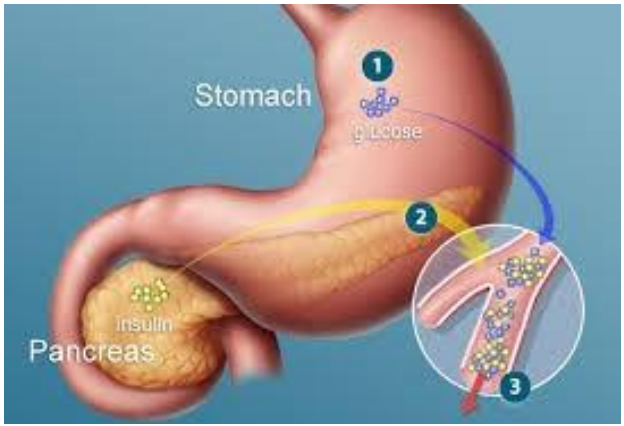
Glycolysis I

Glycolysis = sugar splitting



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Glucose as Energy Substrate

- To function properly, our cells are in need for energy which can be generated from the metabolism of various biomolecules such as carbohydrates, proteins and lipids
- Actually CHO particularly glucose is a major energy substrate in certain tissues like brain
- What are the metabolic pathways of glucose inside our cells?

Carbs —Enters body→ catabolism reaction

*cerebral tissues depend on glucose

*other tissues: doesn't matter what's the type of energy

Glycolysis

- Glycolysis is the metabolic pathway which converts glucose (6C) into 2 pyruvate molecules (3C)
- It occurs in the cell cytosol
- Glycolysis takes place in nearly all organisms both aerobic and anaerobic (i.e. microorganisms live in O_2 free environments)
- Glycolysis is a sequence of ten oxygen-independent and enzyme-catalyzed steps
- The intermediates either provide entry points to the cycle or themselves directly useful (biosynthetic intermediates)

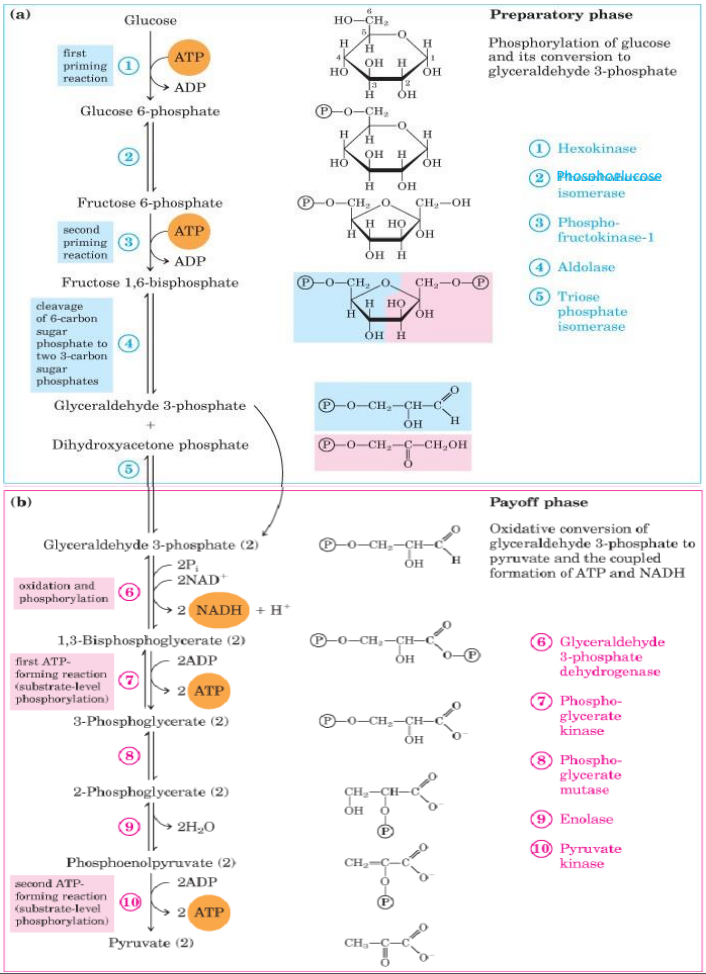
Intermediates → * biosynthesis = anabolic role
***connect to other pathways**

