CDL Advances Chemical Studies (ACS)

Metabolic Biochemistry 7 CFU = 6+1

Lecture

Laboratory

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The Pentose Phosphate Pathway

Pentose phosphate pathway

Phosphogluconate pathway

Hexose Monophosphate Shunt (HMP Shunt)

Metabolic pathway parallel to glycolysis

The Pentose Phosphate Pathway

About half of the glucose metabolized by the liver enters the pentose phosphate pathway

It occurs in the cytoplasm of cells.

Neither ATP is consumed nor directly formed.

Functions:

Produce NADPH

Produce pentoses (ribose 5P, nucleotide synthesis)

Convert pentoses into glycolytic intermediates

The Pentose Phosphate Pathway

Its primary role is anabolic rather than catabolic

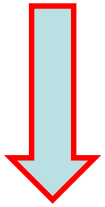
The pathway provides NADPH for reductive biosynthesis and ribose-5-phosphate for nucleotide biosynthesis in the quantities that the cell requires.

NADPH

Reducing power used for reductive biosynthesis (fatty acid, cholesterol, steroid hormon) and to counteract oxidative damage (erythrocyte, lens, cornea)

NADPH derives from vitamin B3

ribose-5-phosphate

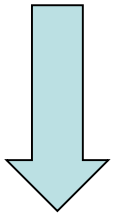


Nucleotid synthesis in cells that have high division velocity

The pathway is especially important in red blood cells (erythrocytes).

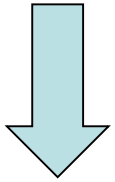
Require the NADPH provided by this pathway.

Tissues that carry out extensive fatty acid synthesis



liver, adipose, lactating mammary gland

Tissue very active synthesis of cholesterol and steroid hormone



liver, adrenal glands, gonads

Rapidly dividing cells

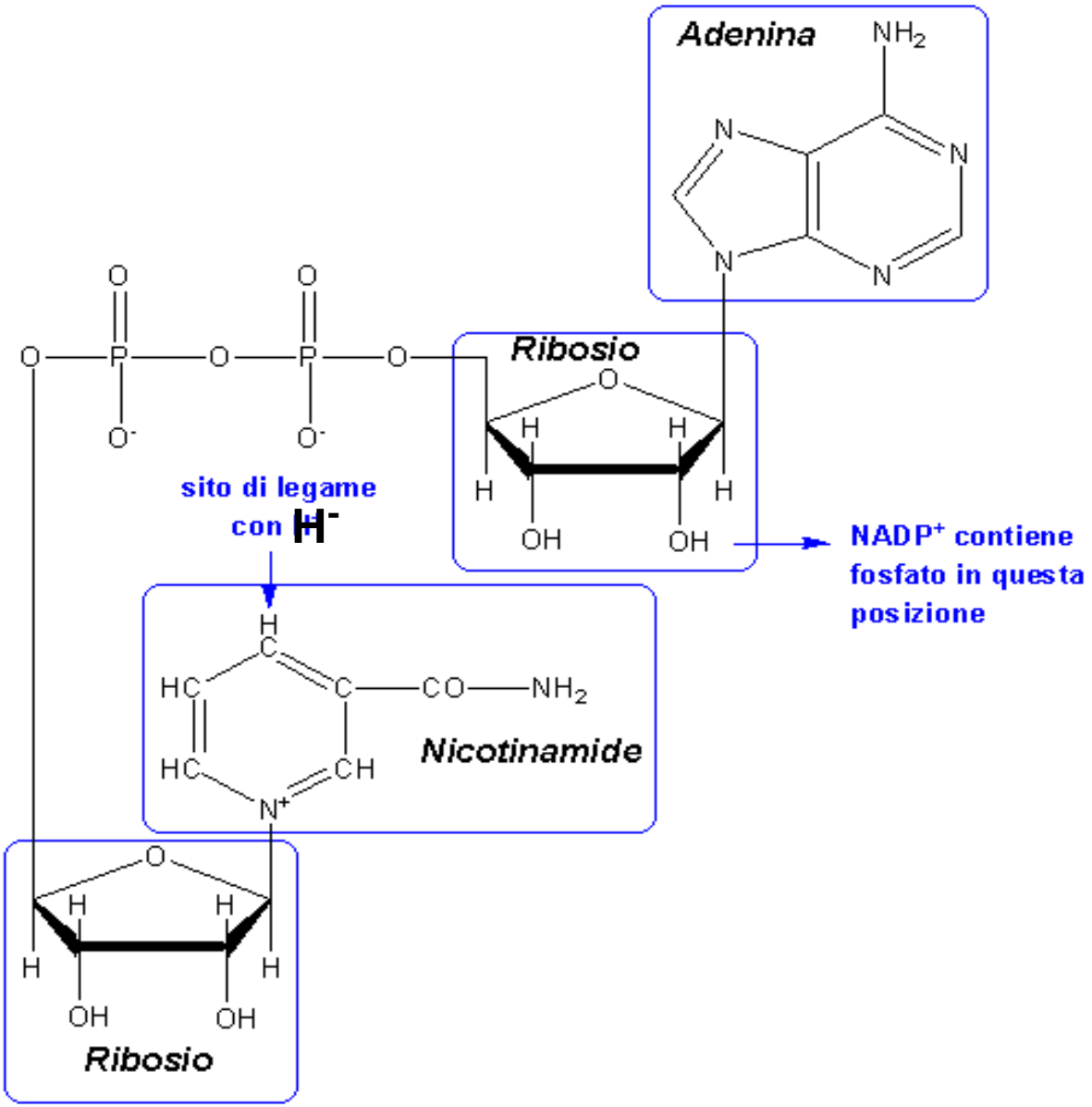
- bone marrow
- skin
- intestinal mucosa
- tumors

Rapidly dividing cells use the pentose ribose 5-phosphate to make

- RNA, DNA
- coenzymes (ATP, NADH, FADH2, and coenzyme A)

NAD $^{+}$ e NADP^{+}

Trasferiscono un
H^-
forma
fosforilata=
$NADP^+$



NADPH

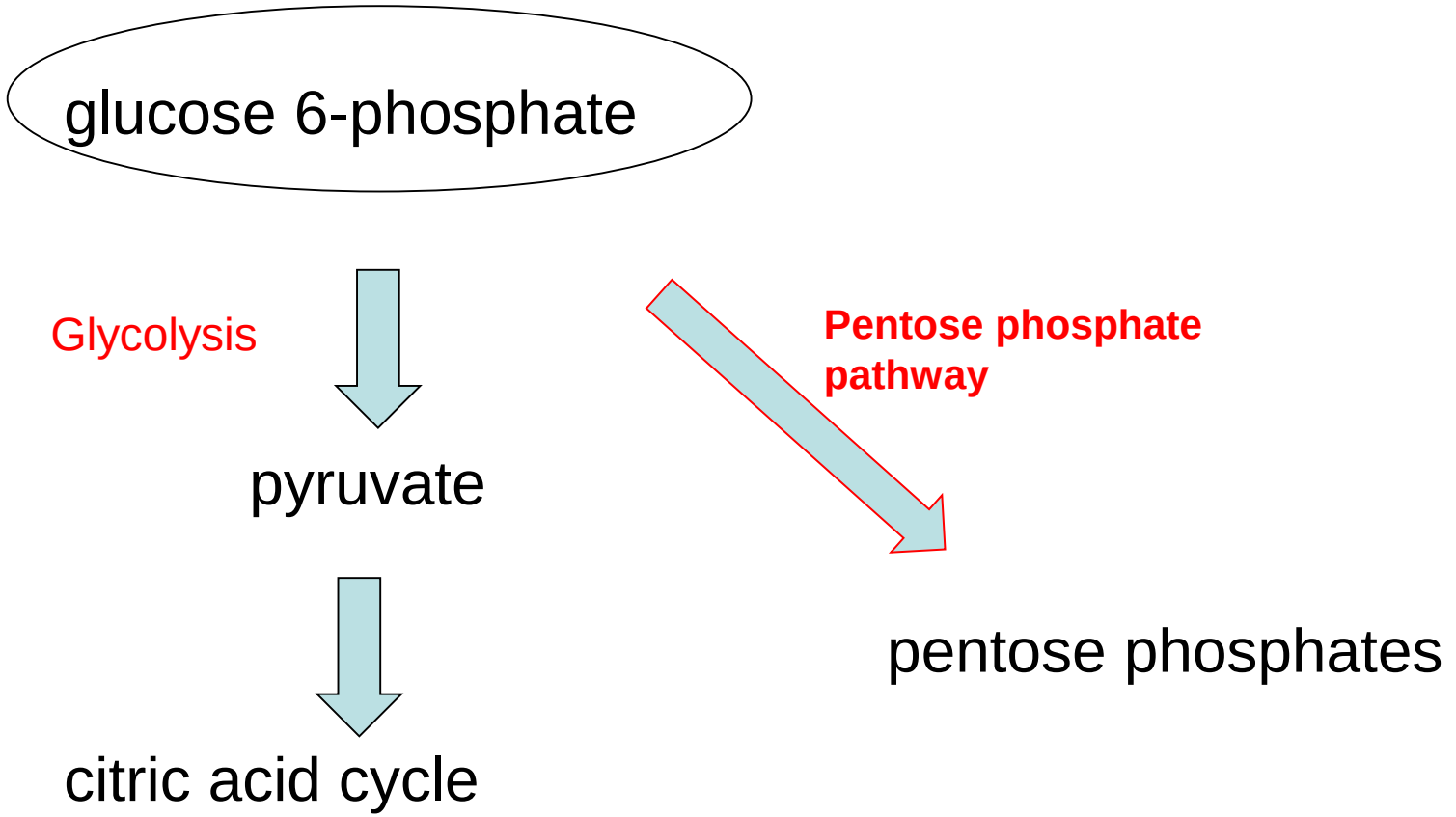
It is an electron-transporting coenzyme.

