King Saud University
College of Sciences
Biochemistry Department

الأيض (١)
Metabolism (1)
BCH 340

Lecture 8: Pentose phosphate pathway

Intended learning outcomes (ILOs)

By the end of this lecture, students will be able to:

- Explain the overall purpose and significance of the pentose phosphate pathway (PPP) within cellular metabolism.
- Describe the substrates entering the PPP and the products generated, including the interconversion of sugars and production of reducing equivalents.
- Understand the regulation of the PPP, including allosteric regulation and hormonal control.
- Describe the biochemical pathway of uronic acid synthesis, including its intermediates, enzymes involved, and regulatory mechanisms.

Pentose phosphate pathway

- The pentose phosphate pathway (PPP), is also known as the hexose monophosphate shunt or the phosphogluconate pathway.
- It occurs entirely in the cytoplasm of most cells, particularly in liver, adipose tissue, adrenal cortex, and lactating mammary glands.
- The PPP is metabolic pathway parallel to glycolysis (i.e. an alternate route for the oxidation of glucose where ATP (energy) is neither produced nor utilized).
- The PPP plays a critical role in cellular metabolism, the primary functions of this pathway include:
  - Generation of NADPH
  - Synthesis of ribose-5-phosphate

Importance of PPP

Generation of NADPH:

- NADPH (nicotinamide adenine dinucleotide phosphate, a reducing equivalent) is essential for biosynthetic processes such as fatty acid synthesis and the maintenance of cellular redox balance.
- It is also crucial for the regeneration of reduced glutathione, which protects cells from oxidative stress.

Synthesis of ribose-5-phosphate:

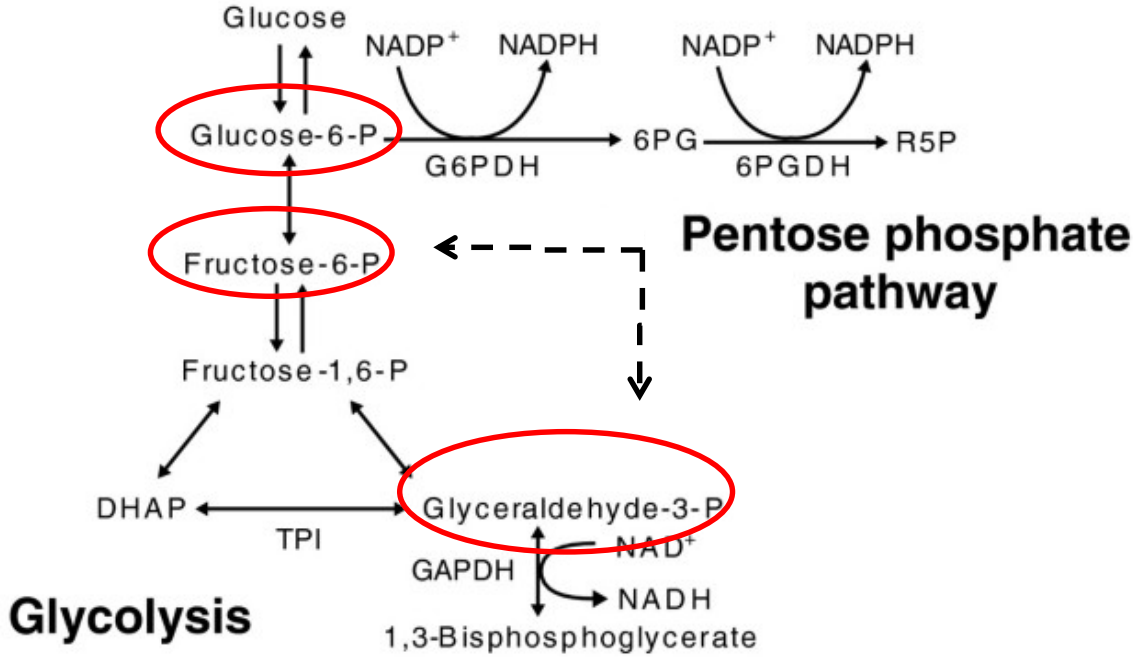
- Ribose-5-phosphate is a precursor for the synthesis of nucleotides (for DNA and RNA).
- This is particularly important in rapidly dividing cells, such as those in the bone marrow or during tissue regeneration.

Pentose phosphate pathway

- The PPP is an alternative metabolic pathway to glycolysis.
  - While both pathways involve the metabolism of glucose, they serve different purposes and occur under different conditions within the cell.
- Glycolysis takes place in almost all cells, while the PPP is particularly active in tissues with high biosynthetic demands, such as liver and adipose tissue.

Pentose phosphate pathway (cont.)

- The PPP branches off from the glycolytic pathway at the level of glucose-6-phosphate.
- The PPP ultimately feeds back into glycolysis (or gluconeogenesis) through intermediates like fructose-6-phosphate and glyceraldehyde-3-phosphate.



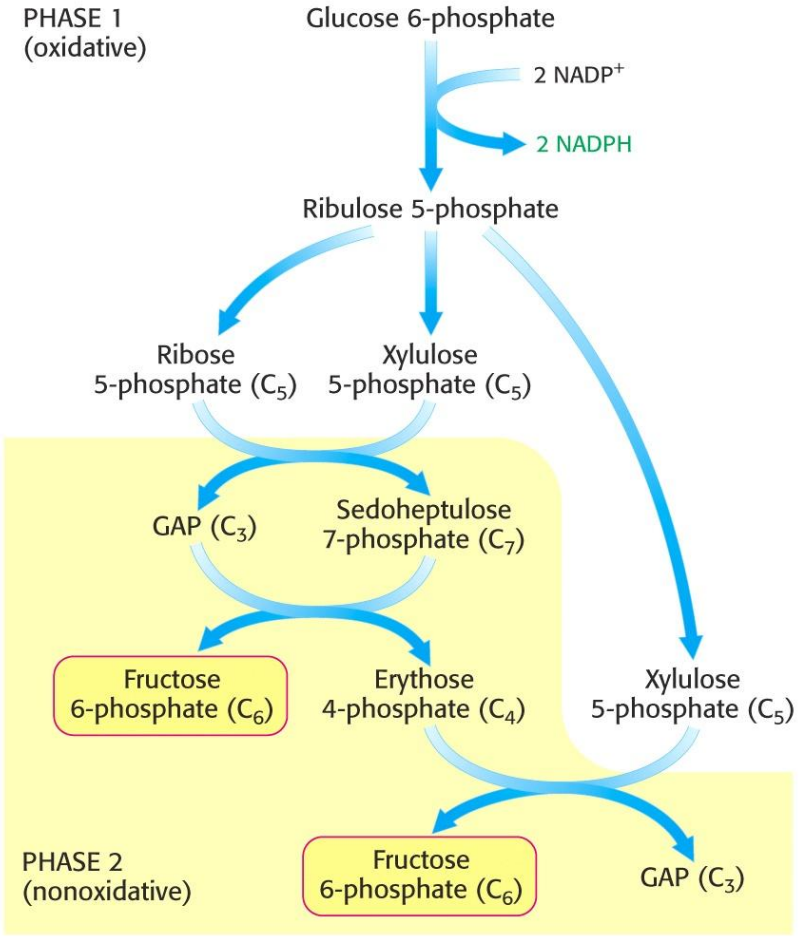
Phases of pentose phosphate pathway

Oxidative phase (irreversible):

- This phase allows the production of NADPH, which is essential for biosynthetic reactions, such as fatty acid synthesis and for detoxification of reactive oxygen species.