Glycolysis

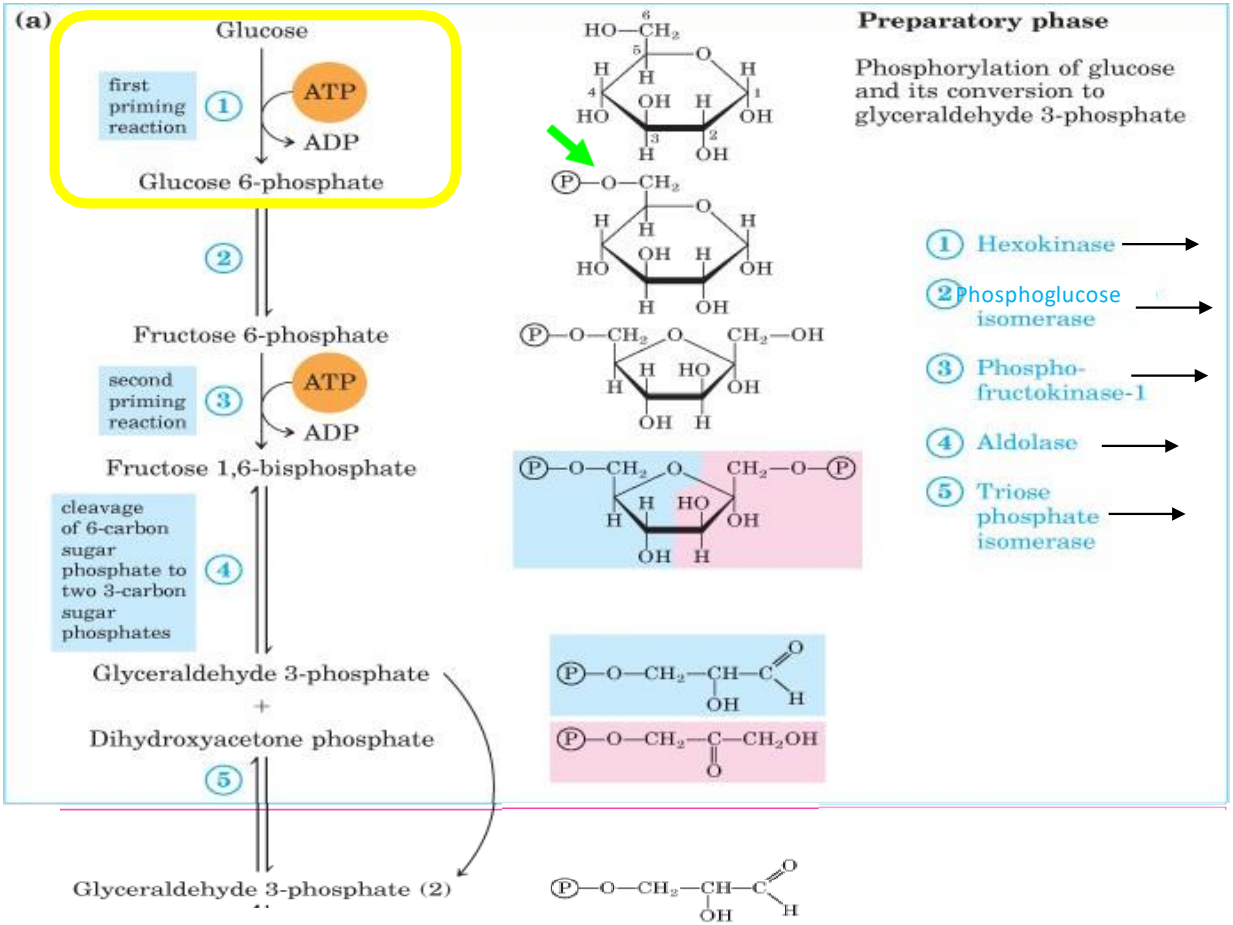
- The entire pathway is divided into two distinct phases:

A. Energy Investment Phase (Preparatory Phase)

B. Energy Generation Phase (Pay Off Phase)



A. Preparatory Phase



phosphorylation

isomerization

phosphorylation

cleavage

isomerization

Slide (5)

**Reaction 1:

r(x) name → ATP -dependent- phosphorylation
catalyzed by → hexokinase

Goal→

*it is (priming/ activation) reaction ?! (add ATP)
We add ATP in each reaction to activate sugar (turn it into active metabolite)

*maintain the influx from out side to inside (glucose), due to low concentration of glucose (Continues influx)

- Trapping of glucose inside the cell

**** reaction 1 important/ structure **not** important

Slide (6)

*irreversible? → hexokinase cant do DE phosphorylation

**G6P do DE phosphorylation Its express in certain type of tissues, (particular tissues in particular time

*regulatory steps? → regulate the pathway by regulation the activity of their catalyze enzyme

Irreversible step —> regulatory step
Enzyme which catalyze irreversible step —> allostric enzyme

A. Preparatory Phase

- **Step 1:** Hexokinases catalyze the ATP- dependent phosphorylation of glucose to produce glucose-6-phosphate (G6P)
- Hexokinase is a transferase enzyme which phosphorylates hexoses by transferring an inorganic phosphate from ATP usually to hydroxyl O at C6
- Irreversible reaction (another enzyme catalyzes the dephosphorylation, only found in specific tissues). Therefore, it is a target site for cycle regulation
- This first priming reaction is important to maintain the influx of glucose through glucose transporters (GLUTs) and at the same time to trap the transported glucose molecules inside the cell

Hexokinases

- 4 isoforms (isozymes) of hexokinase (I, II, III & IV) which differ in their **location**, **catalysis** and **regulation** thereby, contributing to different pattern of glucose metabolism in different tissues
- Hexokinase I, II & III are nonspecific and can phosphorylate a variety of hexoses (e.g. glucose, fructose, mannose) but type I is involved in catabolic pathways like glycolysis whereas type II & III are involved in anabolic pathways like glycogenesis
- Hexokinase IV is called glucokinase expressed in liver and pancreatic cells. It is specific for D-glucose

Liver + pancreatic cells→ control blood glucose level through (isoform) containing glucokinase

***Isoforms are the reason of minor variety in pathways**

Hexokinases

Km: measure for enzyme affinity, inverse relationship (high Km → low affinity)

- Glucokinase has low affinity for glucose (high Km value) compared to others (low Km value)

activated at high level

- Therefore, glucokinase in **liver** is active only at high blood glucose level to accumulate G6P for glycogen synthesis but in the **pancreas** it acts as glucose sensor to control insulin release from beta cells and glucagon release from alpha cells

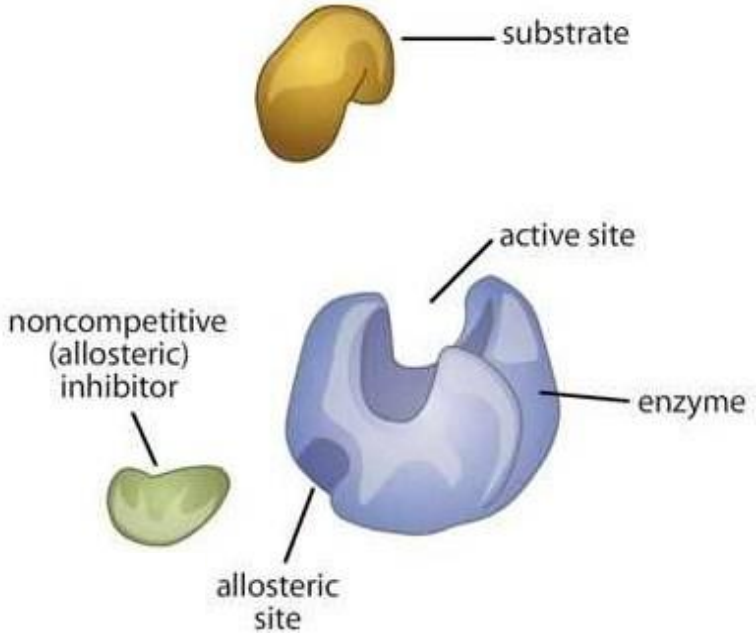
When glucose level is high → activate glucokinase to phosphorylate (in liver & pancreas)k → activation of beta cells to produce insulin (glycogenesis).
Low levels of glucose → alpha cells produce glucagon (glycogen lysis)

