METABOLIC PATHWAYS

CATABOLIC PATHWAYS

Are involved in oxidative breakdown of larger complexes.

They are usually **exergonic** in nature

ANABOLIC PATHWAYS

Are involved in the synthesis of compounds.

They are usually **endergonic** in nature.

CHARACTERISTICS OF METABOLISM

1. Metabolic pathways are mostly irreversible
2. Every metabolic pathway has a committed first step.
3. All metabolic pathways are regulated.
4. Metabolic pathways in eukaryotic cells occur in specific cellular locations.

GLYCOLYSIS

GLYCOLYSIS

Glycolysis comes from a merger of two Greek words:

- **Glykys = sweet**
- **Lysis = breakdown/ splitting**

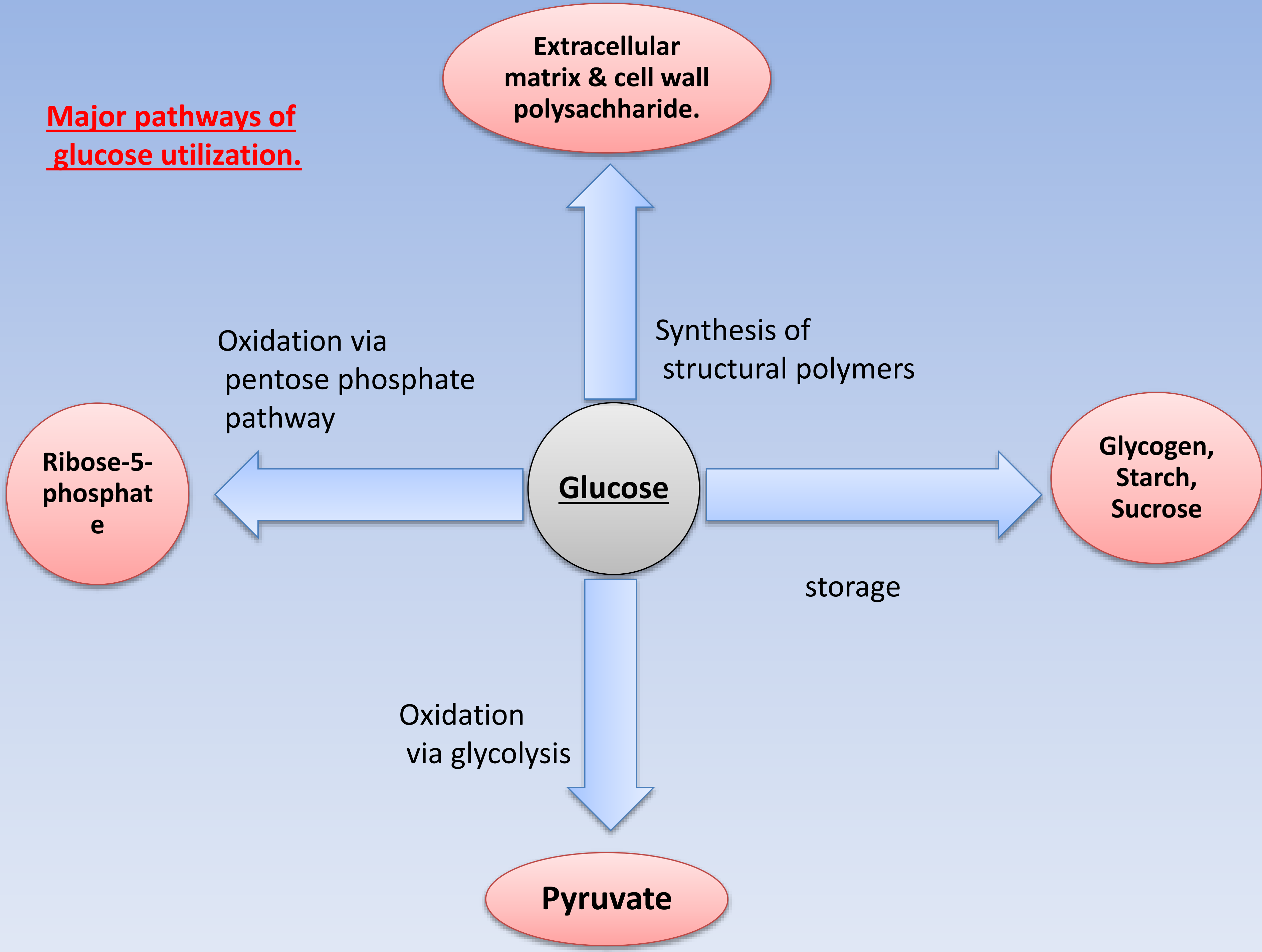
It is also known as Embden-Meyerhof-Parnas pathway or EMP pathway.