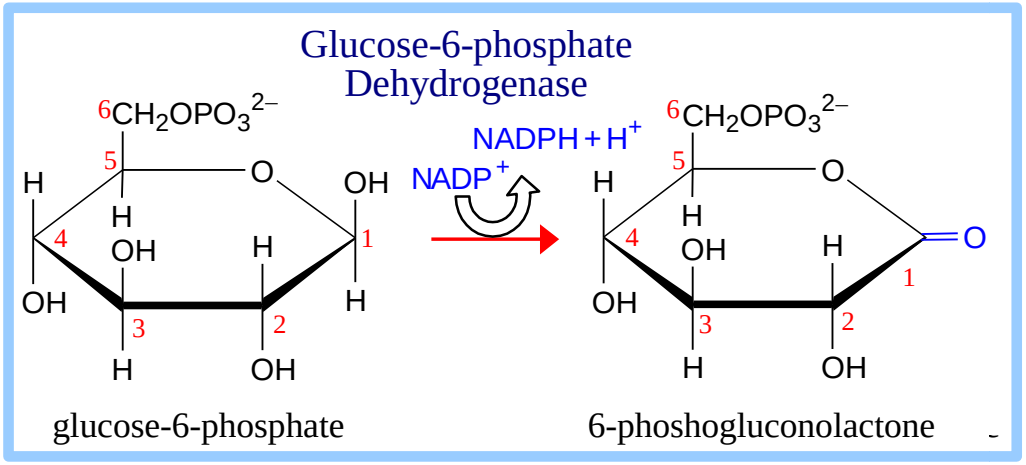
Non-oxidative phase (reversible):

- This phase generates ribose-5-phosphate, which is a precursor for the synthesis of nucleotides.
- It also enables the synthesis of various sugar intermediates used in other metabolic pathways.



The oxidative phase of PPP

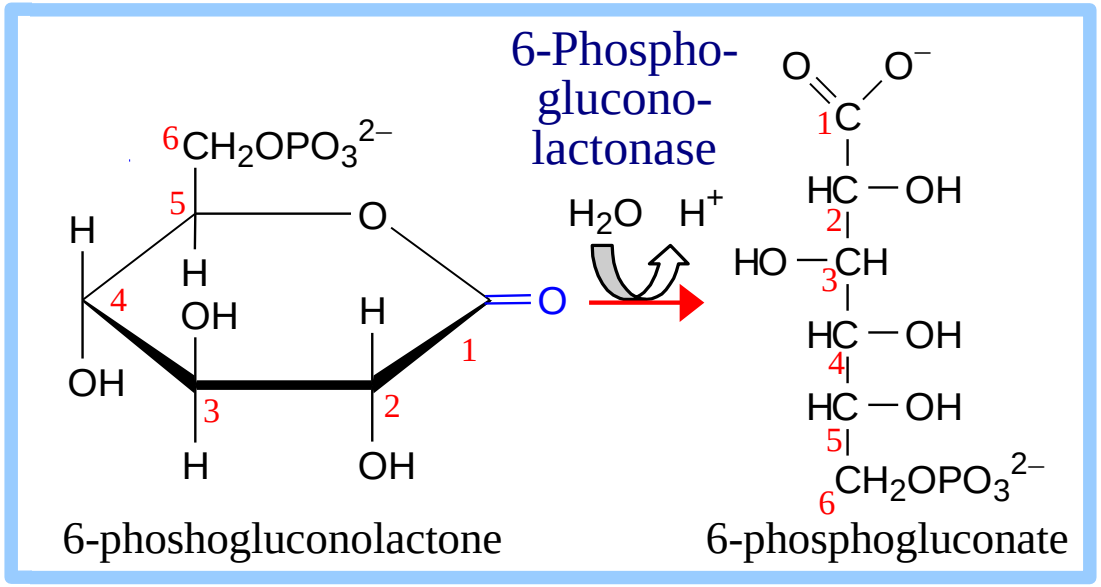
- The oxidative phase involves the conversion of glucose-6-phosphate to ribulose-5-phosphate, producing NADPH in the process.
- The enzyme glucose-6-phosphate dehydrogenase catalyzes the first and rate-limiting step of this phase.
- This enzyme oxidizes glucose-6-phosphate to 6-phosphoglucono-δ-lactone, accompanied by the reduction of $NADP^+$ to NADPH.
- Once produced, 6-phosphoglucono-δ-lactone has no other metabolic fate in the cell but to be converted to 6-phosphogluconate.



The oxidative phase of PPP (cont.)

- The second reaction involves the action of the enzyme 6-phosphogluconolactonase.
- This enzyme catalyzes the rapid hydrolysis of 6-phosphoglucono-δ-lactone to 6-phosphogluconate as following:

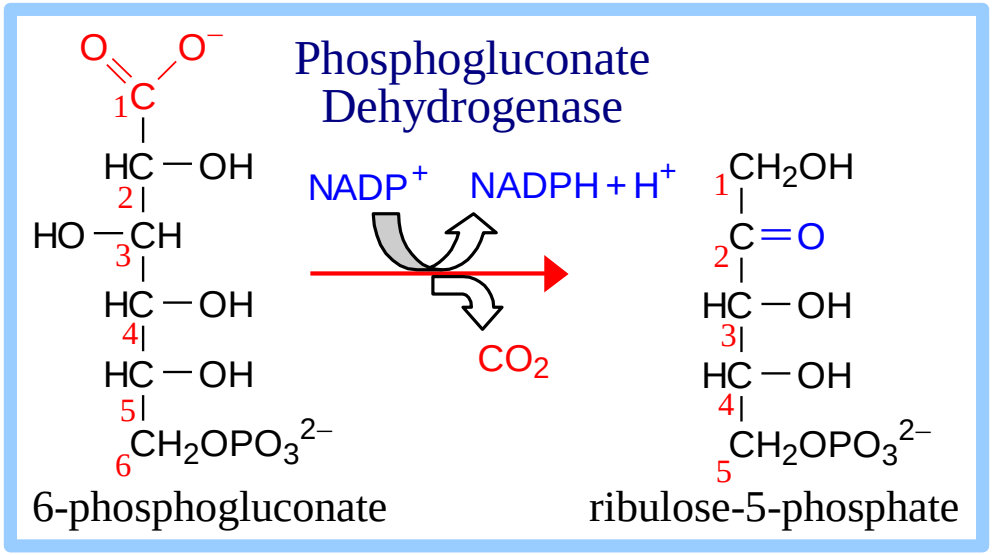
6-phosphoglucono-δ-lactone + H2O → 6-phosphogluconate



The oxidative phase of PPP (cont.)