It is a coenzyme used in reductive biosynthesis
(e.g., fatty acid synthesis, cholesterol synthesis, nucleotide synthesis).

It is used to form reduced glutathione (a molecule with antioxidant functions).

Electrons are not transferred to oxygen
in the mitochondrial respiratory chain (unlike those of NADH).

The Pentose Phosphate Pathway



NADPH,

It’s necessary to reductive biosynthesis or to counter the damaging effects of oxygen radicals.

Erythrocytes and the cells of the lens and cornea that are directly exposed to oxygen to free radicals generated by oxygen.

Erythrocytes (NADPH prevent oxidative damage that a genetic defect in glucose 6-phosphate dehydrogenase, the first enzyme of the pathway, can have serious medical consequences

The pathway comprises two phases:

Oxidative phase:

Production of the coenzyme NADPH

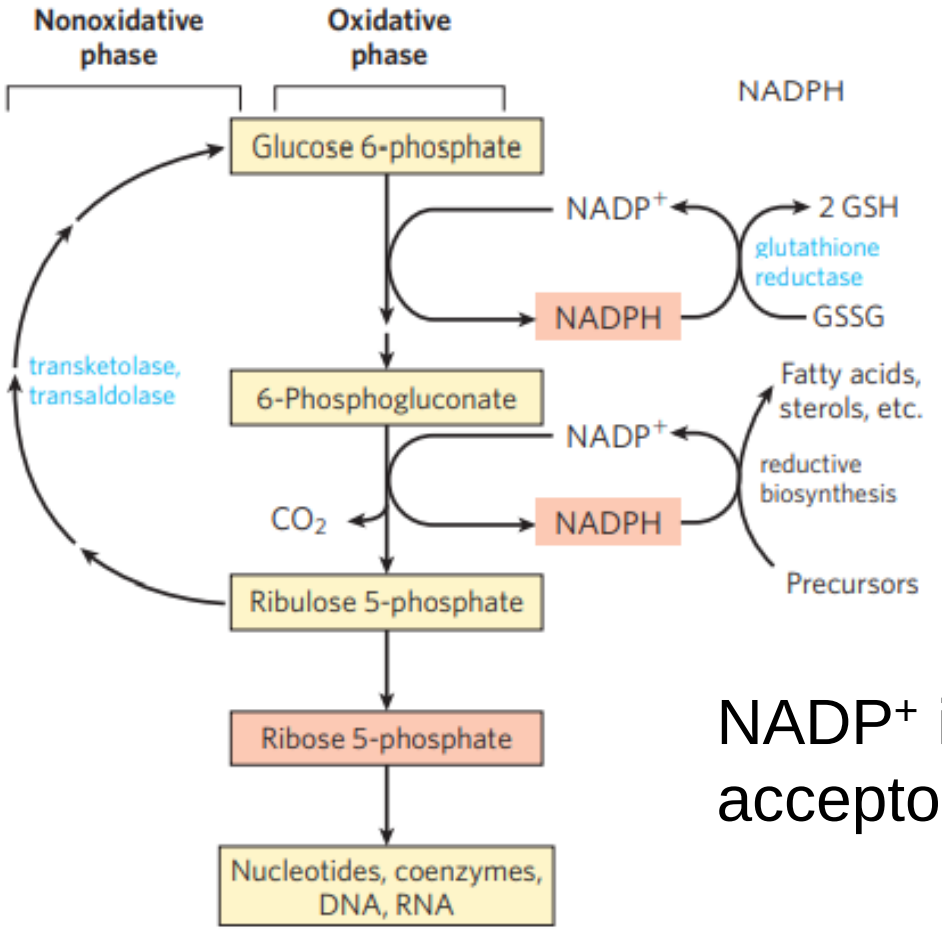
Nonoxidative phase:

Interconversion of 5C sugars into 6C and 3C sugars.

The Pentose Phosphate Pathway

There are two distinct phases in the pathway.

1) Oxidative phase
2) Non-oxidative phase



$NADP^+$ is the electron acceptor, yielding NADPH.

The Pentose Phosphate Pathway

There are two distinct phases in the pathway.

1) Oxidative phase

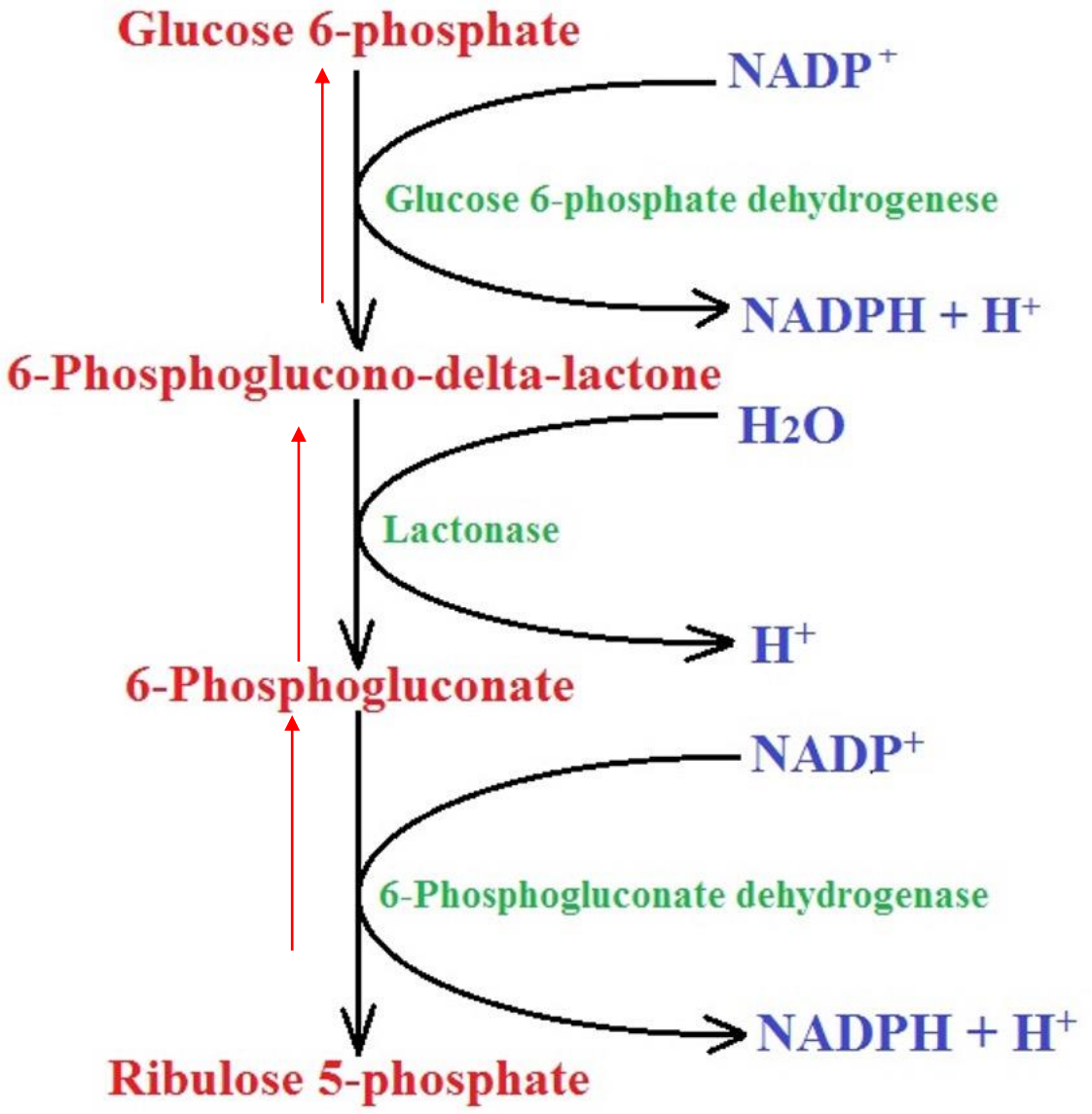
2) Non-oxidative phase

Pentose Phosphate Pathway

PHASE I (Oxidative Phase)

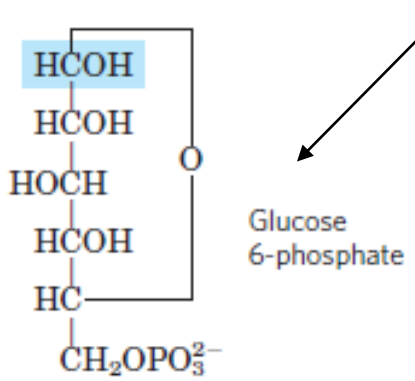
The first is the oxidative phase, in which NADPH is generated.