- Hexokinase isoforms (except isoform IV) are allosterically inhibited by G6P **only** at high level by substrate-level control mechanism (it is controlled by the level of its product)

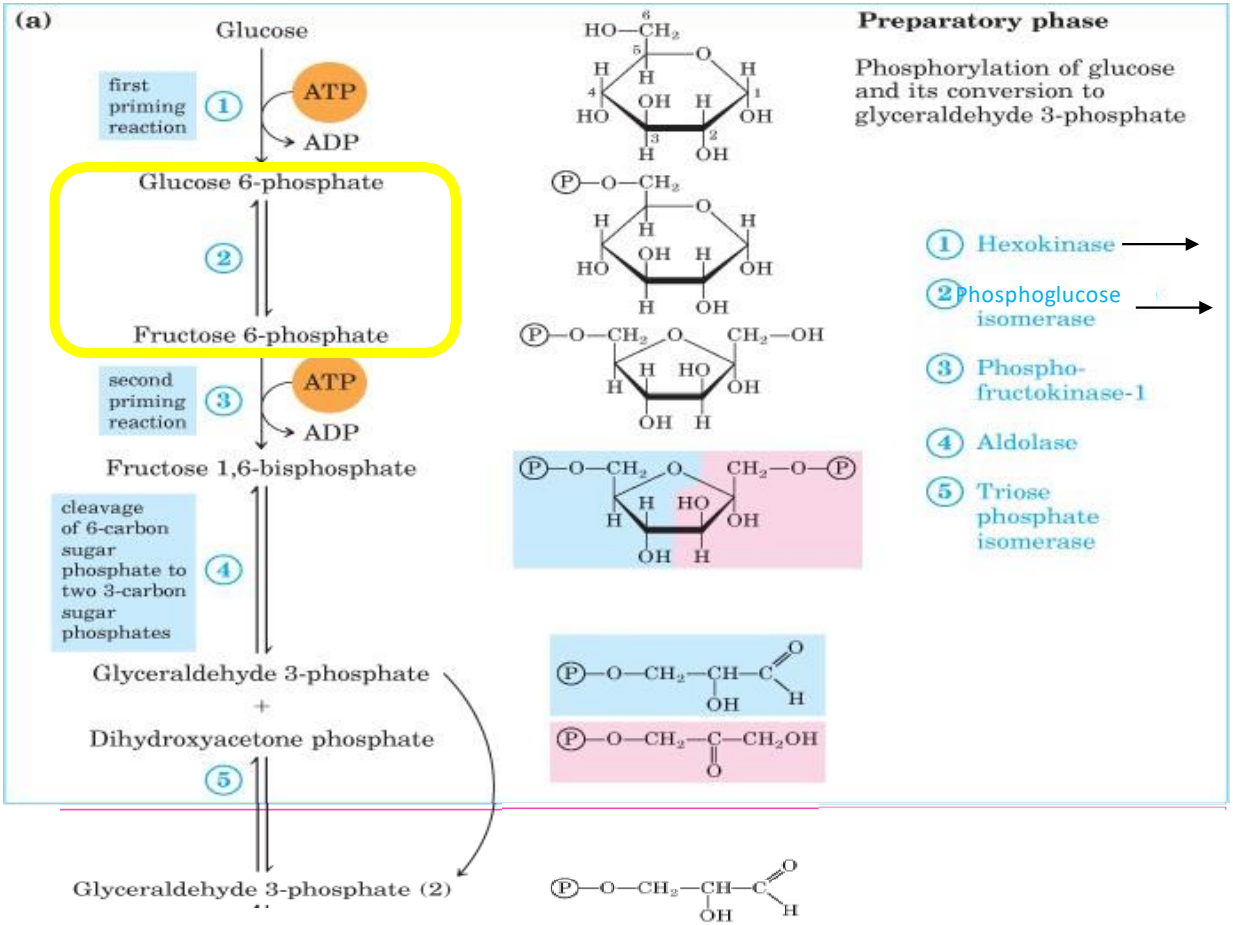
Hexokinases

- Hexokinase is an allosteric enzyme with two binding sites: catalytic site (binds substrate) and regulatory site (allosteric site binds effectors)

***Only hexokinase (I, II, III), When the cell in rest, NO glycolysis down stream, Accumulation of (G6P) → allosteric inhibitor, which binds the enzyme & inhibit it (substrate level control)**
***This don't happen in (IX) because liver require to accumulate of G6P**
even when it accumulates

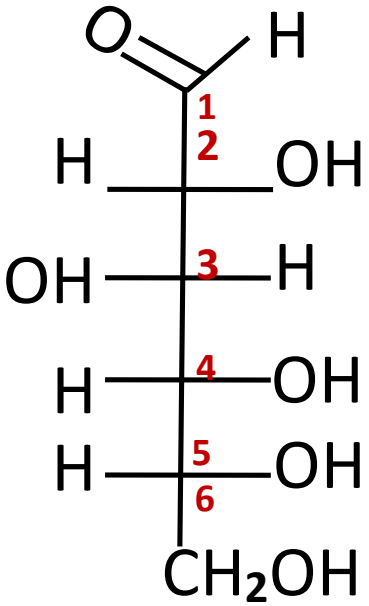


A. Preparatory Phase



A. Preparatory Phase

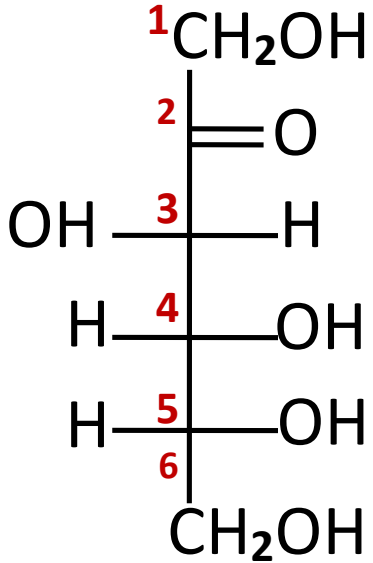
- **Step 2:** Phosphoglucose isomerase (PGI) interconverts G6P and F6P (reversible reaction).
- Indeed, **Mannose and Fructose** can enter the glycolytic pathway at this point