- The third and final reaction of the oxidative phase of PPP is catalyzed by 6-phosphogluconate dehydrogenase.
- This reaction involves the oxidative decarboxylation of 6-phosphogluconate to yield ribulose-5-phosphate (a ketopentose) and another molecule of NADPH. The reaction is as follows:

6-phosphogluconate + $NADP^+$ → ribulose-5-phosphate + CO_2 + NADPH



The non-oxidative phase of PPP

- The non-oxidative phase involves interconversions of several sugar phosphates, particularly those with three to seven carbon atoms.
- Through a series of reversible reactions, this phase can generate ribose-5-phosphate and other sugars that can be utilized for nucleotide synthesis or as intermediates in other metabolic pathways, including glycolysis and gluconeogenesis.
- The two enzymes transketolase and transaldolase catalyze the rearrangement and transfer of carbon units between different sugars.

The non-oxidative phase of PPP (cont.)

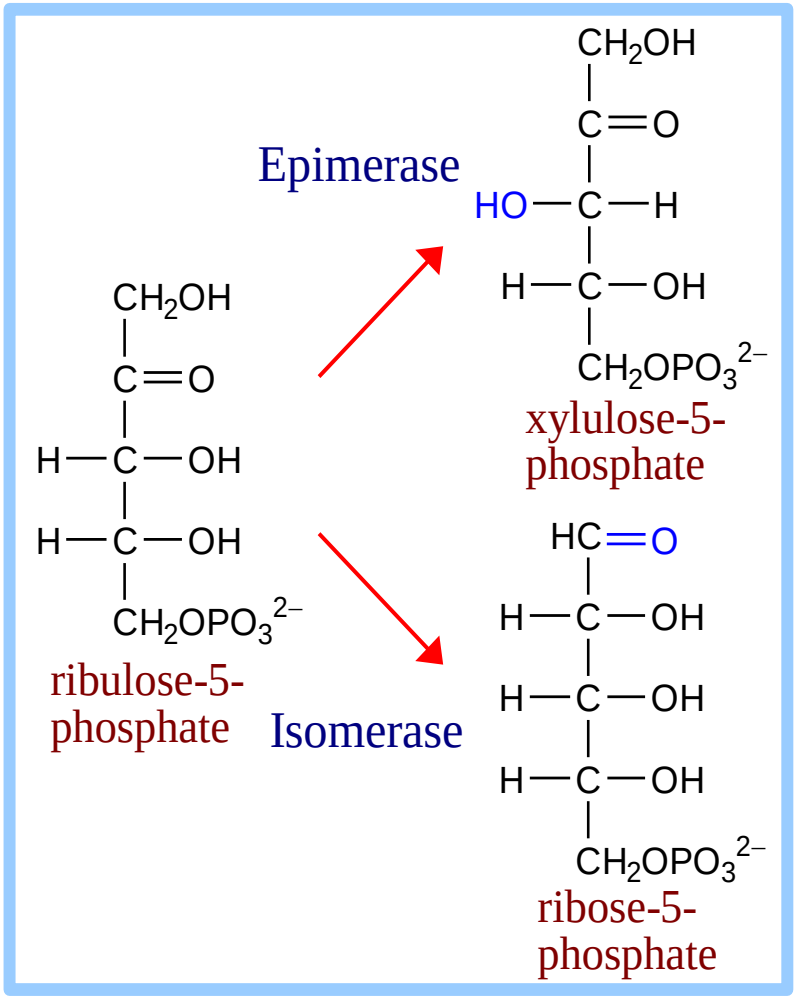
Ribulose-5-phosphate epimerization:

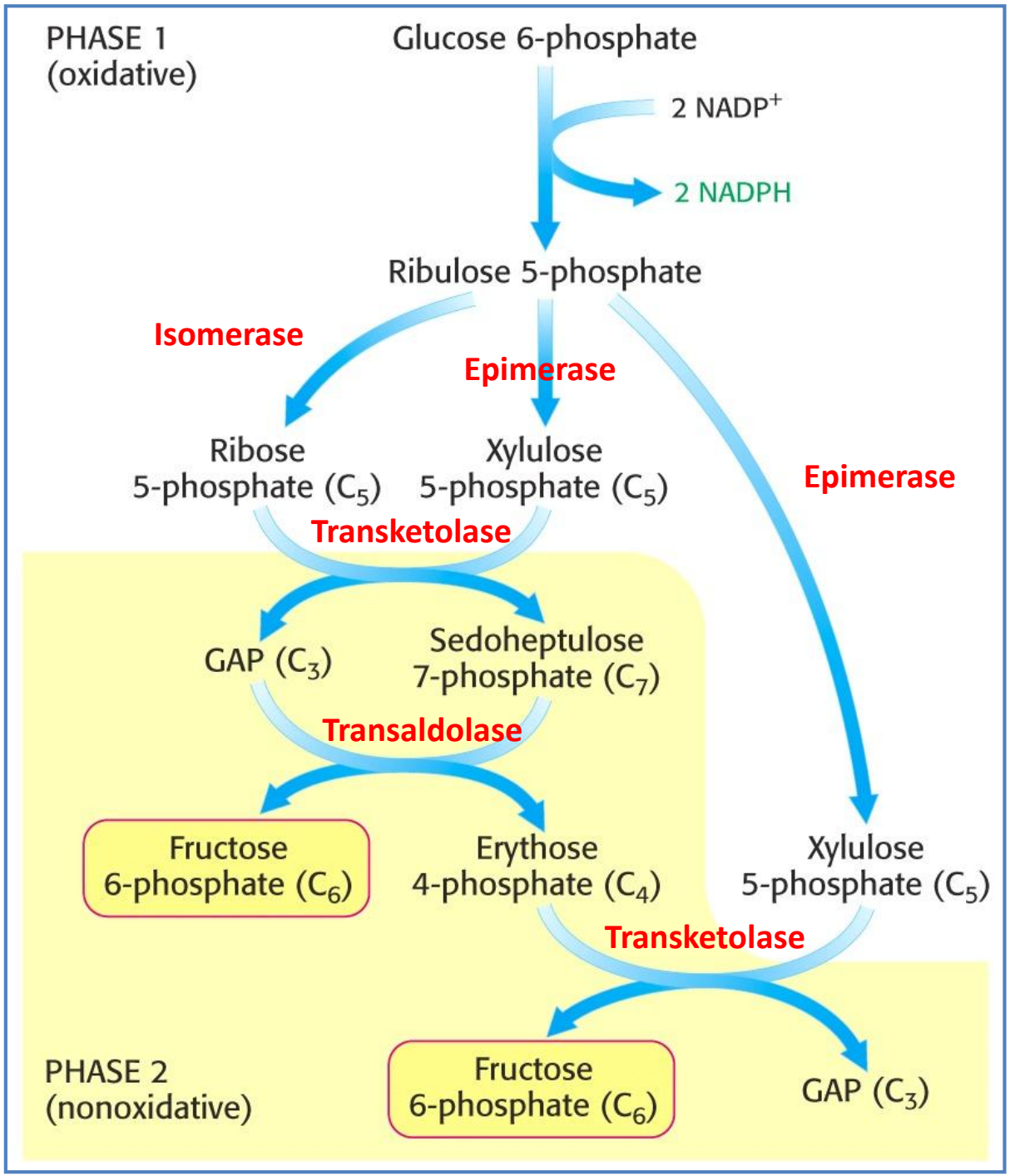
- Ribulose-5-phosphate is converted into xylulose-5-phosphate by the enzyme ribulose-5-phosphate epimerase.