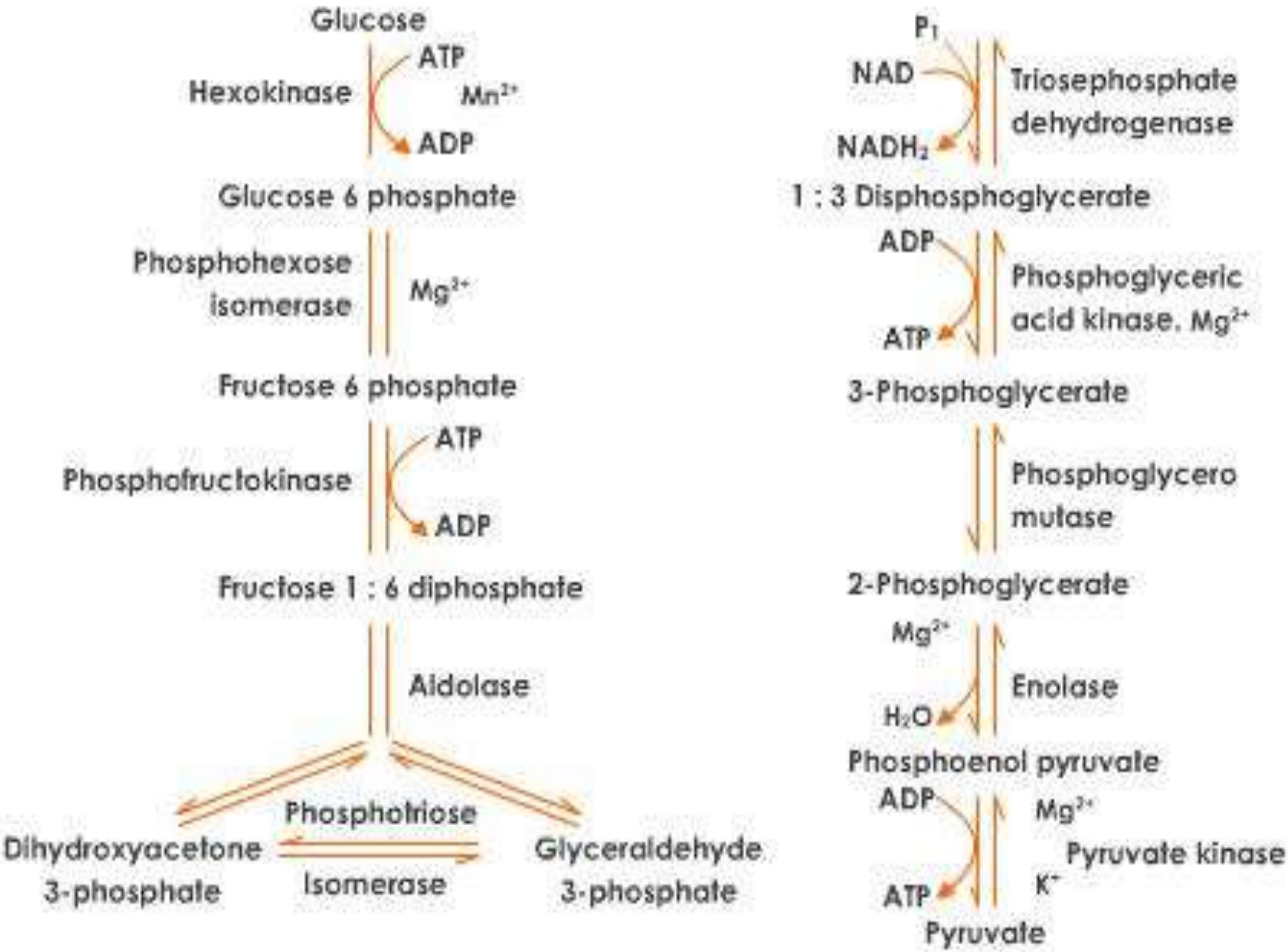
INTRODUCTION

- GLYCOLYSIS is the sequence of 10 enzyme-catalyzed reactions that converts glucose into pyruvate with simultaneous production on of ATP.
- In this oxidative process, 1mol of glucose is partially oxidised to 2 moles of pyruvate.
- This major pathway of glucose metabolism occurs in the cytosol of all cell.
- This unique pathway occurs **aerobically** as well as **anaerobically & doesn't involve molecular oxygen.**

- It also includes formation of Lactate from Pyruvate.
- The glycolytic sequence of reactions differ from species to species only in the mechanism of its regulation & in the subsequent metabolic fate of the pyruvate formed.
- In aerobic organisms, glycolysis is the prelude to Citric acid cycle and ETC.
- Glycolysis is the central pathway for Glucose catabolism.

Major pathways of glucose utilization.





Glucose
ATP
Hexokinase
Mn2+
ADP
Glucose 6 phosphate
Phosphohexose
isomerase
Mg2+
Fructose 6 phosphate
ATP
Phosphofructokinase
ADP
Fructose 1 : 6 diphosphate
Aldolase
Phosphotriose
Dihydroxyacetone
3-phosphate
Isomerase
Glyceraldehyde
3-phosphate
P1
NAD
Triosephosphate
dehydrogenase
NADH2
1 : 3 Disphosphoglycerate
ADP
Phosphoglyceric
acid kinase, Mg2+
ATP
3-Phosphoglycerate
Phosphoglycero
mutase
2-Phosphoglycerate
Mg2+
Enolase
H2O
Phosphoenol pyruvate
ADP
Mg2+
Pyruvate kinase
K+
ATP
Pyruvate

TWO PHASES OF GLYCOLYSIS

- Glycolysis leads to breakdown of 6-C glucose into two molecules of 3-C pyruvate with the enzyme catalyzed reactions being bifurcated or categorized into 2 phases:

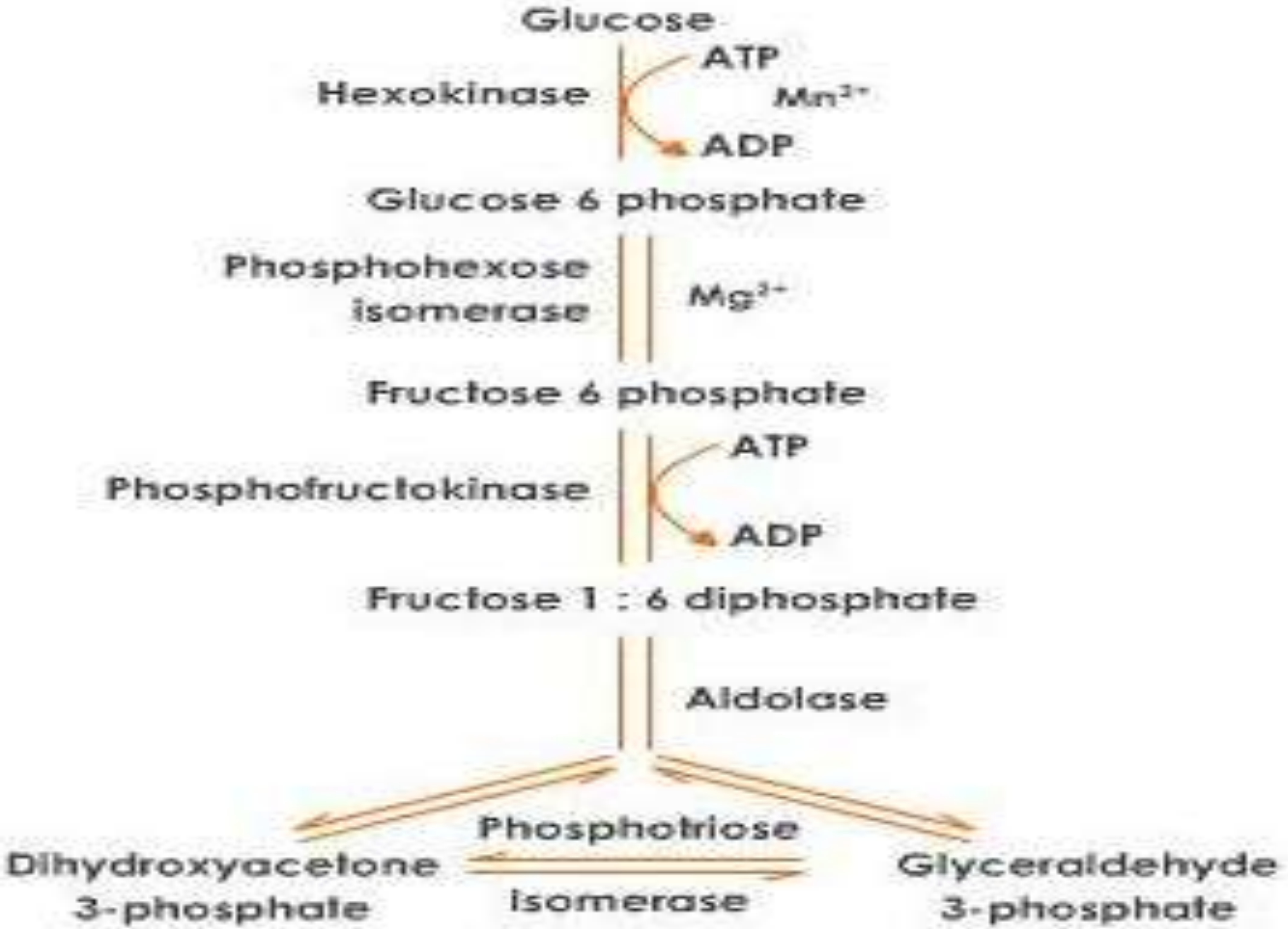
1. **Phase 1- preparatory phase**
2. **Phase 2- payoff phase.**

PREPARATORY PHASE

- It consists of the 1st 5 steps of glycolysis in which the glucose is enzymatically phosphorylated by ATP to yield Fructose-1,6-biphosphate.
- This fructuse-1,6-biphosphate is then split in half to yield 2 molecules of 3-carbon containing Glyceraldehyde-3-phosphate/ dihyroxyacteone phosphate.

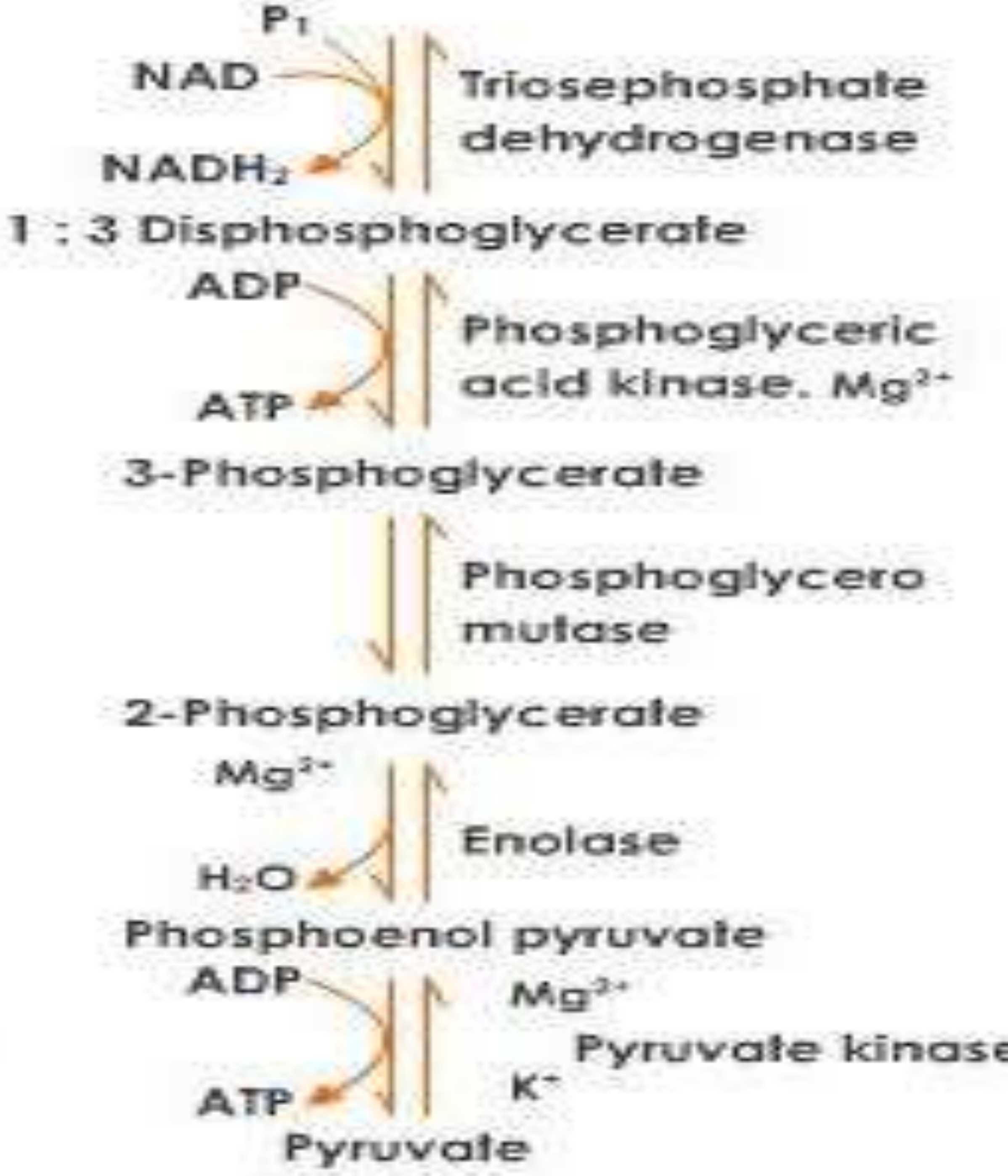
- Thus the first phase **results in cleavage of the hexose chain.**

- This cleavage requires an investment of 2 ATP molecules to activate the glucose mole and prepare it for its cleavage into 3-carbon compound.