D-glucose

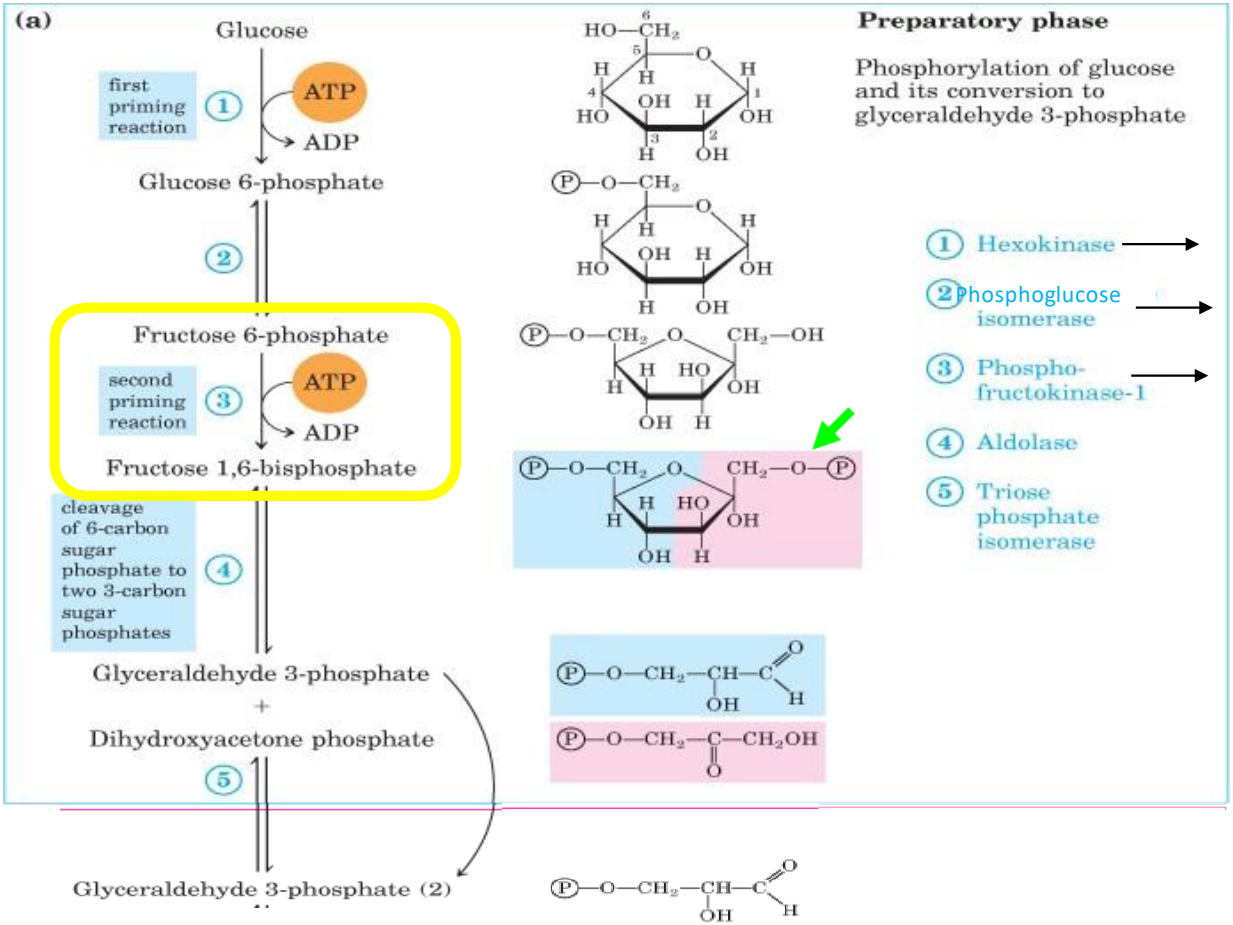
Hexose $C_6H_{12}O_6$

*Goal→ to prepare for the next step (the target site)
*F6P→ entry point for mannose & fructose