Ribulose-5-phosphate isomerization:

- Ribulose-5-phosphate can be converted into ribose-5-phosphate through the enzyme ribose-5-phosphate isomerase.

- Ribose-5-phosphate can be used for nucleotide biosynthesis or it can regenerate glucose-6-phosphate for another round of the oxidative phase.




PHASE 1
(oxidative)
Glucose 6-phosphate
2 NADP+
2 NADPH
Ribulose 5-phosphate
Isomerase
Epimerase
Epimerase
Ribose
5-phosphate (C5)
Xylulose
5-phosphate (C5)
Transketolase
GAP (C3)
Sedoheptulose
7-phosphate (C7)
Transaldolase
Fructose
6-phosphate (C6)
Erythose
4-phosphate (C4)
Xylulose
5-phosphate (C5)
Transketolase
PHASE 2
(nonoxidative)
Fructose
6-phosphate (C6)
GAP (C3)

The non-oxidative phase of PPP (cont.)

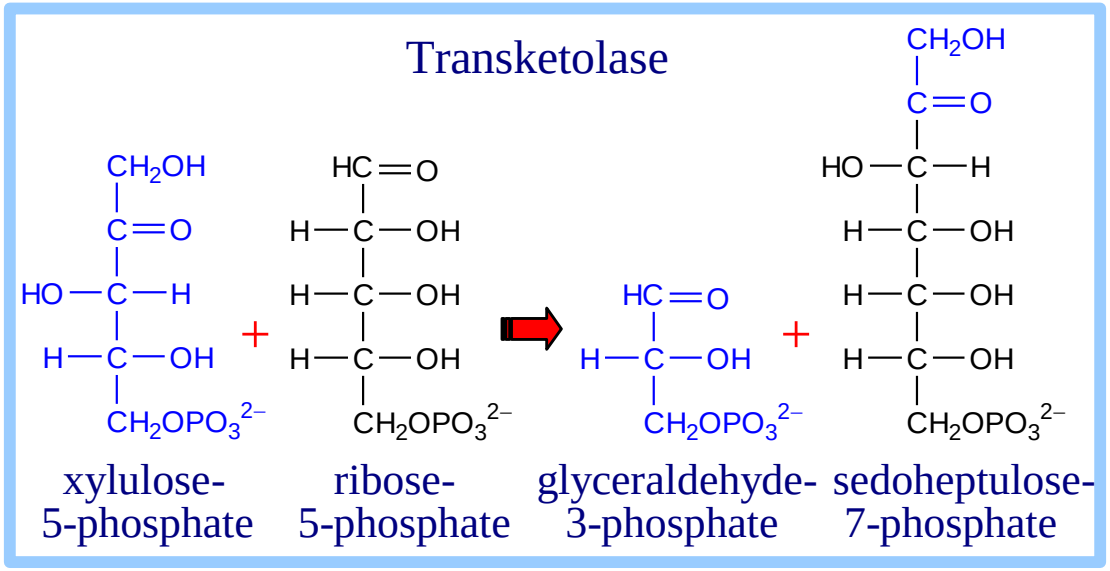
Transketolase reactions:

- Transketolase catalyzes the transfer of a two-carbon ketol (hydroxy ketone) group from a ketose donor to an aldose acceptor, generating a new ketose and aldose.
- This series of reactions involves the transfer of carbon units and serves to interconvert pentose phosphates and hexose phosphates.
- The two-carbon unit is transferred form xylulose-5-phosphate to either:
  - Erythrose-4-phosphate, producing fructose-6-phosphate and glyceraldehyde-3-phosphate.
  - Ribose-5-phosphate, yielding sedoheptulose-7-phosphate and glyceraldehyde-3-phosphate.

The non-oxidative phase of PPP (cont.)

Transketolase reactions (cont.):

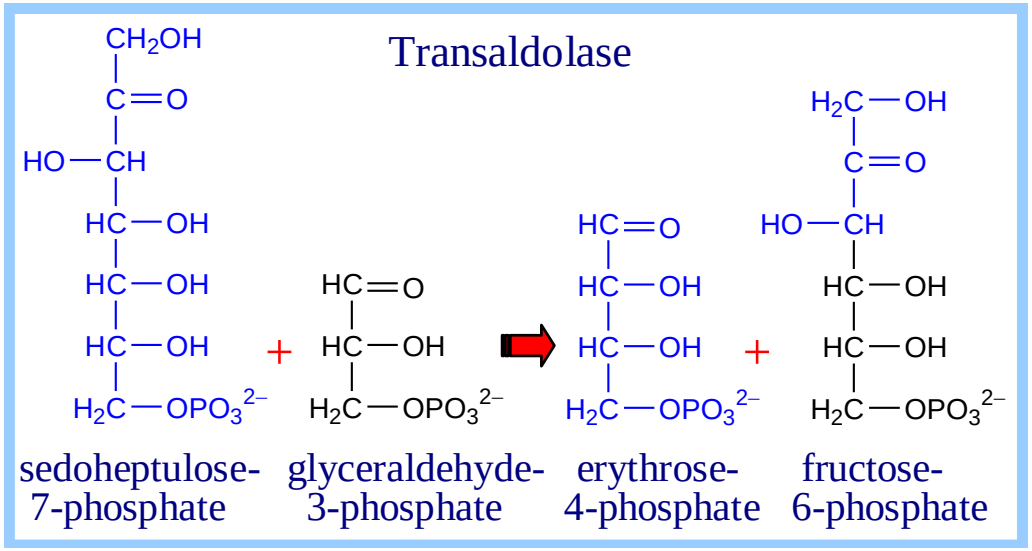
- The transketolase enzyme requires:
  - Mg^{2+} ions to help in stabilizing the enzyme-substrate complex and facilitating the reaction.
  - Thiamine pyrophosphate (TPP), which plays a crucial role in the transfer of two-carbon units in various metabolic reactions.