There are three reversible reactions



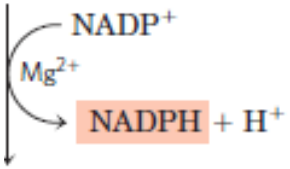
1) Oxidative phase

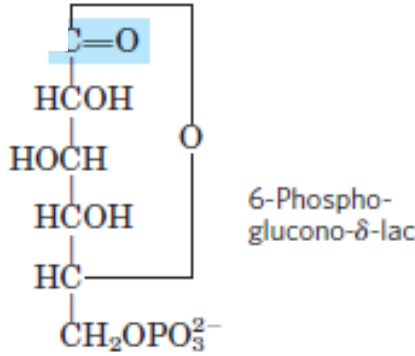
Derived from glycolysis or glicogenolysis



Glucose 6-phosphate

Glucose-6 phosphate dehydrogenase (G6PD)





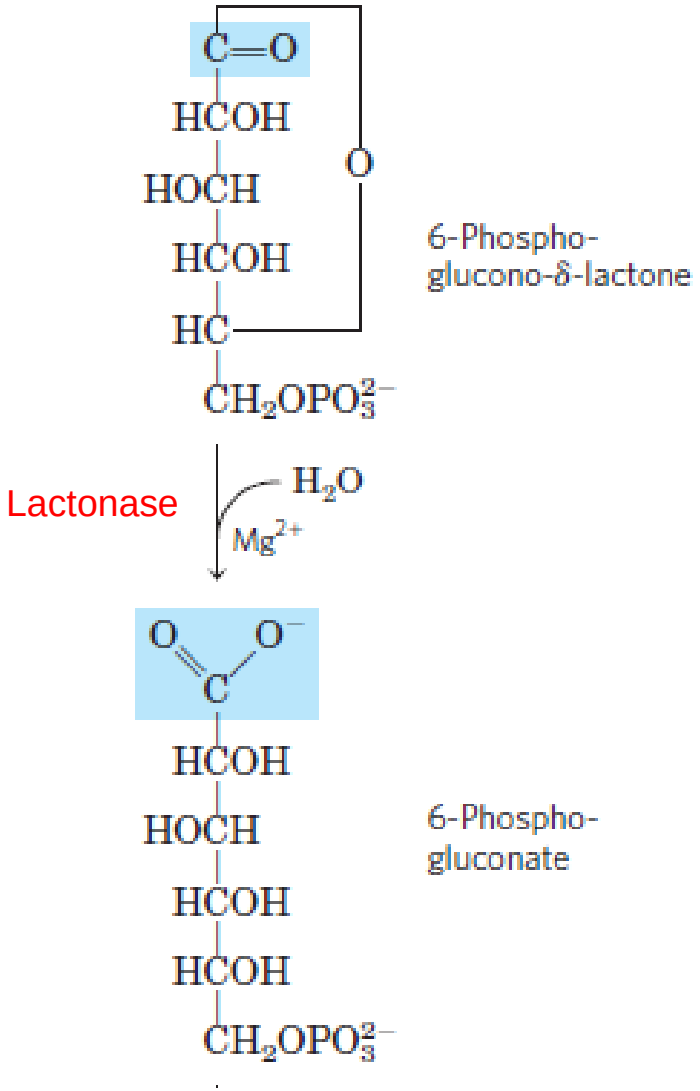
6-Phospho-glucono-δ-lactone (cyclic ester)

1) The first reaction is the oxidation of G6P by **glucose 6-phosphate dehydrogenase (G6PD)**

Transfers hydride ion from C1 of G6P to NADP+

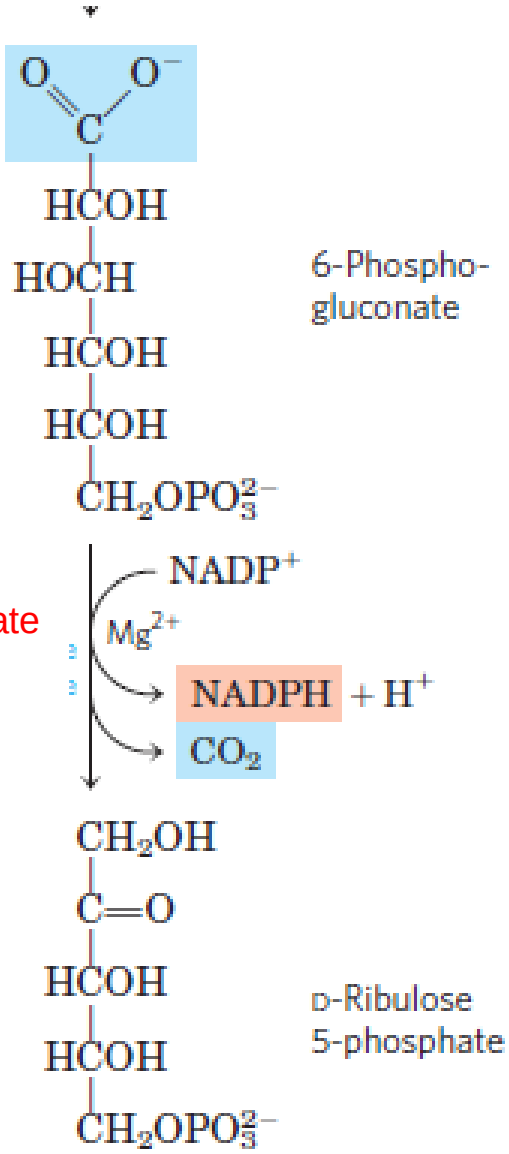
G6PD Generates 1st NADPH

2) Oxidative phase



The lactone is hydrolyzed to the free acid 6-phosphogluconate by a specific lactonase,

3) Oxidative phase



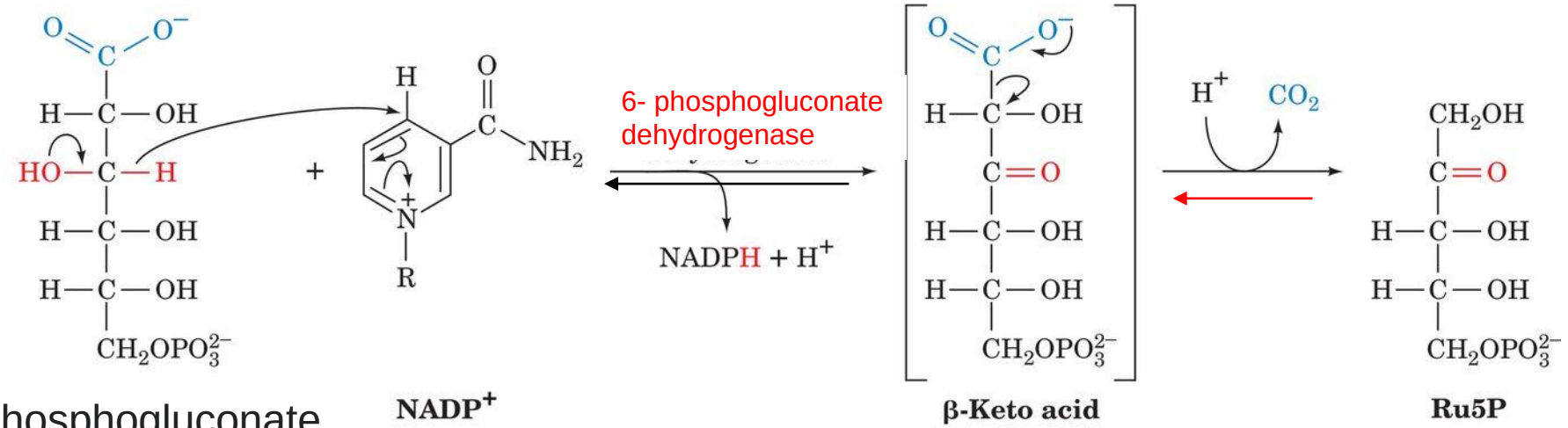
3) oxidation and decarboxylation
by **6-phosphogluconate dehydrogenase**

Generates 2nd NADPH

Oxidative decarboxylation

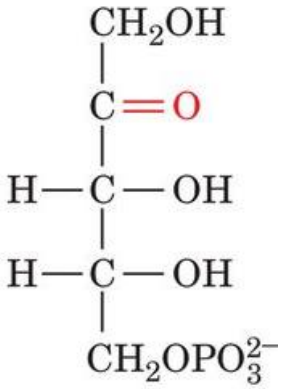
Ribulose 5-phosphate is important in glycolysis and gluconeogenesis regulation

oxidation and decarboxylation



6- phosphogluconate
(β-hydroxy acid)

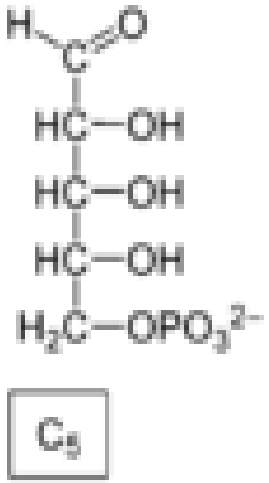
ribulose-5-phosphate



Ru5P

ribulose-5-phosphate

Ribulose 5-phosphate isomerase



ribose-5-phosphate

Phosphopentose isomerase converts

Ribulose 5-phosphate to its aldose isomer, ribose 5-phosphate.