D-fructose

A. Preparatory Phase

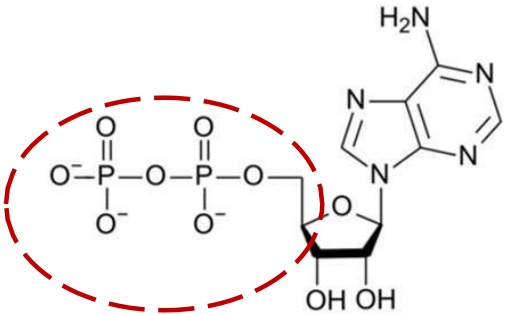


phosphorylation

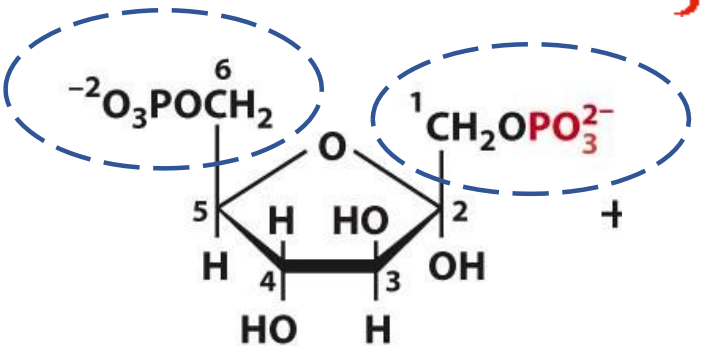
isomerization

phosphorylation

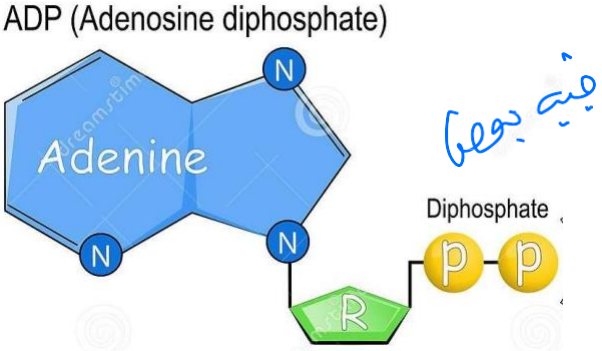
A. Preparatory Phase



ADP

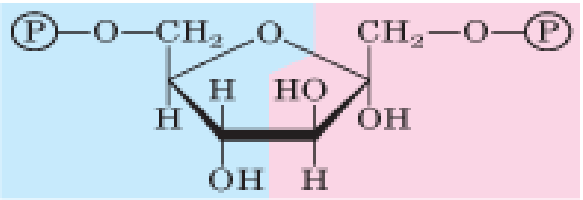


Fructose 1,6 bisphosphate



جنب بعض

بجيب اي حتى بعض



A. Preparatory Phase

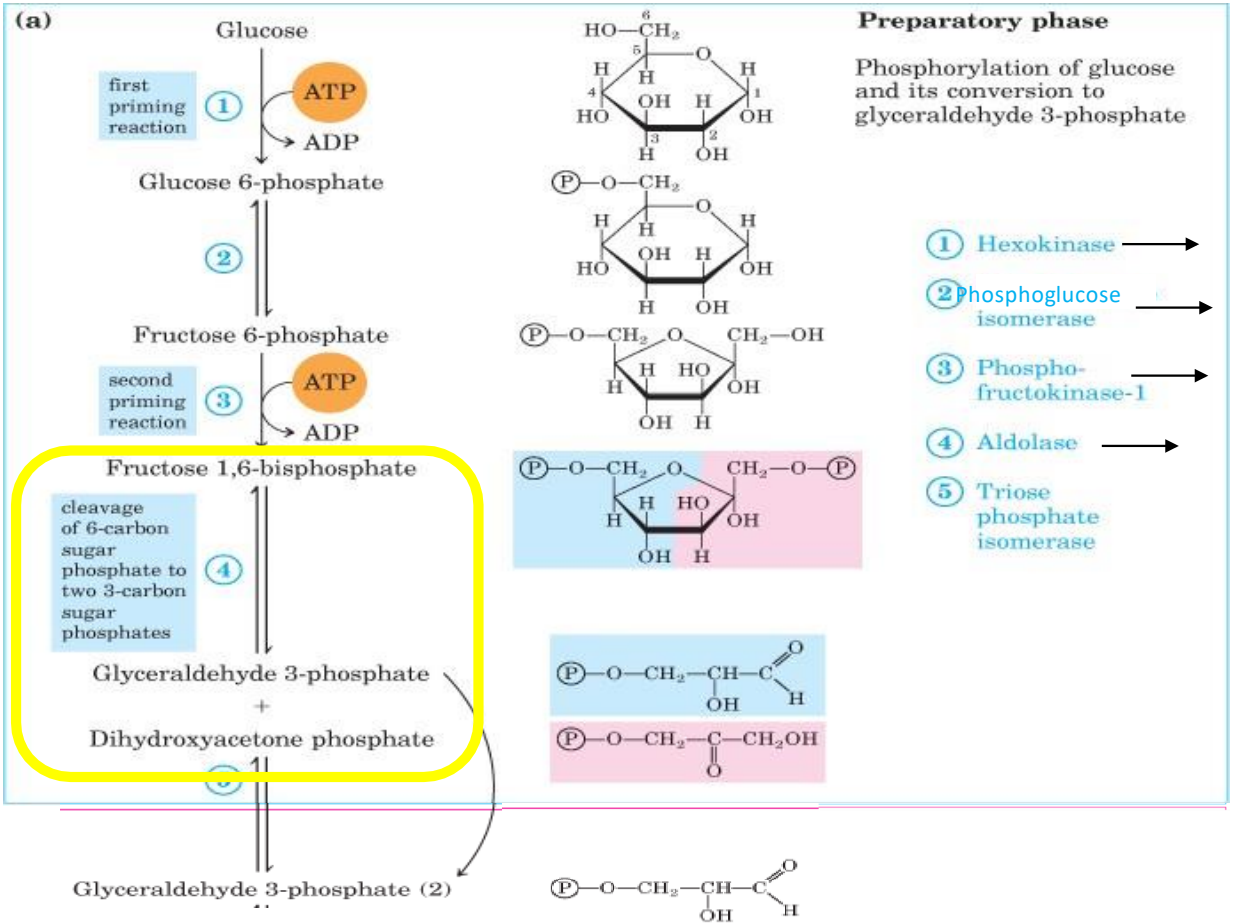
Step 3: bisphosphate → means the two phosphate groups in different directions

Key regulation site/ rate limiting step→ glycolysis, is mainly dependent on regulation of (PFK) activity

- **Step 3:** This is the rate limiting or key regulatory step. The activity of phosphofructokinase-1 (PFK-1) enzyme can be controlled. PFK-1 catalyzes the phosphorylation of hydroxyl oxygen at C1 to produce **fructose-1,6-bisphosphate**
- **Step 4: (splitting)** Aldolase enzyme catalyzes the cleavage to two triose phosphates: DHAP (dihydroxyacetone phosphate) and GADP (glyceraldehyde-3-phosphate)
- The addition of the second phosphate group on C1 from the previous step destabilizes the hexose ring and facilitates the cleavage reaction

Step 4: two bulk groups in front of each other repulsion which decrease the stability

A. Preparatory Phase



phosphorylation

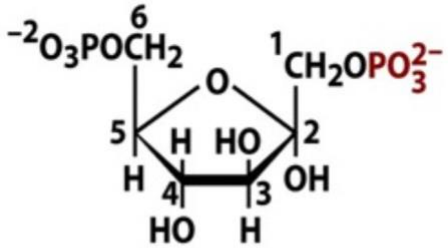
isomerization

phosphorylation

cleavage

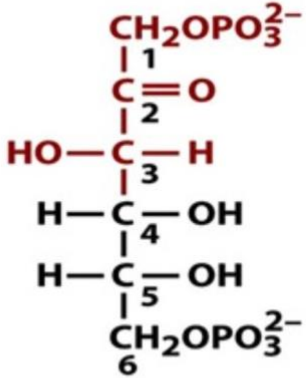
Aldolase Mechanism of Action

Haworth and Fischer Projections Equivalency



Fructose-1,6-bisphosphate
(FBP)