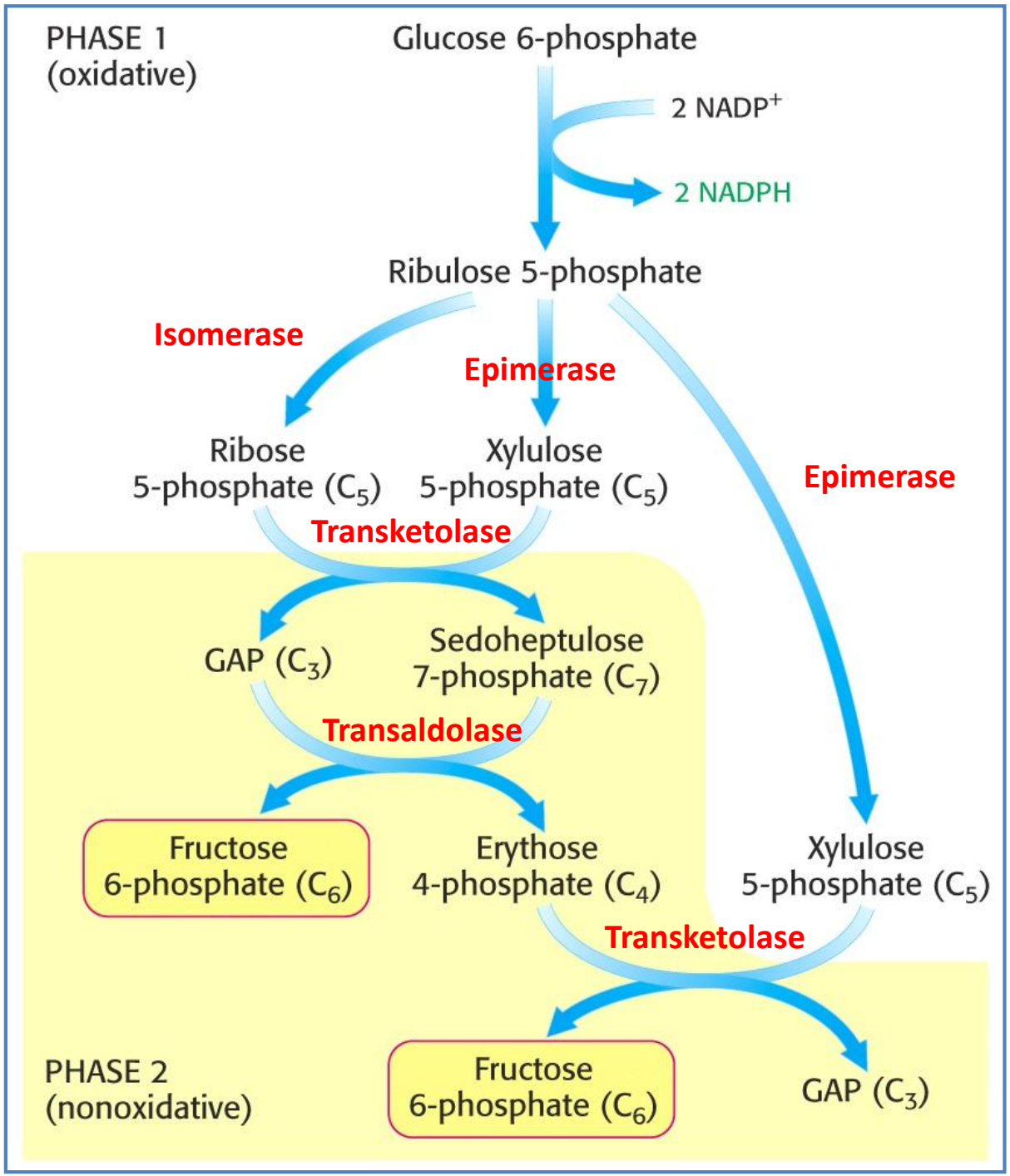


The non-oxidative phase of PPP (cont.)

Transaldolase reactions:

- Transaldolase catalyzes the transfer of a three-carbon dihydroxyacetone group from a ketose donor to an aldose acceptor, generating a new ketose and a new aldose.
- The three-carbon unit is transferred from sedoheptulose-7-phosphate to glyceraldehyde-3-phosphate, resulting in the formation of fructose-6-phosphate and erythrose-4-phosphate.




PHASE 1
(oxidative)
Glucose 6-phosphate
2 NADP+
2 NADPH
Ribulose 5-phosphate
Isomerase
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Xylulose
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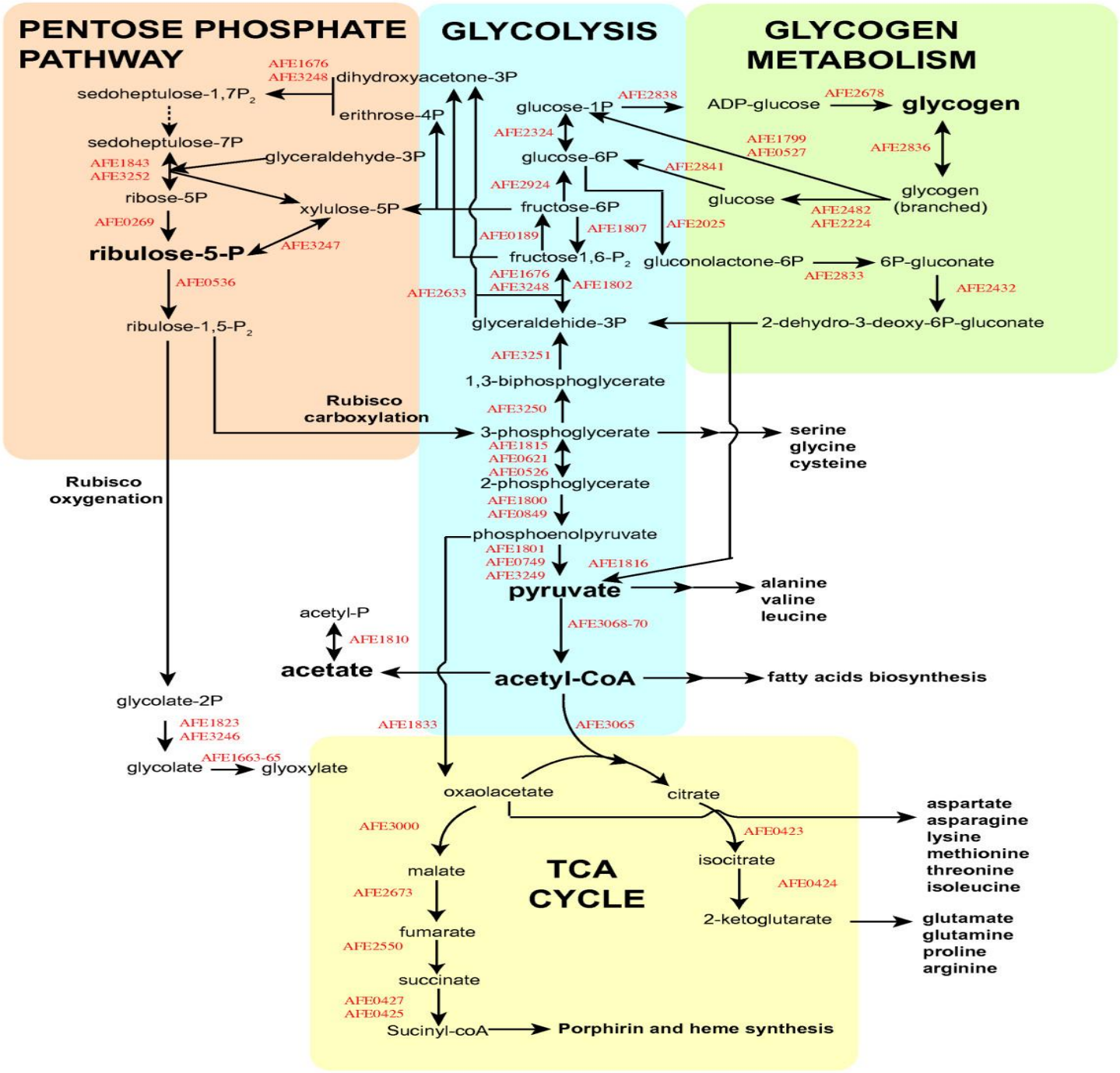
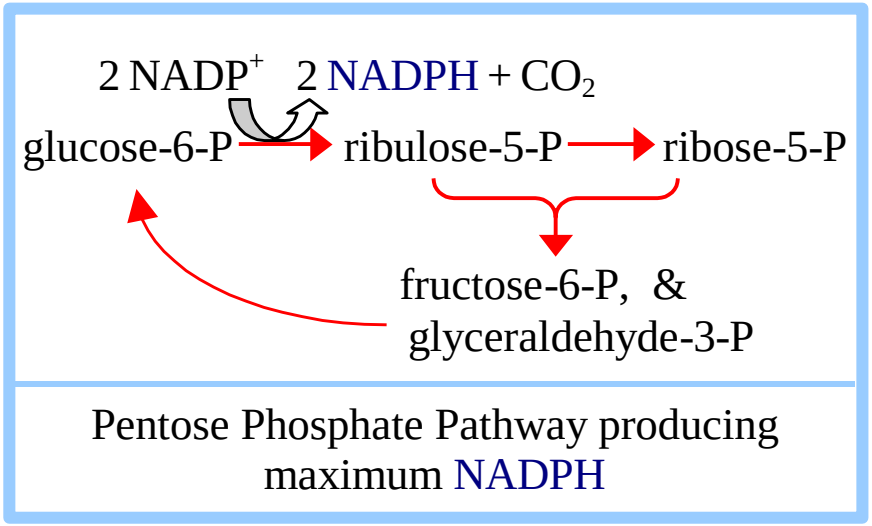
Sedoheptulose
7-phosphate (C7)
Transaldolase
Fructose
6-phosphate (C6)
Erythose
4-phosphate (C4)
Xylulose
5-phosphate (C5)
Transketolase
PHASE 2
(nonoxidative)
Fructose
6-phosphate (C6)
GAP (C3)

