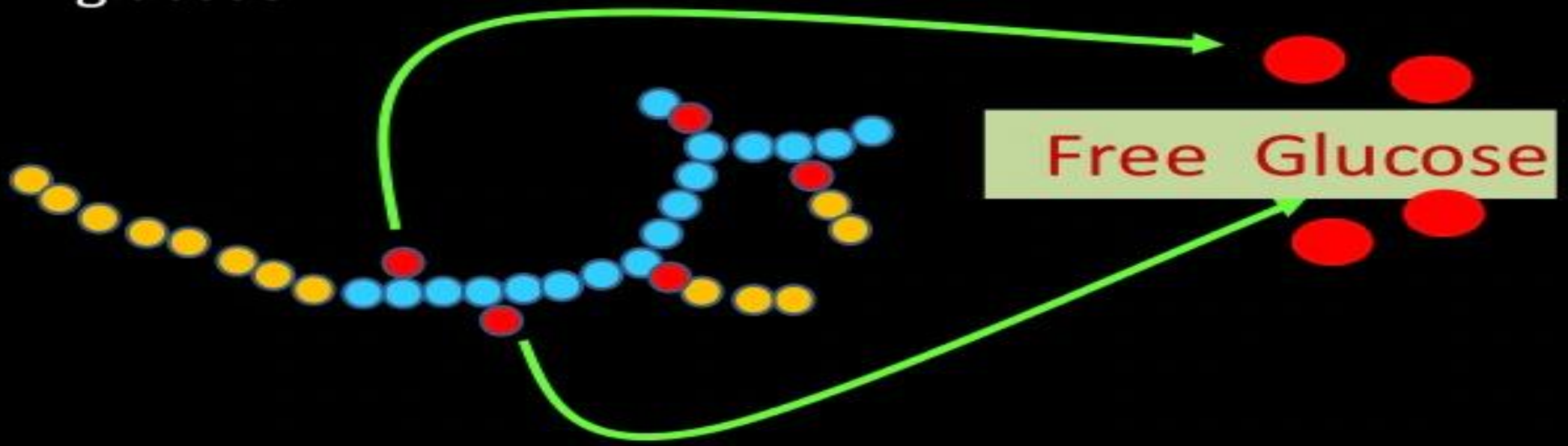
Action of debranching enzyme

- Shifts a block of three glucosyl unit from outer branch to other
- Exposes α 1—6 linkage
- α 1—6 Glucosidase or debranching enzyme hydrolyses α 1—6 bond and releases free glucose



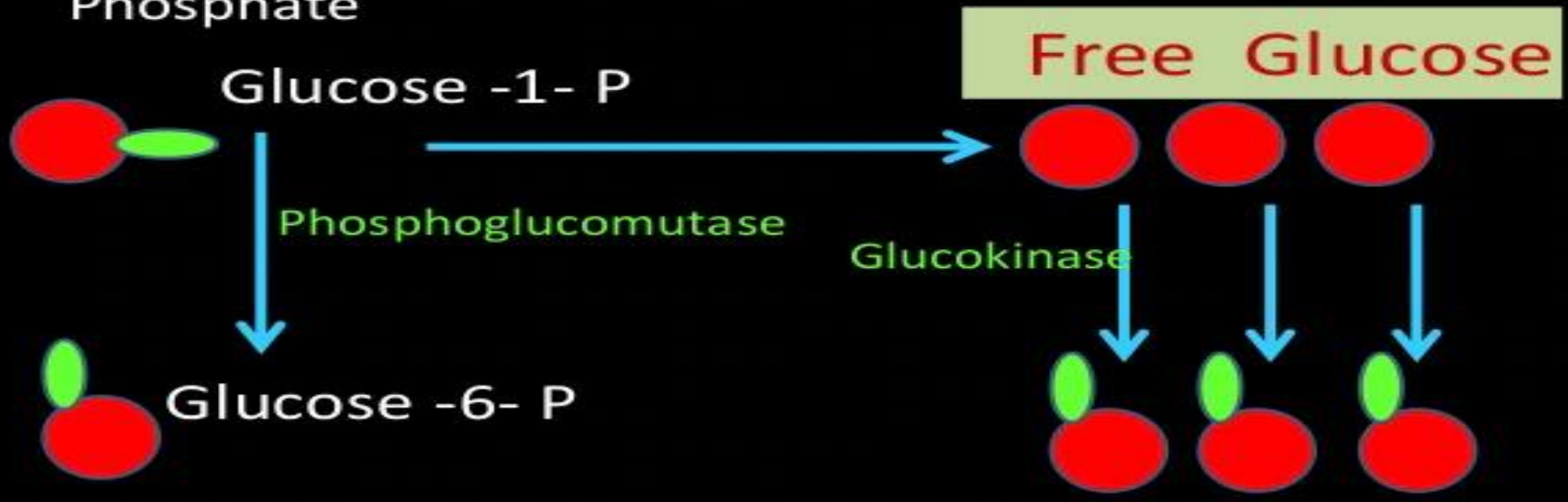
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Action of phospho-gluco-mutase enzyme

- Free glucose is phosphorylated
- Glucose -1-Phosphate is converted to Glucose -6-Phosphate



Glycogen storage diseases

- Group of inherited disorders characterized by deficient mobilization of glycogen or deposition of abnormal type or quantity of glycogen in tissues.
- Leads to liver damage or muscle weakness. Some GSD resulting in early death.
- Though there are more than 10 type six are classical type

❖E.g Gierke's disease has **Glucose 6 phaophatase defect** that lead Glycogen accumulates in liver & renal cell, Hypoglycemia, lipidemia, Ketoacidosis , Fatty liver , early death