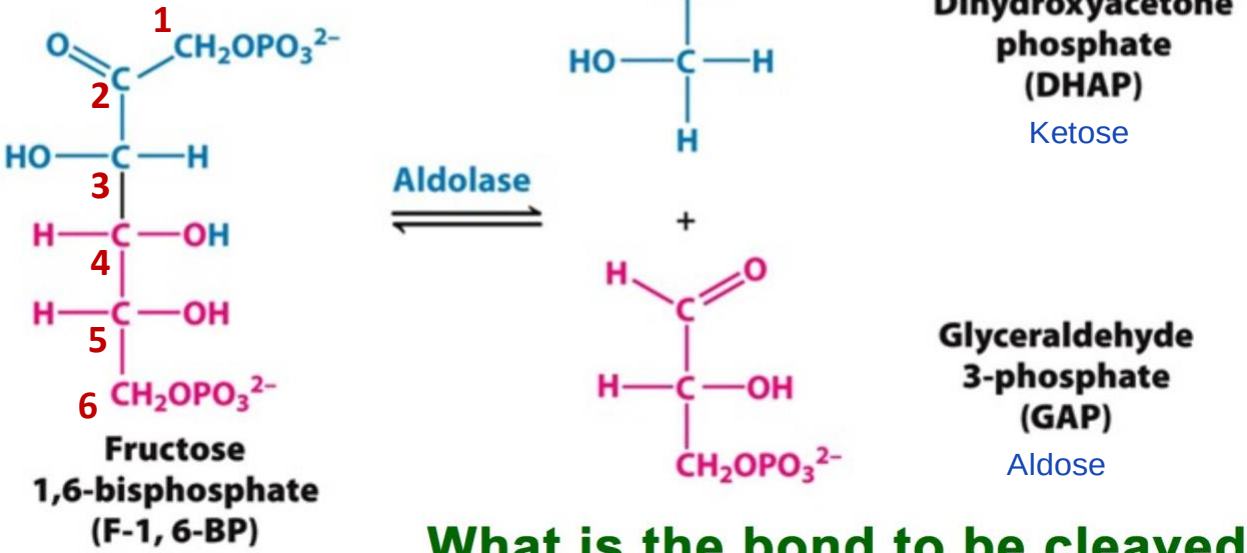
≡



Fructose-
1,6-bisphosphate
(FBP)

Aldolase Mechanism of Action

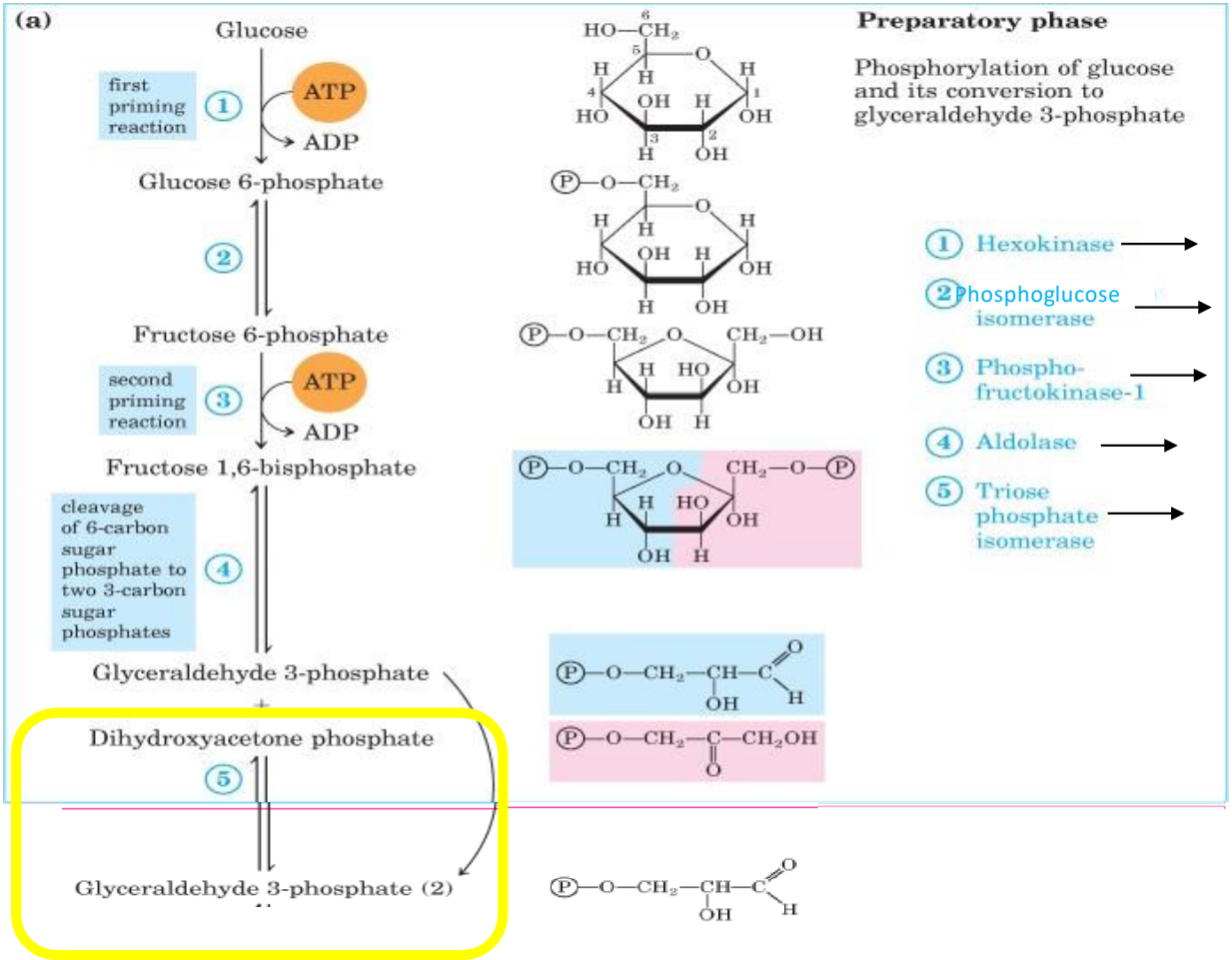
Six Carbon Sugar Cleaved to Two Three Carbon Units



What is the bond to be cleaved?