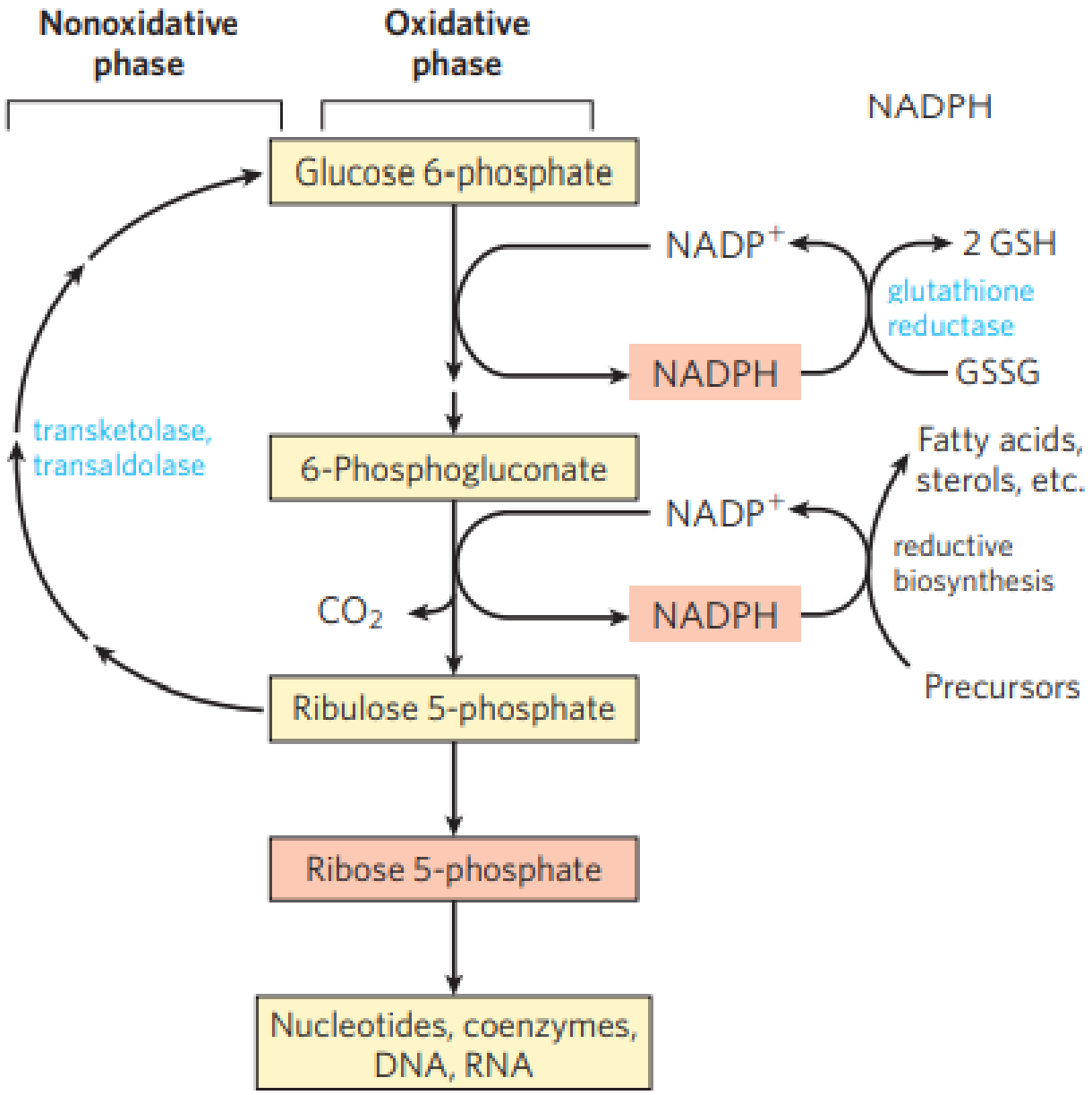
The Pentose Phosphate Pathway

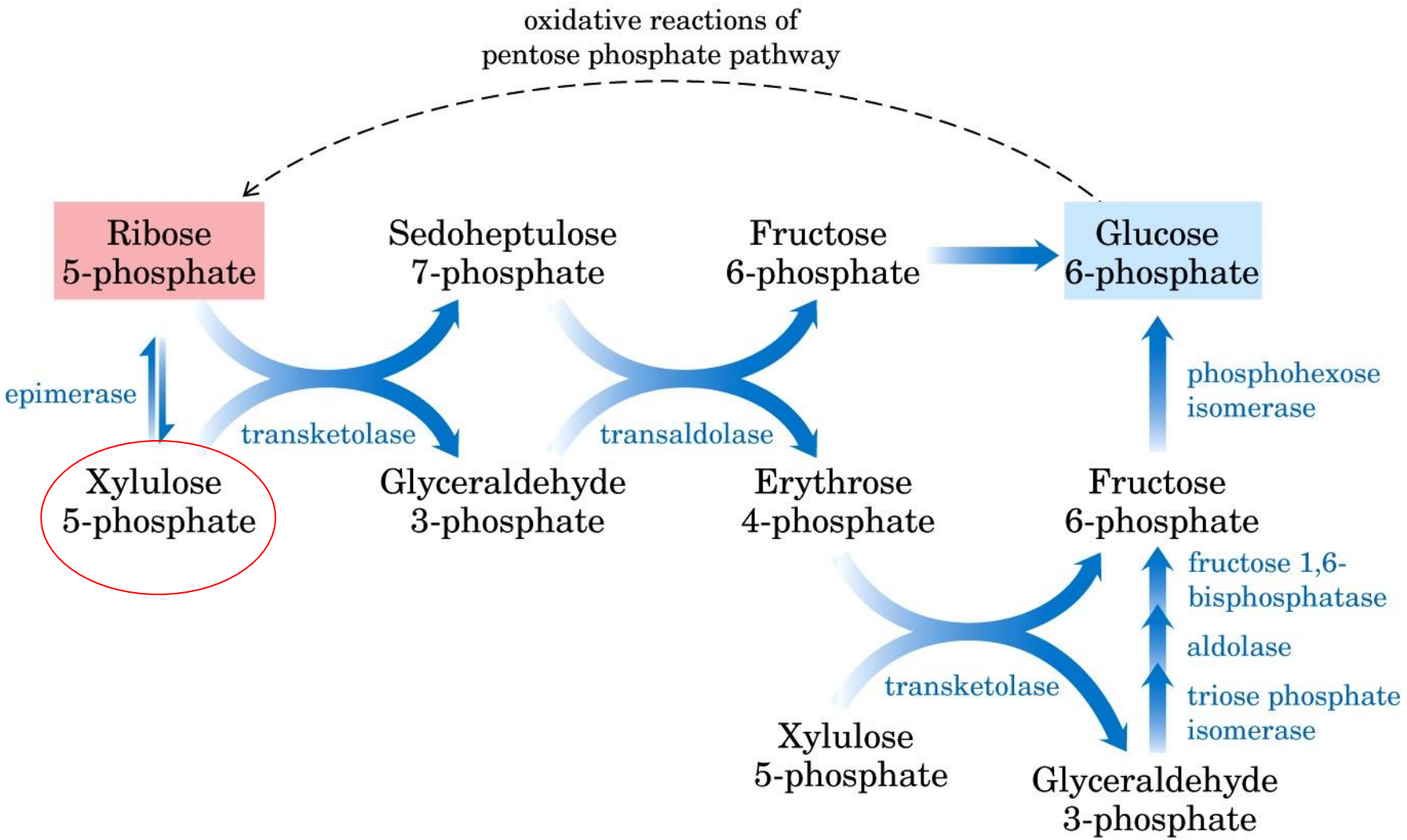
Glucose 6-phosphate + 2NADP+ + H_2O -> ribose 5-phosphate + CO_2 + 2NADPH + $2H^+$

The Nonoxidative Phase Recycles Pentose Phosphates to Glucose 6-Phosphate

Pentose phosphates produced in the oxidative phase

are recycled into glucose 6-phosphate.

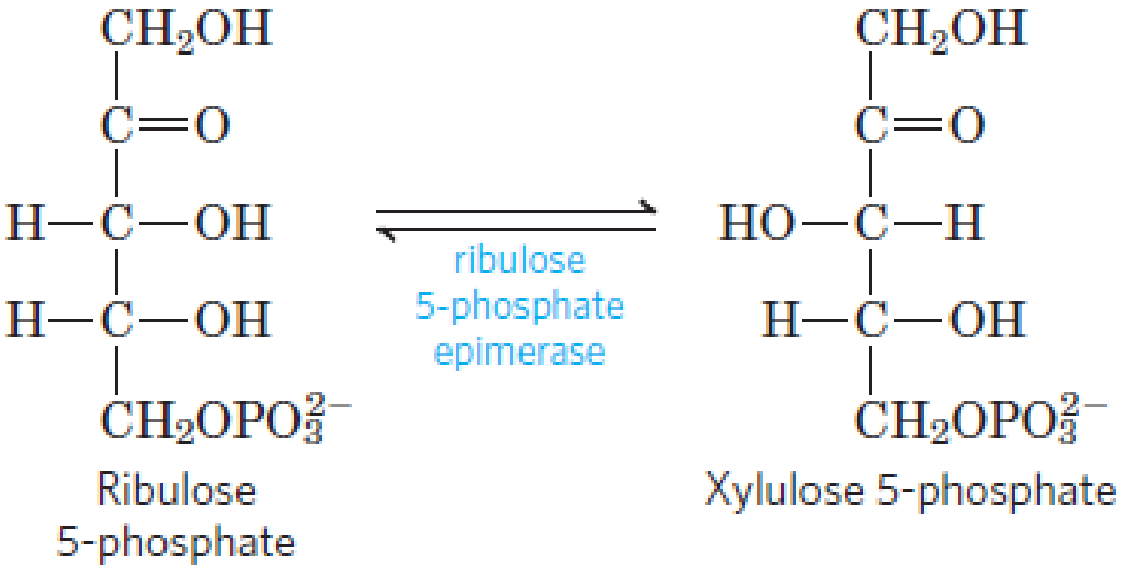

Nonoxidative phase
Oxidative phase
NADPH
Glucose 6-phosphate
NADP+
2 GSH
glutathione reductase
NADPH
GSSG
transketolase, transaldolase
6-Phosphogluconate
Fatty acids, sterols, etc.
NADP+
reductive biosynthesis
CO2
NADPH
Precursors
Ribulose 5-phosphate
Ribose 5-phosphate
Nucleotides, coenzymes, DNA, RNA


oxidative reactions of
pentose phosphate pathway
Ribose
5-phosphate
Sedoheptulose
7-phosphate
Fructose
6-phosphate
Glucose
6-phosphate
epimerase
transketolase
transaldolase
phosphohexose
isomerase
Xylulose
5-phosphate
Glyceraldehyde
3-phosphate
Erythrose
4-phosphate
Fructose
6-phosphate
fructose 1,6-
bisphosphatase
aldolase
triose phosphate
isomerase
transketolase
Xylulose
5-phosphate
Glyceraldehyde
3-phosphate