Glucose
ATP
Hexokinase
Mn2+
ADP
Glucose 6 phosphate
Phosphohexose
isomerase
Mg2+
Fructose 6 phosphate
ATP
Phosphofructokinase
ADP
Fructose 1 : 6 diphosphate
Aldolase
Phosphotriose
Dihydroxyacetone
3-phosphate
isomerase
Glyceraldehyde
3-phosphate

PAYOFF PHASE

- This phase constitutes the last 5 reactions of Glycolysis.
- This phase marks the release of ATP molecules during conversion of Glyceraldehyde-3-phosphtae to 2 moles of Pyruvate.
- Here 4 moles of ADP are phosphorylated to ATP. Although 4 moles of ATP are formed, the net result is only 2 moles of ATP per mole of Glucose oxidized, since 2 moles of ATP are utilized in Phase 1.

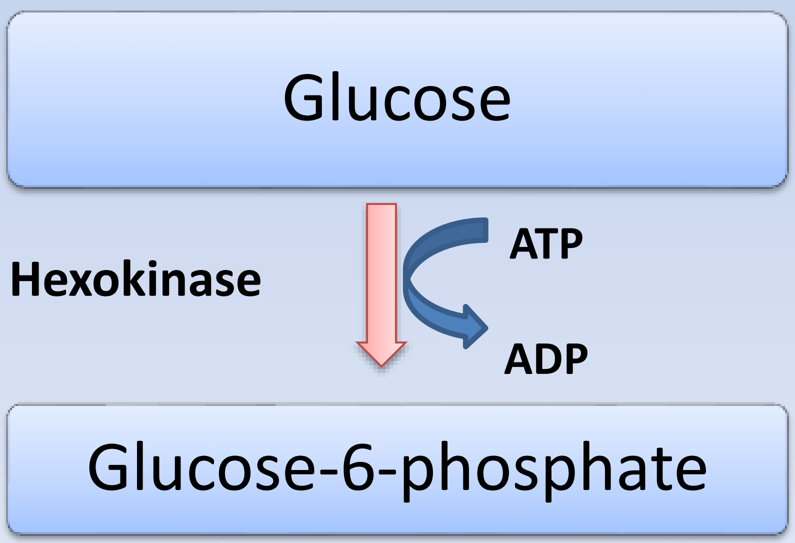


P_i
NAD
$NADH_2$
Triosephosphate dehydrogenase
1 : 3 Disphosphoglycerate
ADP
ATP
Phosphoglyceric acid kinase. Mg^{2+}
3-Phosphoglycerate
Phosphoglycero mutase
2-Phosphoglycerate
Mg^{2+}
H_2O
Enolase
Phosphoenol pyruvate
ADP
ATP
Mg^{2+}
Pyruvate kinase
K^+
Pyruvate

STEPWISE EXPLAINATION OF GLYCOLYSIS

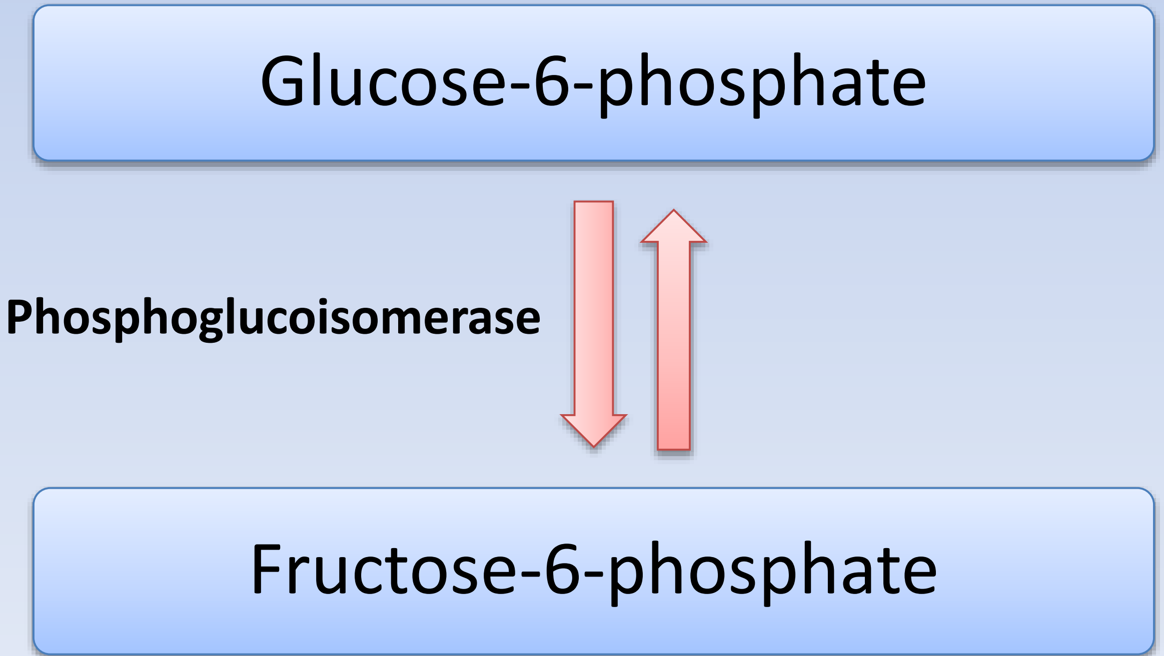
STEP 1: PHOSPHORYLATION

- Glucose is phosphorylated by ATP to form sugar phosphate.
- This is an irreversible reaction & is catalyzed by ***hexokinase.***
- Thus the reaction can be represented as follows:



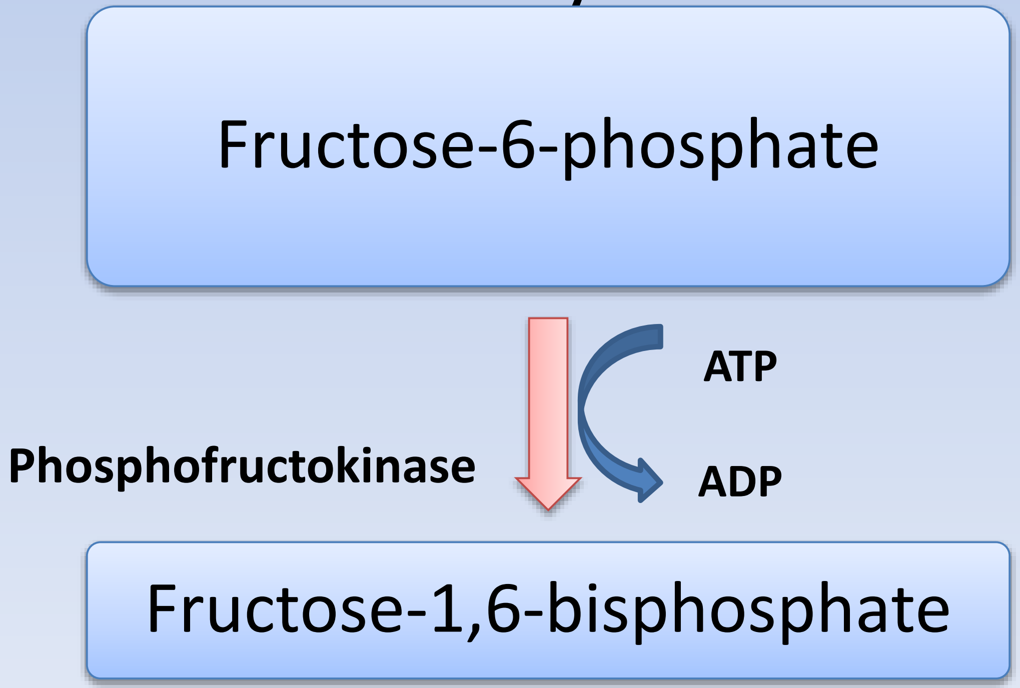
STEP 2: ISOMERIZATION

- It is a reversible rearrangement of chemical structure of carbonyl oxygen from C1 to C2, forming a Ketose from the Aldose.
- Thus, isomerization of the aldose Glucose6-phosphate gives the ketose, Fructose-6-phoshphate.



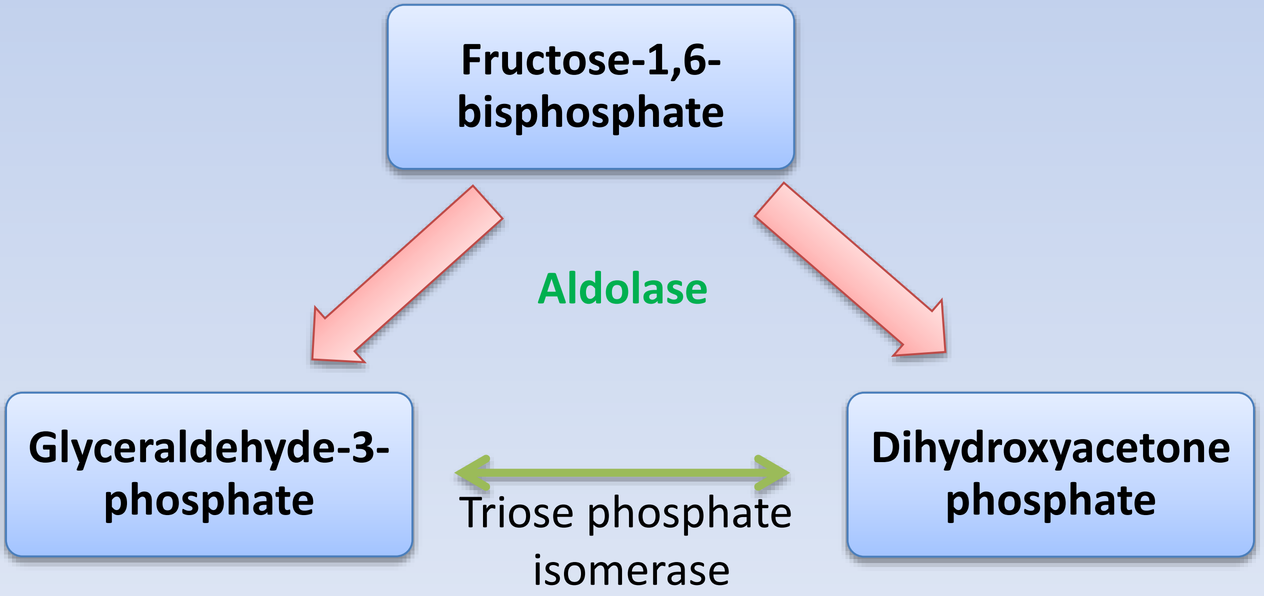
STEP 3: PHOPHORYLATION

- Here the ***Fructose-6-phosphate*** is phosphorylated by ATP to ***fructose-1,6-bisphosphate.***
- This is an ***irreversible reaction*** and is catalyzed by ***phosphofructokinase*** enzyme.