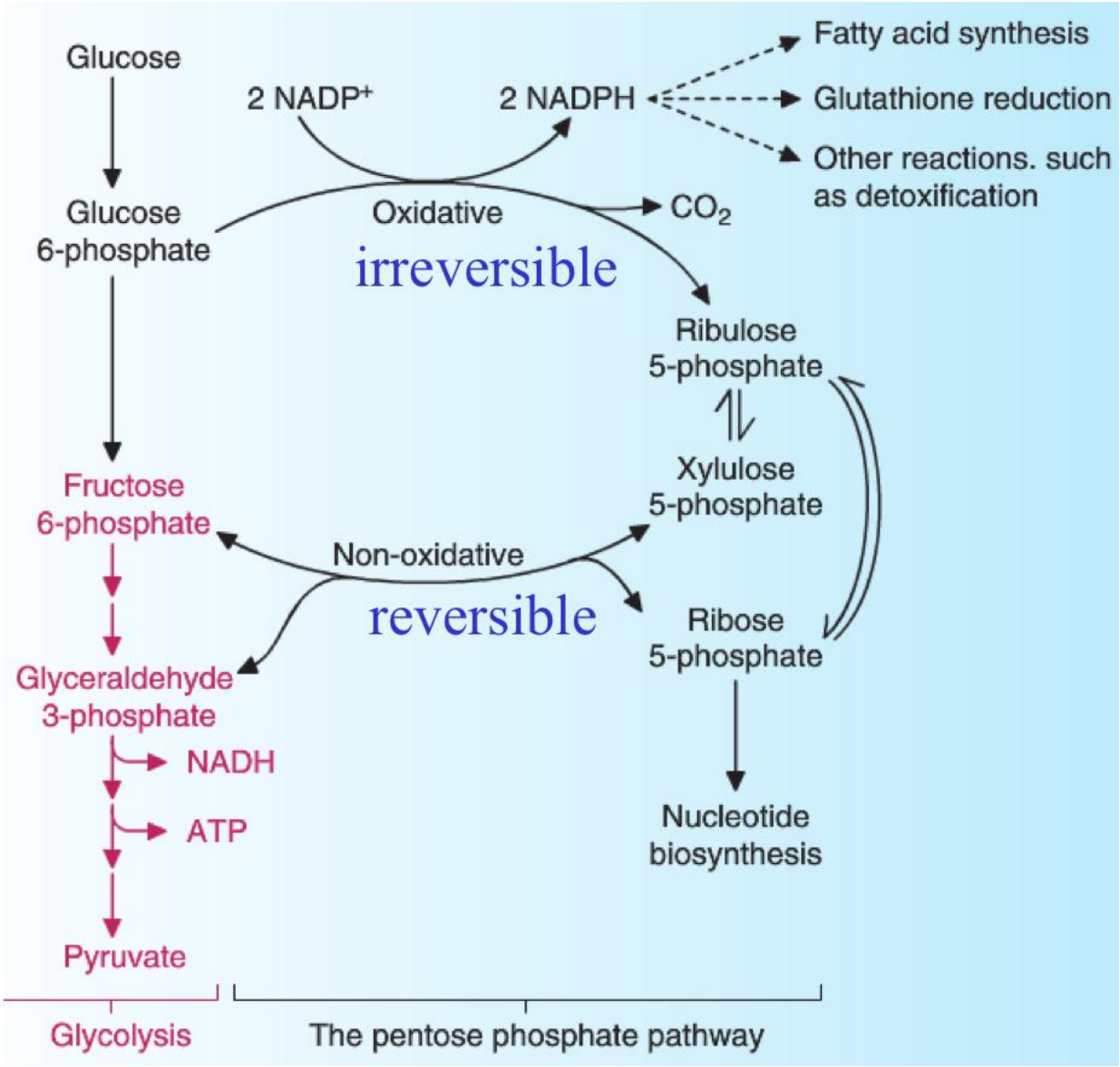

PENTOSE PHOSPHATE PATHWAY
GLYCOLYSIS
GLYCOGEN METABOLISM
AFE1676
AFE3248
dihydroxyacetone-3P
sedoheptulose-1,7P2
erithrose-4P
sedoheptulose-7P
AFE1843
AFE3252
glyceraldehyde-3P
ribose-5P
xylulose-5P
AFE0269
ribulose-5-P
AFE3247
AFE0536
ribulose-1,5-P2
Rubisco carboxylation
Rubisco oxygenation
AFE2838
glucose-1P
ADP-glucose
AFE2678
glycogen
AFE2324
glucose-6P
AFE1799
AFE0527
AFE2836
AFE2841
AFE2924
fructose-6P
glucose
glycogen (branched)
AFE2482
AFE2224
AFE0189
AFE1807
AFE2025
fructose1,6-P2
gluconolactone-6P
AFE2833
6P-gluconate
AFE1676
AFE3248
AFE1802
AFE2633
AFE2432
glyceraldehide-3P
2-dehydro-3-deoxy-6P-gluconate
AFE3251
1,3-biphosphoglycerate
AFE3250
3-phosphoglycerate
serine
glycine
cysteine
AFE1815
AFE0621
AFE0526
2-phosphoglycerate
AFE1800
AFE0849
phosphoenolpyruvate
AFE1801
AFE0749
AFE3249
AFE1816
pyruvate
alanine
valine
leucine
acetyl-P
AFE1810
AFE3068-70
acetate
acetyl-CoA
fatty acids biosynthesis
glycolate-2P
AFE1823
AFE3246
AFE1833
AFE3065
AFE1663-65
glycolate
glyoxylate
oxaolacetate
citrate
aspartate
asparagine
lysine
methionine
threonine
isoleucine
AFE3000
AFE0423
malate
TCA CYCLE
isocitrate
AFE0424
AFE2673
2-ketoglutarate
glutamate
glutamine
proline
arginine
fumarate
AFE2550
succinate
AFE0427
AFE0425
Sucinyl-coA
Porphirin and heme synthesis

Maximizing the production of NADPH

- Glyceraldehyde-3-phosphate and fructose-6-phosphate (generated in the PPP) can be converted into glucose-6-phosphate (through gluconeogenesis), which can then:
  - Reenter the PPP, maximizing the production of NADPH and ribose-5-phosphate.
  - Enter glycolysis for the production of ATP.