(a)

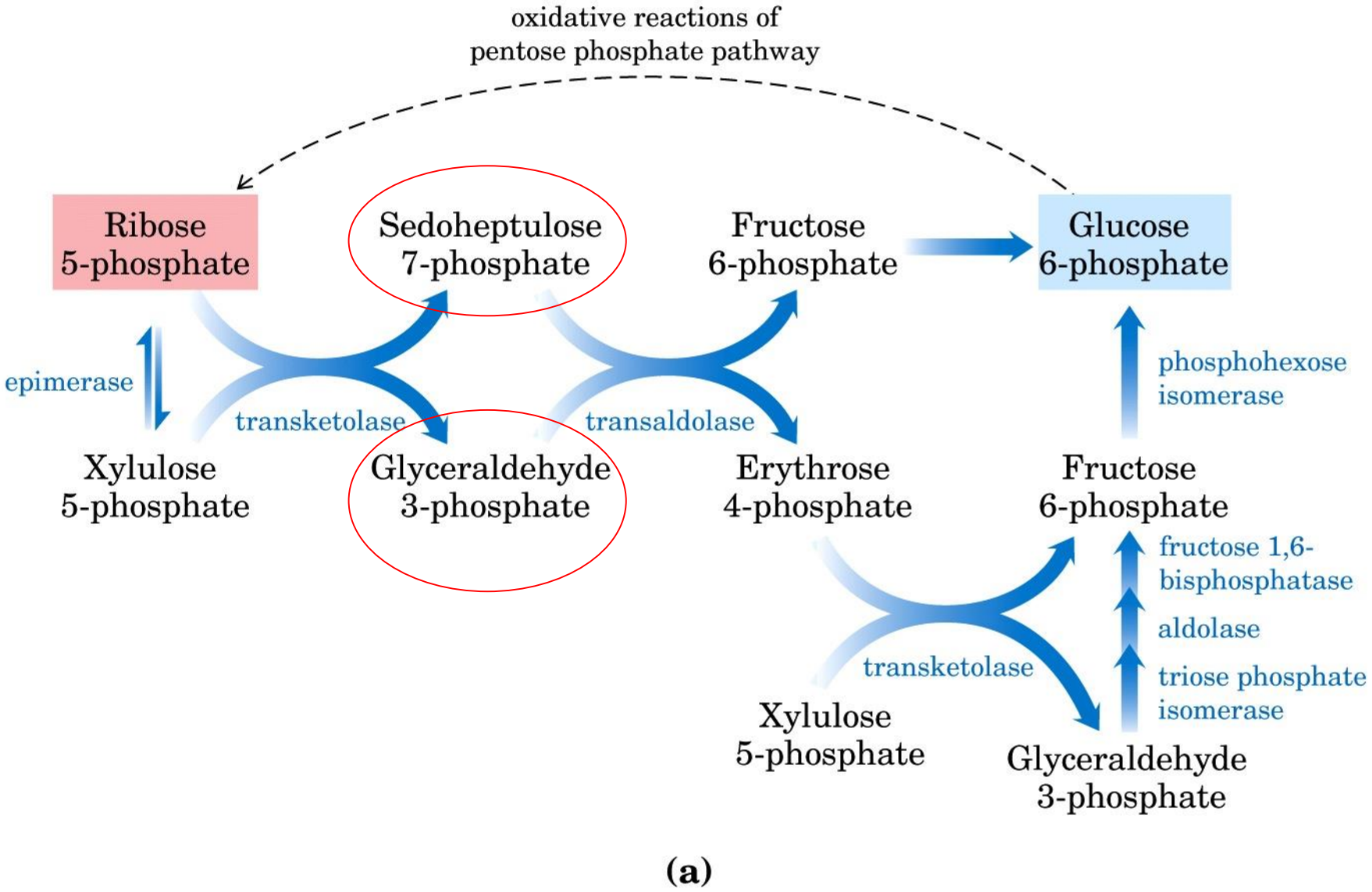
PHASE II: Non-oxidative phase

Ribulose 5-phosphate is first epimerized to **xylulose 5-phosphate:**



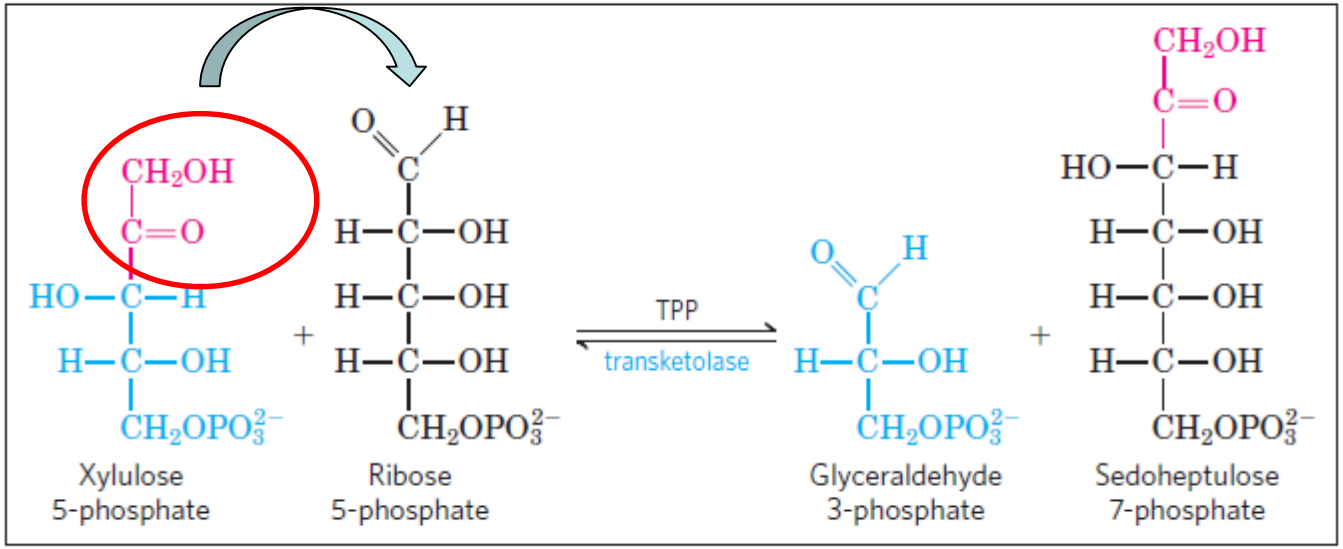
Transketolase transfers a two-carbon unit

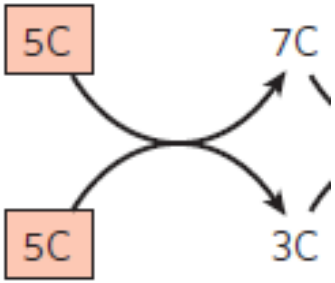
Transaldolase transfers a three-carbon unit