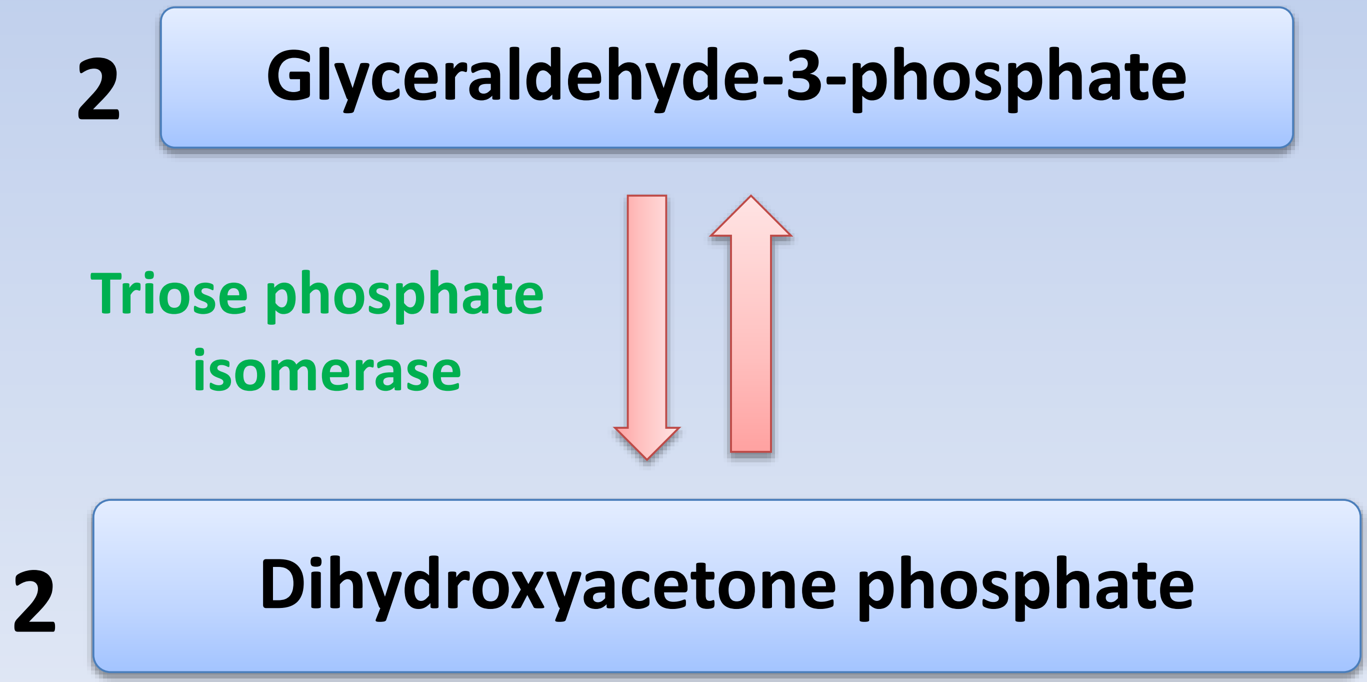
STEP 4: BREAKDOWN

- This six carbon sugar is cleaved to produce two 3-C molecules: ***glyceradldehyde-3-phosphate (GAP)*** & ***dihydroxyacetone phosphate(DHAP).***
- This reaction is catalyzed by ***Aldolase.***



STEP 5: ISOMERIZATION

- Dihydroxyacetone phosphate is oxidized to form Glyceraldehyde-3-phosphate.
- This reaction is catalyzed by ***triose phosphate isomerase*** enzyme.



STEP 6

- 2 molecules of Glyceraldehyde-3-phosphate are oxidized.
- ***Glyceraldehyde-3-phosphate dehydrogenase*** catalyzes the conversion of Glyceraldehyde3-phosphate into ***1,3-bisphosphoglycerate.***

Aldehyde → Carboxylic acid

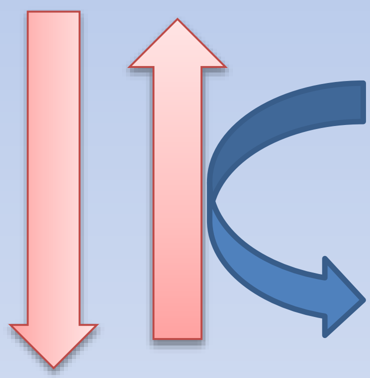
Carboxylic acid + Ortho-phosphate = Acyl-phosphate product

Joining)

Resultant reaction

2 **Glyceraldehyde-3-phosphate**

Glyceraldehyde-3-phosphate dehydrogenase



$2NAD^+$ + 2Pi

2NADH + $2H^+$

2 **1,3-bisphosphoglycerate**

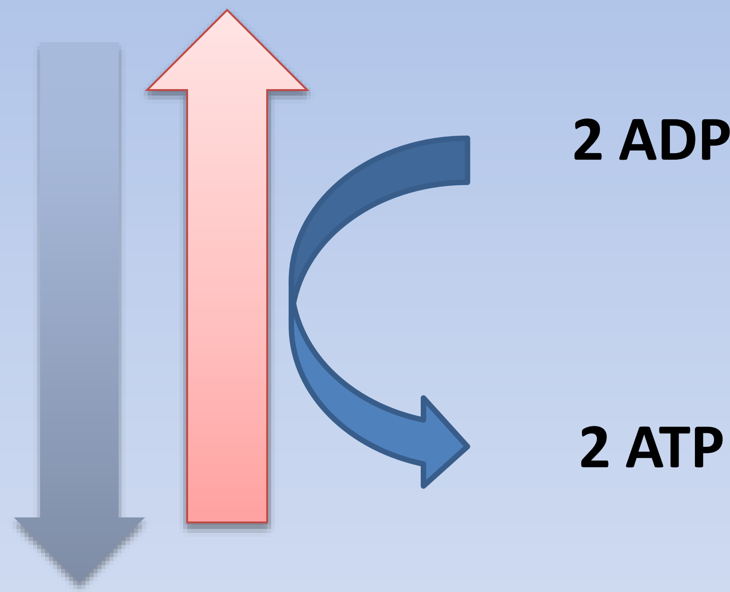
STEP 7

- The transfer of high-energy phosphate group that was generated earlier to ADP, form ATP.
- This phosphorylation i.e. addition of phosphate to ADP to give ATP is termed as ***substrate level phosphorylation*** as the phosphate donor is the substrate ***1,3-bisphosphoglycerate (1,3-BPG).***
- The product of this reaction is 2 molecules of ***3-phosphoglycerate.***