Bond between C3 with C4

A. Preparatory Phase



phosphorylation

isomerization

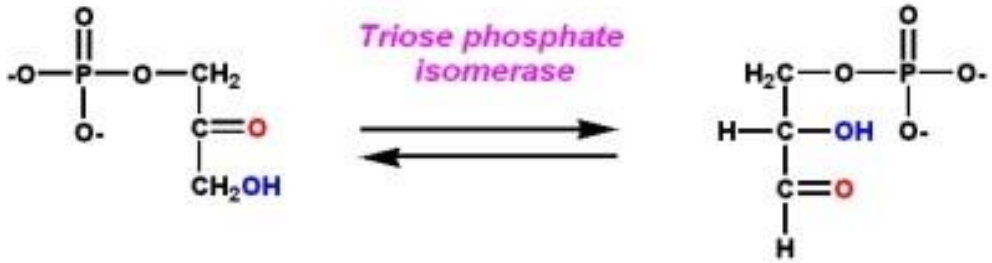
phosphorylation

cleavage

isomerization

A. Preparatory Phase

- **Step 5:** Isomerization of DHAP by triose phosphate isomerase (TPI) to GADP to proceed further in glycolysis as GADP is the substrate for the next reaction. This reaction is reversible

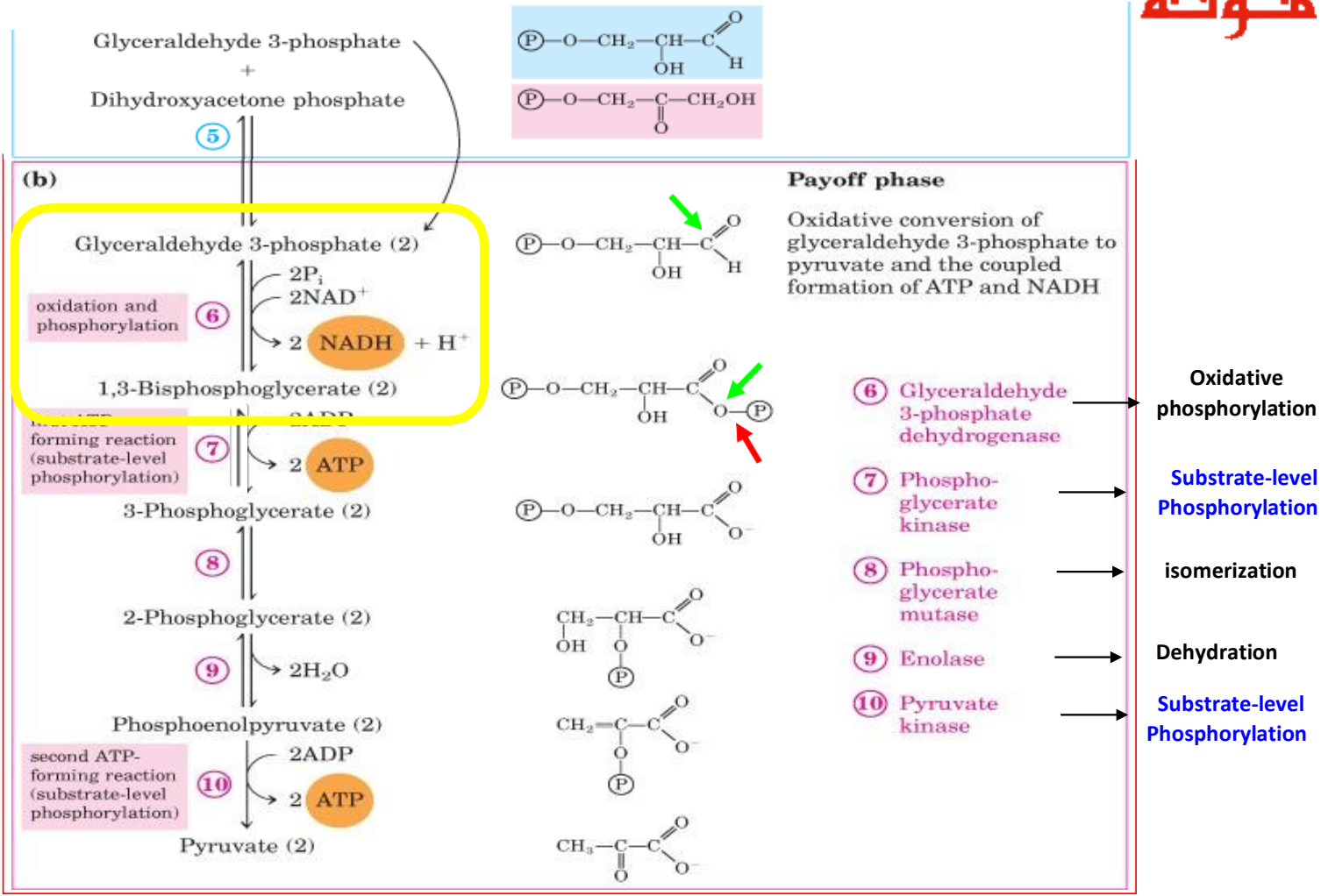


When the cell accumulates without needed for energy production, the increase converts to DHAP

***DHAP (NOT substrate) precursor of glycerol which is part of triglycerides (link between lipid & carbs metabolism)**

B. Pay Off Phase

In Pay off phase → each product out from reaction will be (X2)