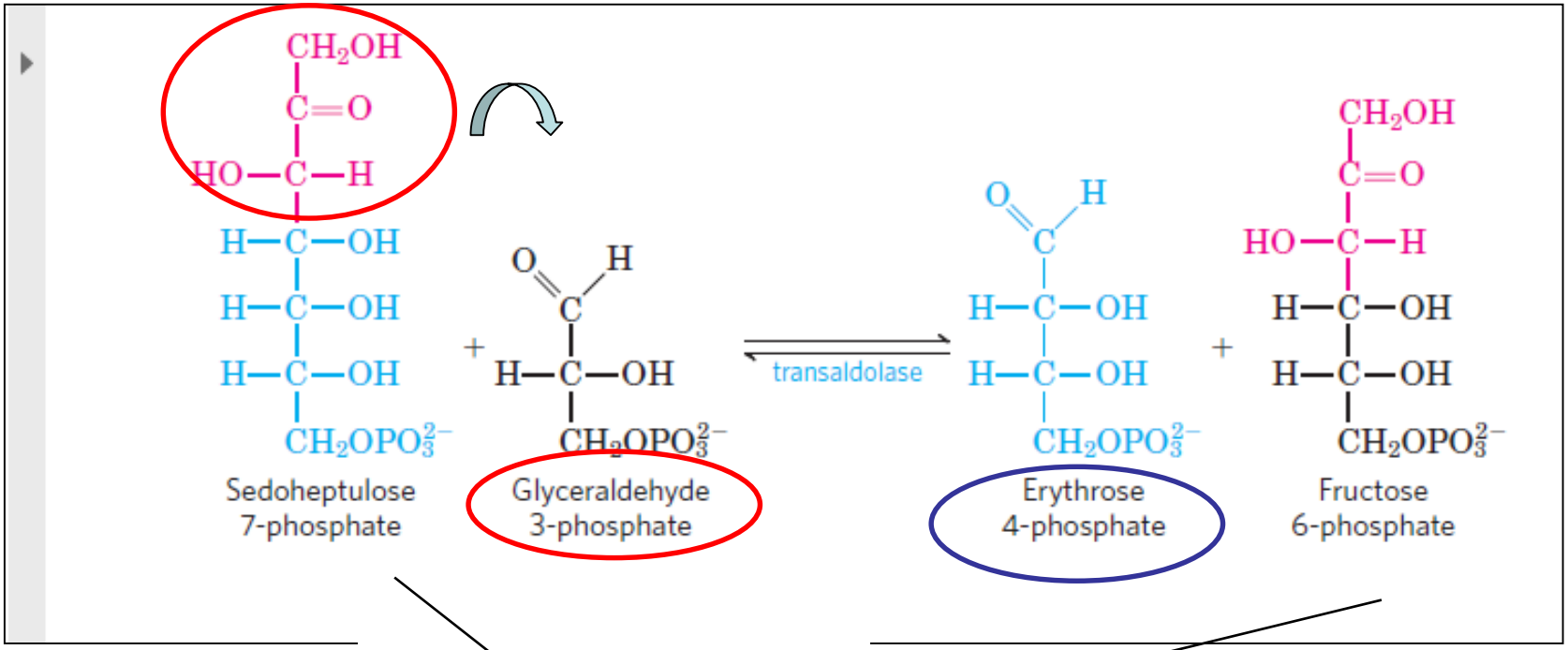
(a)

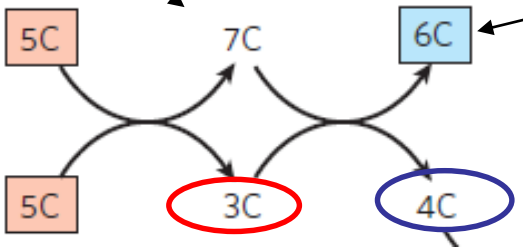
Conversion of two pentose phosphates to a triose phosphate and a seven-carbon sugar phosphate, sedoheptulose 7-phosphate.(Tranketolase)



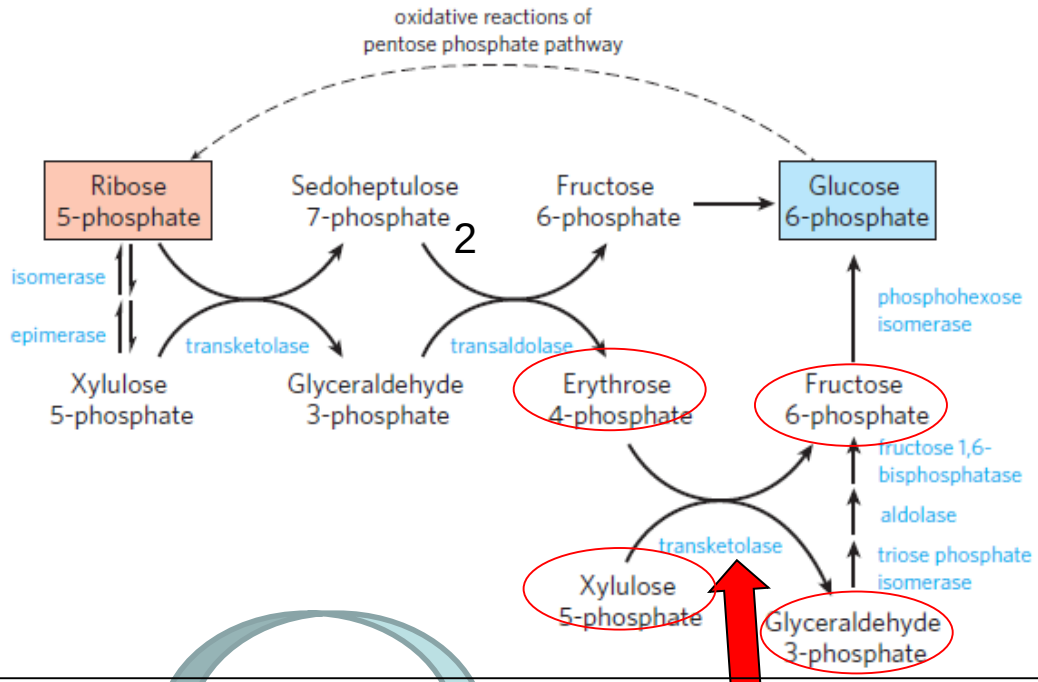


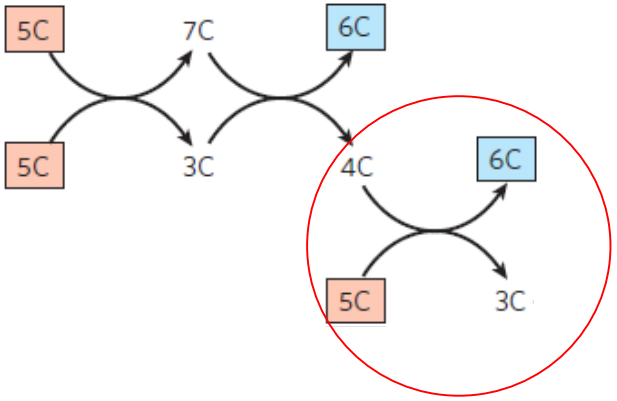
Transaldolase transfers 3 carbon atoms of sedoheptulose 7-phosphate on C1 of acceptor aldose (glyceraldehyde 3-phosphate)