2 **1,3-bisphosphoglycerate**

Phosphoglycerate kinase

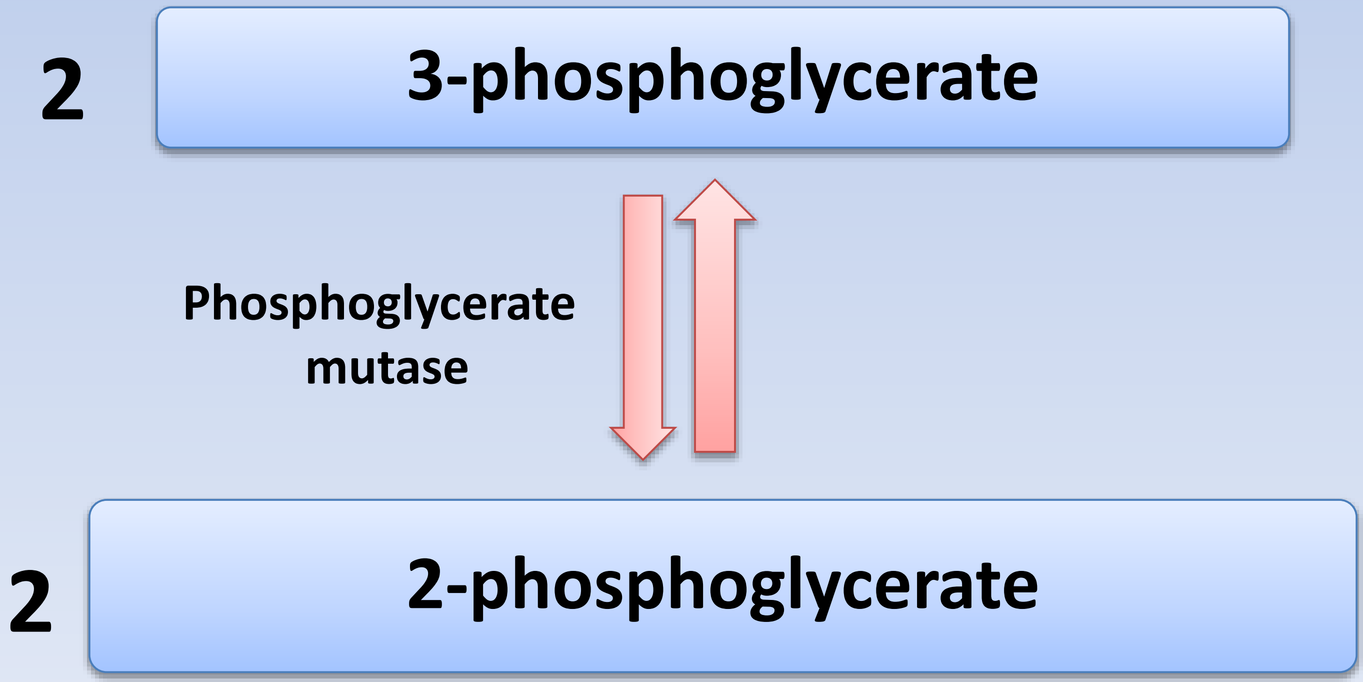
FIRST SUBSTRATE LEVEL PHOSPHORYLATION



2 **3-phosphoglycerate**

STEP 8

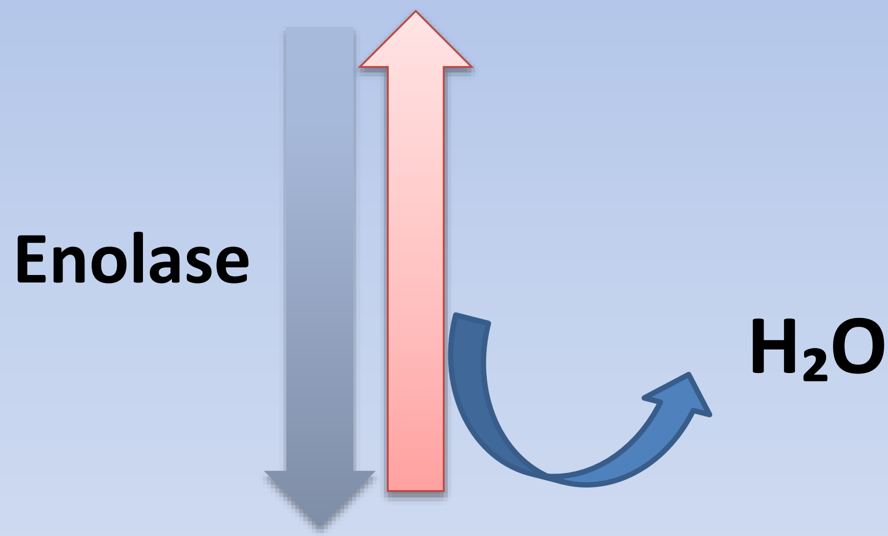
- The remaining phosphate-ester linkage in 3-phosphoglycerate, is moved from carbon 3 to carbon 2 ,because of relatively low free energy of hydrolysis, to form ***2-phosphoglycerate*****(2-PG).**



STEP 9: DEHYDRATION OF 2-PG

- This is the second reaction in glycolysis where a high-energy phosphate compound is formed.
- The 2-phosphoglycerate is dehydrated by the action of ***enolase*** to ***phosphoenolpyruvate(PEP)***. This compound is the phosphate ester of the enol tautomer of pyruvate.
- This is a reversible reaction.