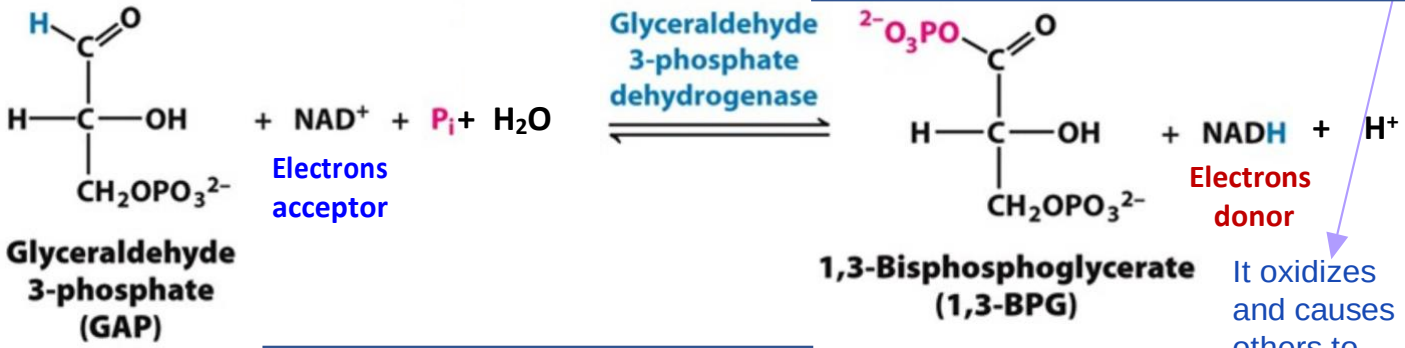


B. Pay Off Phase

Step 6: oxidative phosphorylation reaction / reversible
*goal of phosphorylation is to make super high energy molecule (1,3- BPG) 1st one

- **Step 6**: GADP dehydrogenase enzyme catalyzes the oxidative phosphorylation of GADP (electron donor) into super-high-energy compound **(1,3-BPG)** and the transfer of electrons into the coenzyme NAD^+(electron acceptor) forming **NADH**
- Dehydrogenases are named as electrons donor substrate -dehydrogenase

*mcq → Bisphosphoglycerate NOT bi
*mcq → in dehydrogenases dr. nessren will ask about (oxidizing, reducing agents)



It oxidizes and causes others to reduce

Electrons donor (reductant)

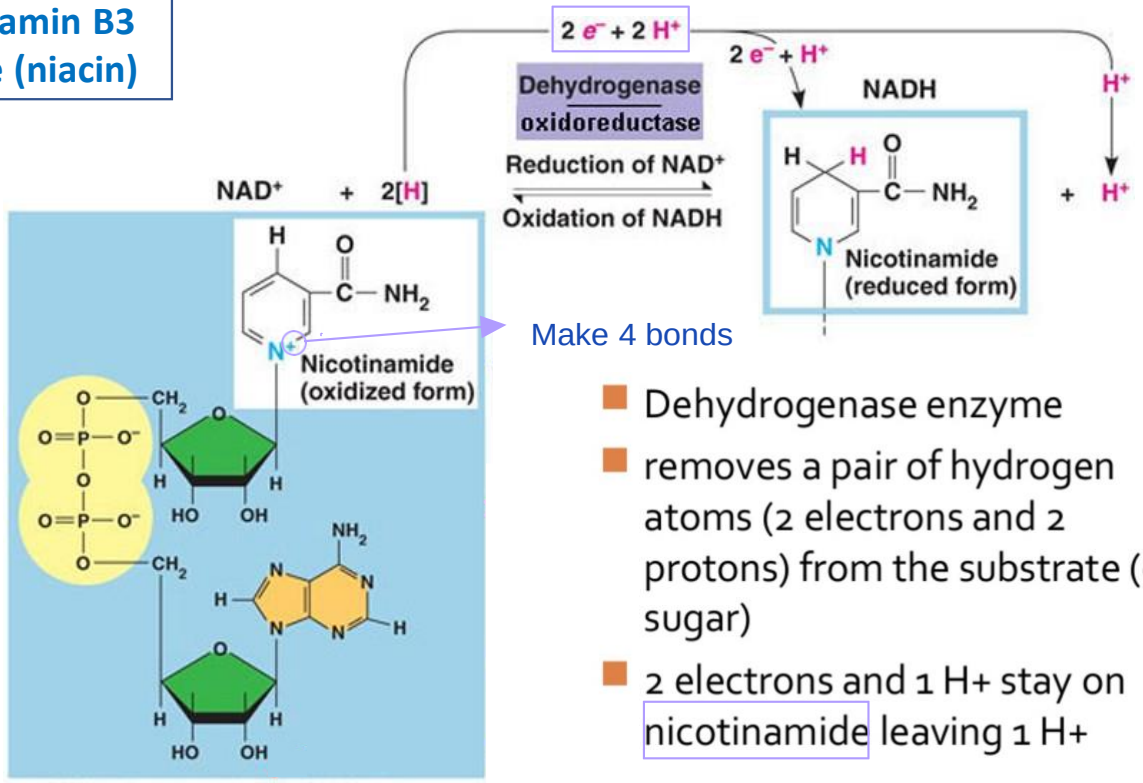
NAD+ (coenzyme)→ NADH2 (indirect form of energy)

Electrons acceptor (oxidant)

Nicotinamide Adenine Dinucleotide

- **NAD (Nicotinamide adenine dinucleotide)** is a coenzyme which exists in two forms: NADH (the reduced form) and NAD^+ (the oxidized form)

***NAD: vitamin B3 derivative (niacin)**



- Dehydrogenase enzyme
- removes a pair of hydrogen atoms (2 electrons and 2 protons) from the substrate (eg. sugar)
- 2 electrons and 1 H+ stay on nicotinamide leaving 1 H+

Nicotinamide Adenine Dinucleotide

- **NAD (Nicotinamide adenine dinucleotide)** is a coenzyme of dehydrogenases
- The reduced form NADH is electrons carrier and it is called **energy rich molecule**. It is an indirect form of energy

1 NADH = 2.5 ATP

Slide 23:- The reaction

1- 2 electrons and 2 protons are released from GAP
2- an electron binds with the N+ of nicotinamide→ rearrangement of double bonds of the ring
3- The opposite C atom of the ring is now capable for binding with another atom
4- The opposite C atom binds with a proton and electron, results in extra proton that is still released freely
5- products are NADH + H*
6- NADH reaches the mitochondria to participate in electron transport chain
7- NADH now loses its proton and electrons in order to produce energy
8- NADH is converted to NAD+ again to regenerate the pool of NAD+ in glycolysis