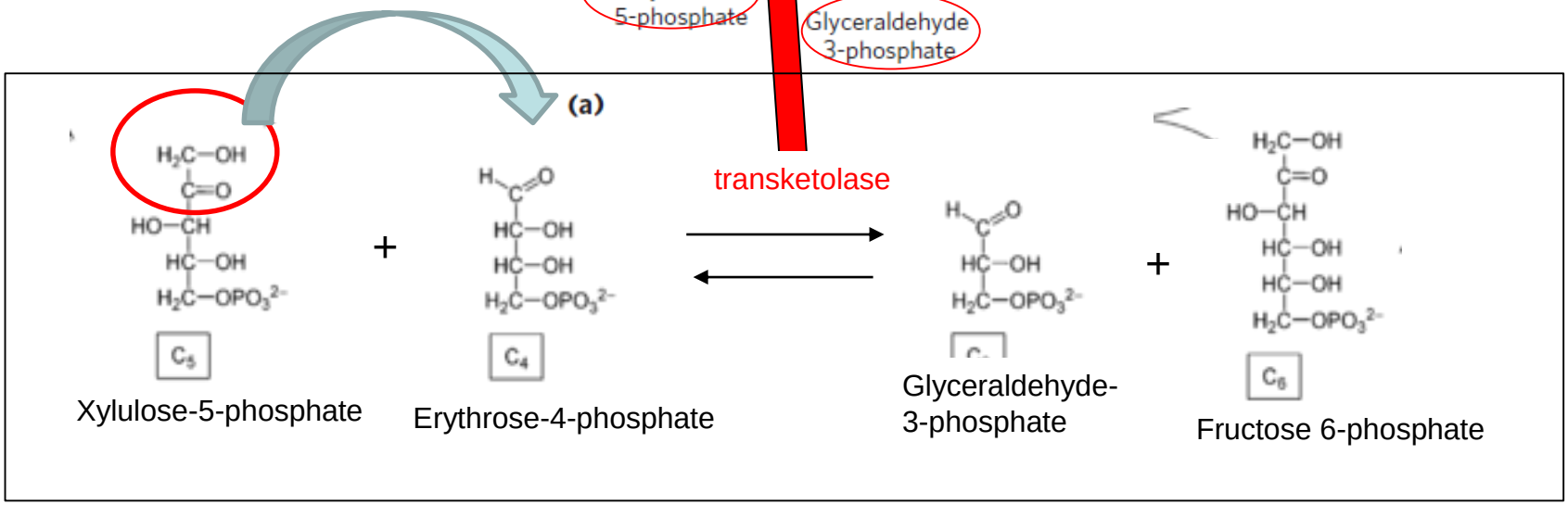




A series of rearrangements of the carbon skeletons







Xylulose-5-phosphate

Erythrose-4-phosphate

Glyceraldehyde-3-phosphate

Fructose 6-phosphate

Principal products of this way

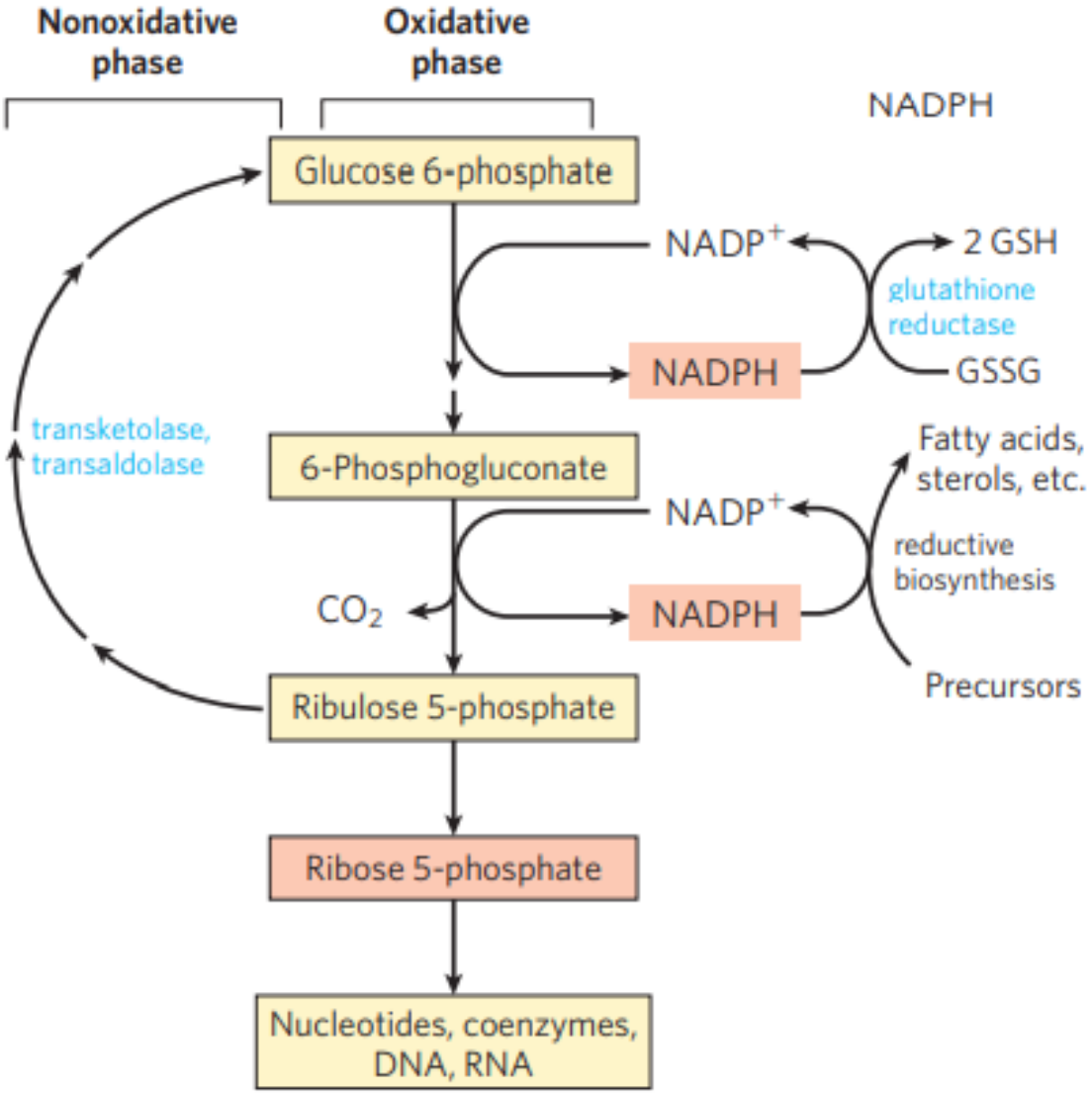
NADPH and Ribose-5-P

Reducing equivalents

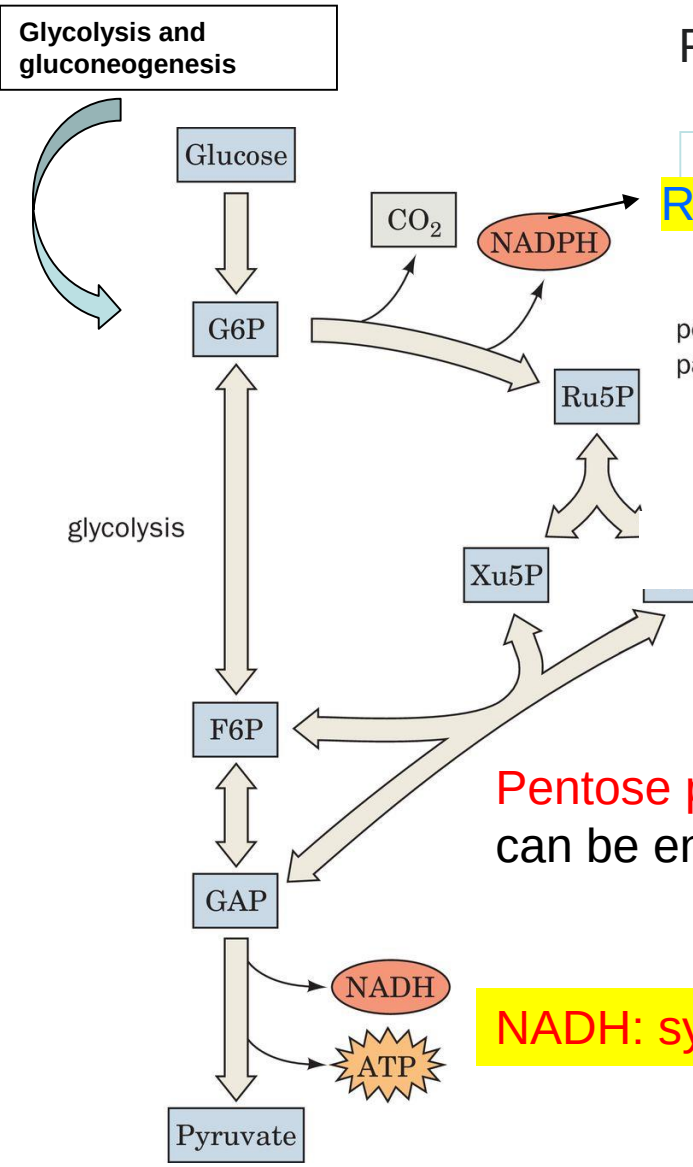
•Nucleotid and nucleic acid

•Produces erythrose 4-phosphate
•Precursor of aminoacid and vitamin B6 synthesis .

Transketolases and Transaldolases are activated to produce metabolic intermediates of glycolysis.


Nonoxidative phase
Oxidative phase
NADPH
Glucose 6-phosphate
NADP+
2 GSH
glutathione reductase
NADPH
GSSG
transketolase, transaldolase
6-Phosphogluconate
Fatty acids, sterols, etc.
NADP+
reductive biosynthesis
CO2
NADPH
Precursors
Ribulose 5-phosphate
Ribose 5-phosphate
Nucleotides, coenzymes, DNA, RNA

Relationship Between Glycolysis & Pentose Phosphate Pathway



Principal function:

Reducing power used for reductive biosynthesis

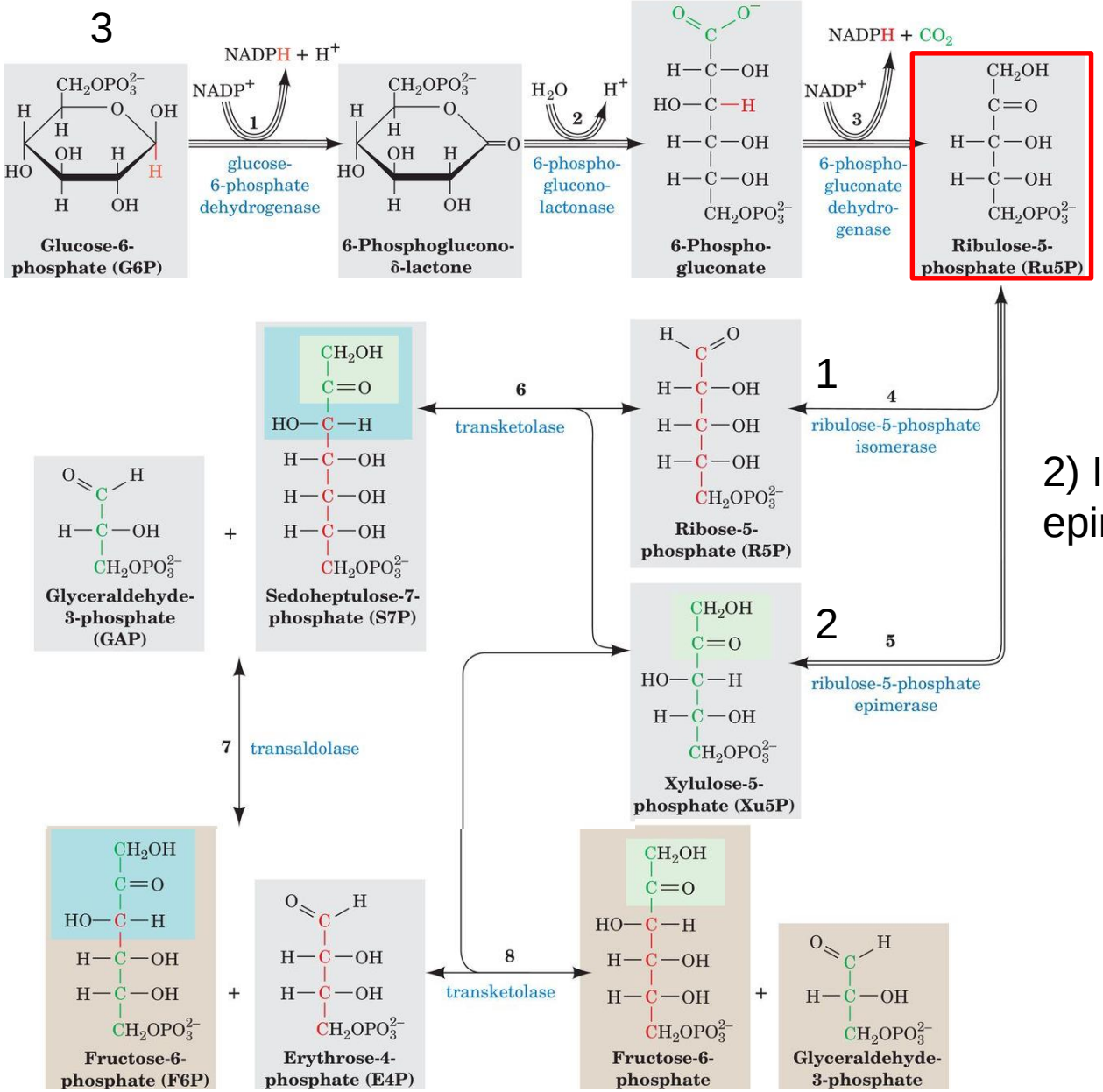
ribose-5 phosphate (R5P)