Pentose phosphate pathway and its link to glycolysis

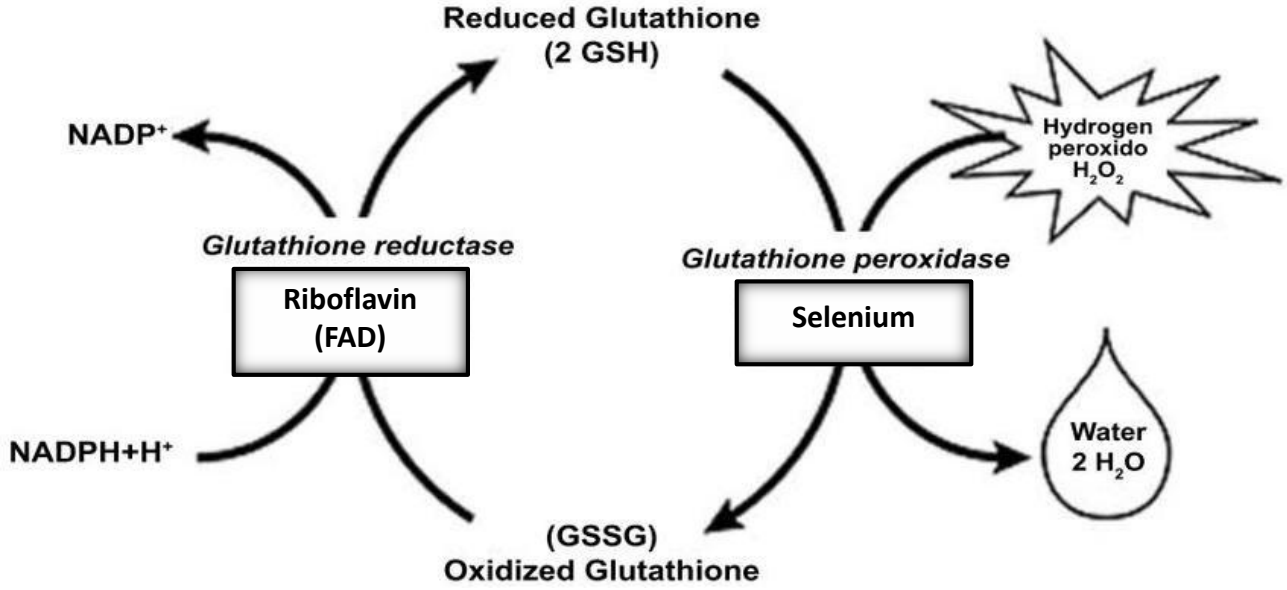


Importance of PPP to erythrocytes

- RBCs are particularly vulnerable to oxidative stress due to their high metabolic activity and exposure to high oxygen levels.
- The primary function of the PPP in erythrocytes is to generate NADPH, a reducing agent.
- NADPH is essential to maintain glutathione (GSH) and thioredoxin in their reduced states, which is critical for protecting RBCs from oxidative damage.
- Glutathione serves as a major antioxidant in erythrocytes, protecting them from oxidative damage caused by reactive oxygen species (ROS) generated during normal cellular metabolism and exposure to environmental stressors.

Importance of PPP to erythrocytes (cont.)

- The PPP provides NADPH, which is necessary for the regeneration of reduced glutathione (GSH) from its oxidized form (GSSG), thus maintaining the cellular antioxidant defense system.



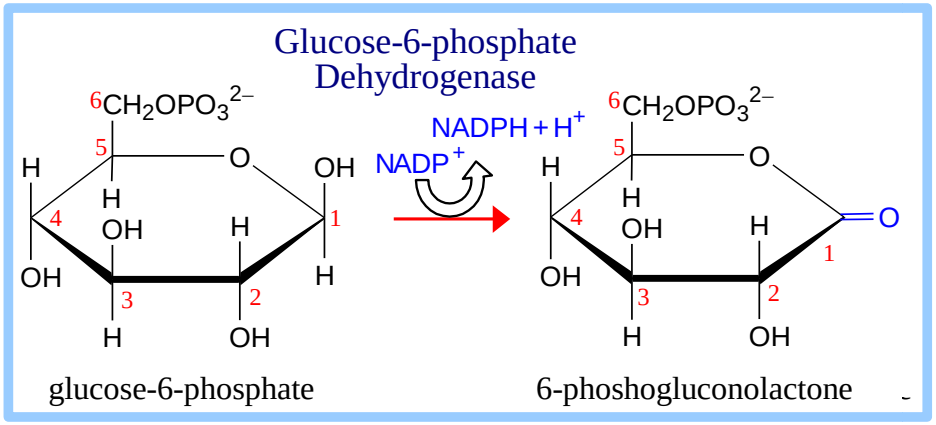
- NADPH is required to keep glutathione in the reduced state, and glucose-6-phosphate dehydrogenase is needed to reduce $NADP^+$ to NADPH.