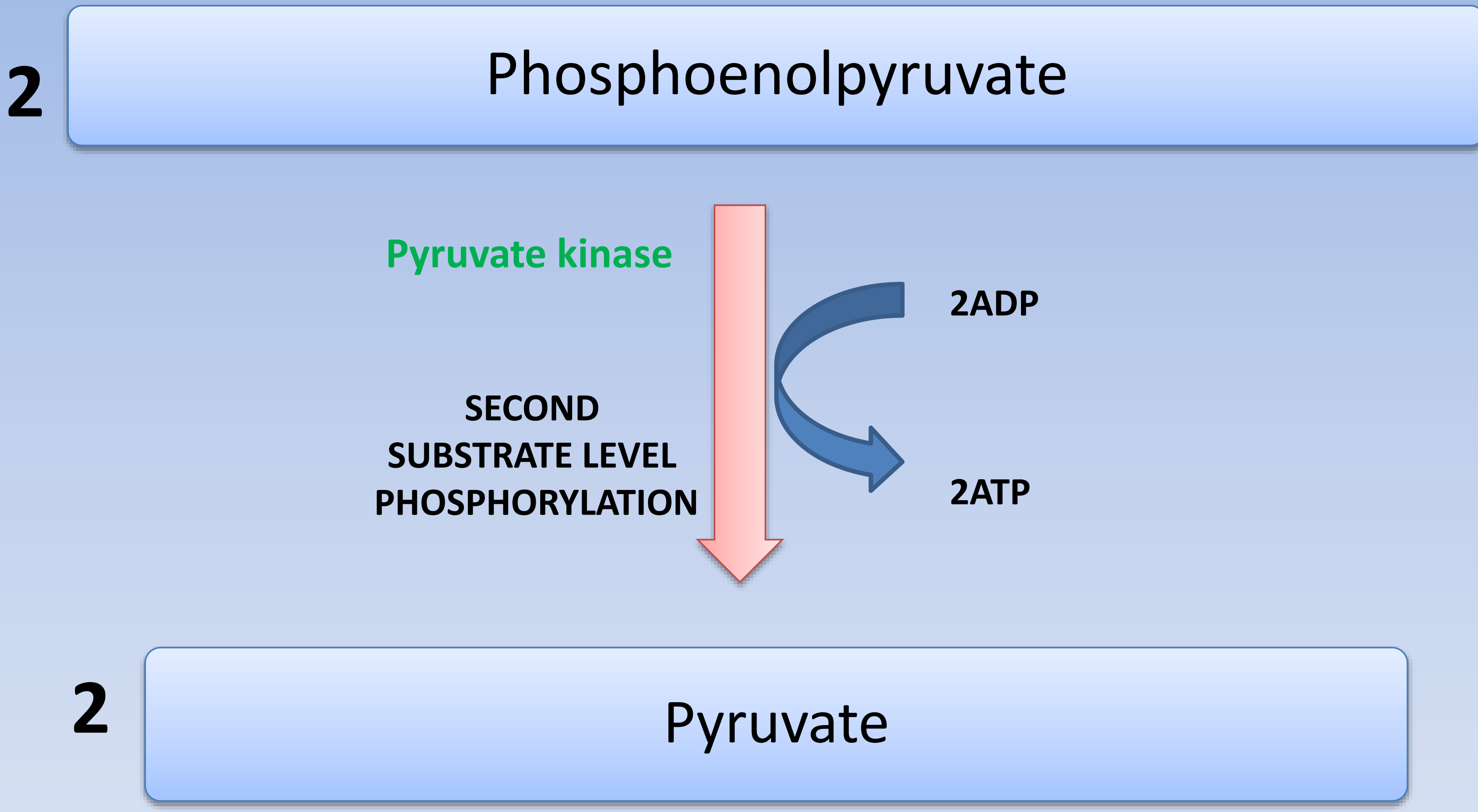
2 2-phosphoglycerate



2 Phosphoenol pyruvate

STEP 10: TRANSFER OF PHOSPHATE FROM PEP to ADP

- This last step is the irreversible transfer of high energy phosphoryl group from phosphoenolpuruvate to ADP.
- This reaction is catalyzed by ***pyruvate kinase.***
- This is the ***2nd substrate level phosphorylation*** reaction in glycolysis which yields ATP.
- This is a non-oxidative phosphorylation reaction.


2
Phosphoenolpyruvate
Pyruvate kinase
2ADP
SECOND
SUBSTRATE LEVEL
PHOSPHORYLATION
2ATP
2
Pyruvate

OVERALL BALANCE SHEET OF GLYCOLYSIS

- Each molecule of glucose gives 2 molecules of Glyceraldehyde-3-phosphate. Therefore , the total input of all 10 reactions can be summarized as:

$$\text{Glucose} + 2\text{ATP} + 2\text{Pi} + 2\text{NAD}^{+} + 2\text{H}^{+} + 4\text{ADP}$$

$$\downarrow$$

$$2\text{Pyruvate} + 2\text{H}^{+} + 4\text{ATP} + 2\text{H}_2\text{O} + 2\text{NADH} + 2\text{ADP}$$

On cancelling the common terms from the above equation, we get the net equation for Glycolysis:

Glucose+ 2Pi+ 2ADP+ $2NAD^+$

↓

2Pyruvate+ 2NADH+ 2ATP+ $2H^+$ + $2H_2O$

THUS THE SIMULTANEOUS REACTIONS INVOLVED IN GLYCOLYSIS ARE:

- ***Glucose is oxidized to Pyruvate***
- ***NAD^+ is reduced to NADH***
- ***ADP is phosphorylated to ATP***

- **ENERGY YIELD IN GLYCOLYSIS:**

STEP NO.	REACTION	CONSUMPTION of ATP	GAIN of ATP
1	Glucose → glucose-6-phosphate	1	-
3	Fructose-6-phosphate → fructose-1,6-biphosphate	1	-
7	1,3-diphosphoglycerate → 3-phosphoglycerate	-	1x2=2
10	Phosphoenolpyruvate → pyruvate	-	1x2=2
		2	4
		Net gain of ATP=4-2= 2	

-- REGULATION OF GLYCOLYSIS --

Three irreversible kinase reactions primarily drive glycolysis forward.

- hexokinase or glucokinase
- phosphofructokinase
- pyruvate kinase

Three of these enzymes regulate glycolysis as well.

2. HEXOKINASE

Phosphorylation of glucose.

◆Inhibited by its product, glucose 6-phosphate, as a response to slowing of glycolysis

Not GLUCOKINASE – as discussed

2. PHOSPHOFRUCTOKINASE

- major regulatory enzyme,

rate limiting for glycolysis

- an allosteric regulatory enzyme.
- measures adequacy of energy levels.

Inhibitors: ATP by decreasing fructose 6-phosphate binding

and **citrate**

both indicate high energy availability

- **Activators:** ADP, AMP, low energy

AMP and ADP reverse ATP inhibition

And another activator

Fructose 2,6 bisphosphate is a very important regulator, controlling the relative flux of carbon through glycolysis versus gluconeogenesis.
- It also couples these pathways to hormonal regulation.

3. PYRUVATE KINASE PEP + ADP→ pyruvate + ATP

- An allosteric tetramer

- **inhibitor:** ATP
- **inhibitors:** acetyl CoA and fatty acids (alternative fuels for TCA cycle)

- **activator:** fructose 1,6-bisphosphate ("feed-forward")

- Phosphorylation (inactive form) and dephosphorylation (active form) under **hormone control**.

- Also highly regulated at the level of gene expression ("carbohydrate loading")