Regulation of PPP

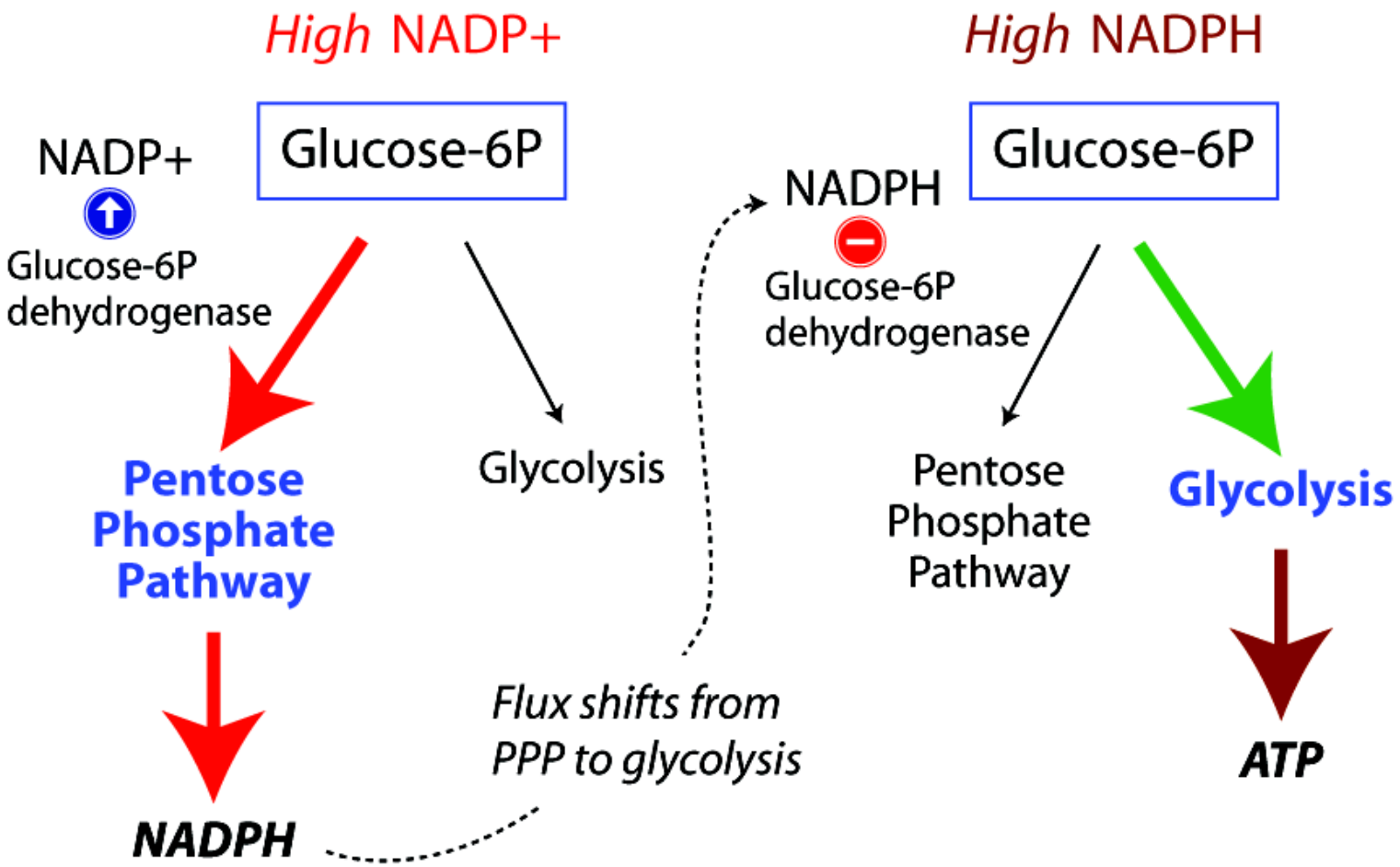
- The entry of glucose 6-phosphate into the pentose phosphate pathway is controlled by the cellular demand for ribose-5-phosphate (for nucleotide synthesis) and NADPH (for biosynthetic reactions or antioxidant defenses).
- The regulation of the PPP involves various factors, including:
  - The availability of glucose 6-phosphate (substrate or the first enzyme of the pathway, glucose 6-phosphate dehydrogenase).
  - The energy status of the cell (such as the ratio of ATP to ADP/AMP).
  - The levels of $NADP^+$ and NADPH.
  - Other factors such as hormones and nutrients.

Regulation of PPP (cont.)

- The glucose-6-phosphate dehydrogenase enzyme requires Mg^{2+} and $NADP^+$ as coenzymes.
- The activity of the rate-limiting enzyme, glucose-6-phosphate dehydrogenase, is allosterically regulated by $NADPH/NADP^+$:
  - NADPH is an allosteric inhibitor of the enzyme, providing feedback inhibition to prevent excessive NADPH production.
  - Conversely, $NADP^+$ can act as an allosteric activator, stimulating the enzyme activity.



Regulation of glucose-6-phosphate dehydrogenase activity controls flux through the glycolytic pathway and pentose phosphate pathways



Regulation of PPP (cont.)

- The increased insulin/glucagon ratio (after a high-carbohydrate meal) influences the synthesis of glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase, promoting glucose utilization through the PPP.
- The deficiency of glucose-6-phosphate dehydrogenase leads to the impairment of NADPH production and the inhibition of the detoxification process (resulting in cell damage).
  - Individuals with this deficiency have erythrocytes that are more prone to breaking down prematurely, a condition known as hemolytic anemia.

Differences between PPP and glycolysis

	PPP	Glycolysis
Location	In certain cells	In all cells
Oxidation of glucose	Occurs in the first reaction	Phosphorylation occurs first then oxidation
Coenzyme	$NADP^+$	NAD^+
Energy	No energy production	8 ATP
CO2	Produced	Not produced
Pentose sugars	Produced	Not produced

Uronic Acid Pathway

Uronic acid pathway

- The uronic acid pathway (also known as the glucuronic acid pathway) is a metabolic pathway that mainly occurs in the cytoplasm of cells in the liver.
- The uronic acid pathway involves the conversion of glucose to glucuronic acid, followed by the conjugation of glucuronic acid with xenobiotics and waste products to facilitate their excretion.
- Like the pentose phosphate pathway, the uronic acid pathway does not lead to generation of ATP, however, it uses ATP for the generation of the activated form of glucuronate (UDP-glucuronate).

Importance of the uronic acid pathway

- The uronic acid pathway plays diverse and essential roles in metabolism, detoxification, and physiological processes. These roles include:

1. Synthesis of substances:

- Glucuronic acid is an important precursor molecule for the synthesis of substances such as glycosaminoglycans (hyaluronic acid and heparin) and ascorbic acid (vitamin C).

2. Conjugation reactions:

- UDP-glucuronic acid is conjugated with various endogenous and exogenous compounds, making them more water-soluble to facilitate their excretion (e.g. UDP-glucuronic conjugates with bilirubin to form bilirubin glucuronide, which is excreted in the bile).

Importance of the uronic acid pathway (cont.)

3. Detoxification reactions:

- One of the primary functions of the uronic acid pathway is the detoxification and elimination of various toxic and foreign compounds (xenobiotics) from the body.

- Through a process called glucuronidation, UDP-glucuronic acid is conjugated to drugs, toxins, and other harmful substances, rendering them less toxic and facilitating their excretion via the urine or bile.

Formation of UDP-glucuronic acid

- The pathway begins with the phosphorylation of glucose to glucose-6-phosphate by the enzyme hexokinase or glucokinase.
  - This step traps glucose within the cell and primes it for further metabolic processing.
- Glucose-6-phosphate is first converted to glucose-1-phosphate by phosphoglucomutase, which then reacts with UTP to form UDP-glucose in a reaction catalyzed by UDP-glucose pyrophosphorylase.
- Finally, UDP-glucose is oxidized to UDP-glucuronic acid by UDP-glucose dehydrogenase.

Fate of glucuronic acid

- UDP-glucuronate can be converted to glucuronate by the enzyme UDP-glucuronate 4-epimerase.
- Glucuronate can then enter various metabolic pathways depending on cellular needs.
  - It can be converted to D-xylulose-5-phosphate through a series of reactions.
  - D-xylulose-5-phosphate enter the pentose phosphate pathway for further metabolism.

Summary

- The pentose phosphate pathway is a metabolic pathway that operates alongside glycolysis, generating NADPH and ribose 5-phosphate for nucleotide synthesis, lipid biosynthesis, and antioxidant defense.

- The uronic acid pathway is a metabolic route primarily involved in the conversion of glucose to various uronic acids, such as glucuronic acid. These uronic acids serve essential functions in detoxification, conjugation of xenobiotics, and extracellular matrix formation.