Nucleotid synthesis in cells

Pentose phosphate pathway produts can be entered to glycolysis

NADH: synthesis ATP

Pentose Phosphate Pathway

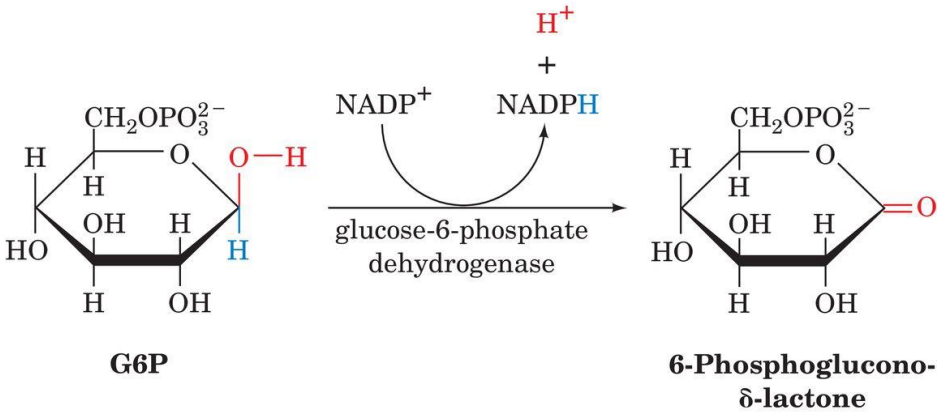


1) Oxidative phase

2) Isomerization and epimerization

Glucose 6-phosphate dehydrogenase (G6PD) deficiency

Glucose 6-phosphate dehydrogenase catalyzes the first step in the pentose phosphate pathway which produces NADPH.

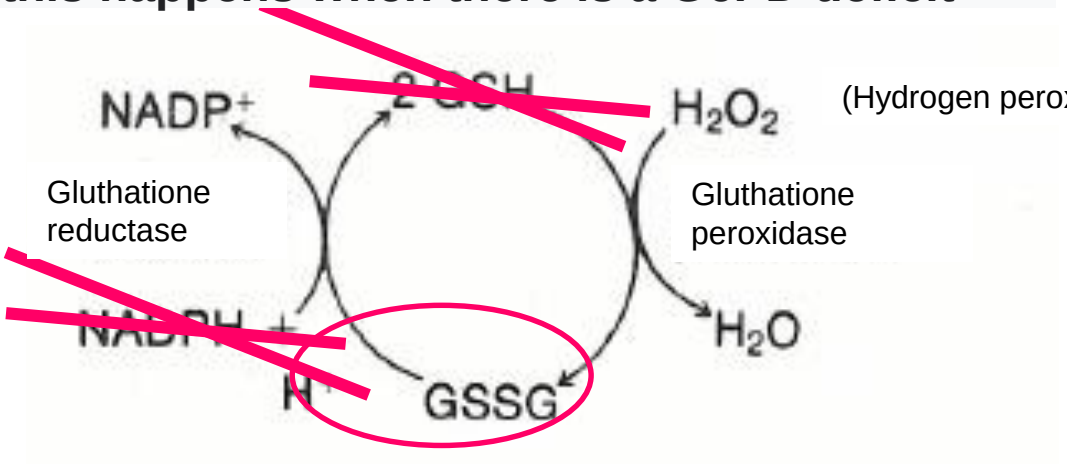


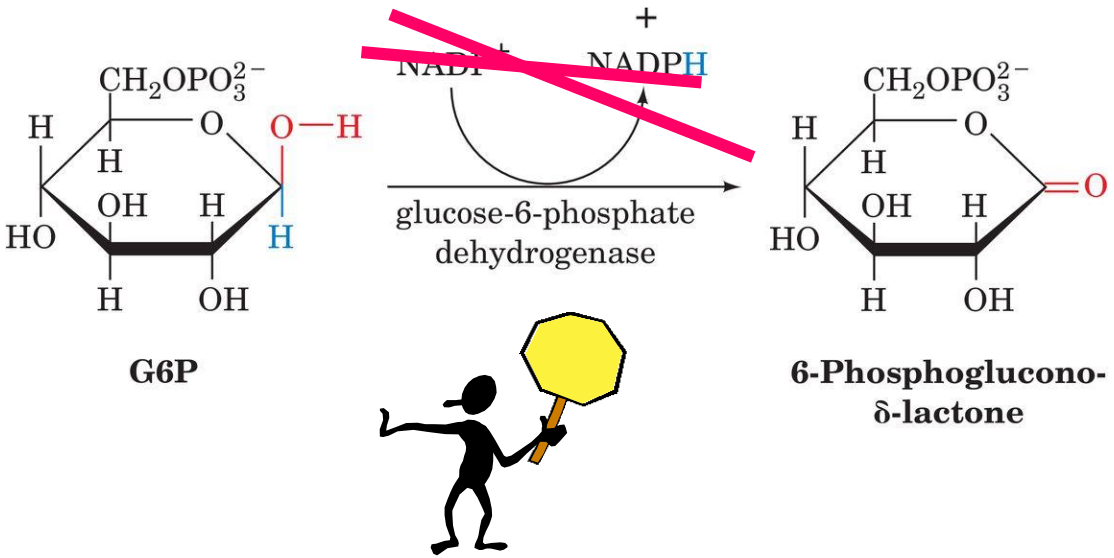
G6PD: Essential in many biosynthetic pathways, also protects cells from oxidative damage by hydrogen peroxide (H_2O_2) and superoxide free radicals,

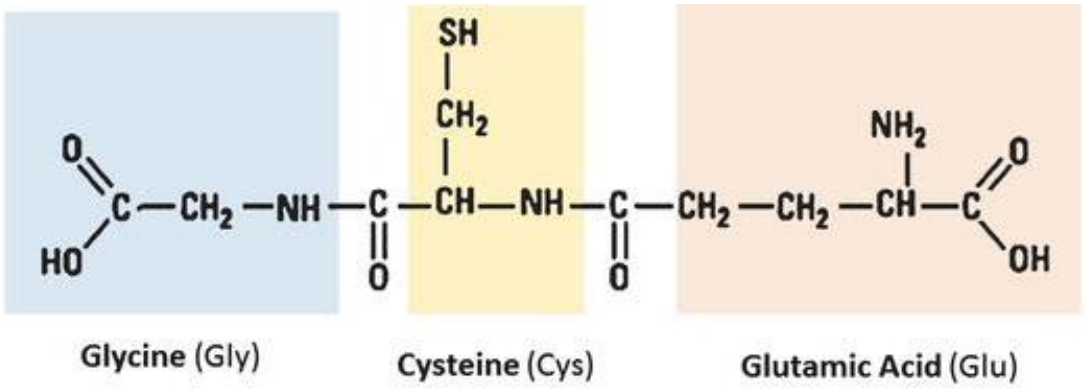
Highly reactive oxidants generated as metabolic byproducts and through the actions of drugs such as primaquine and natural products such as divicine (the toxic ingredient of fava beans)

NADPH is important to obtain GSH (glutathione)

GSH is not produced when pentoso phosphate pathway is broken and this happens when there is a G6PD deficit

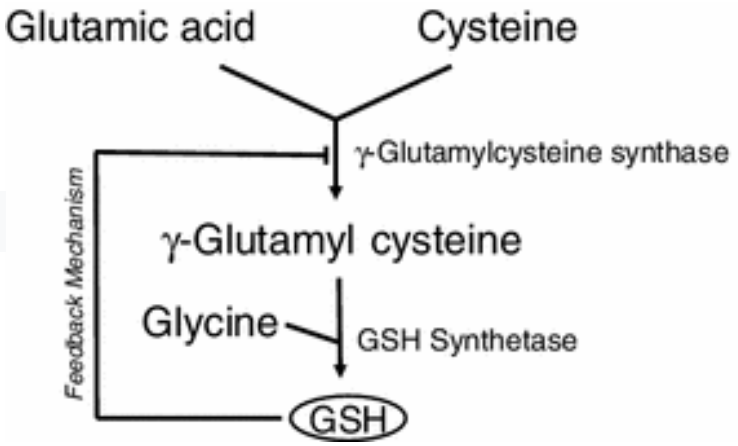






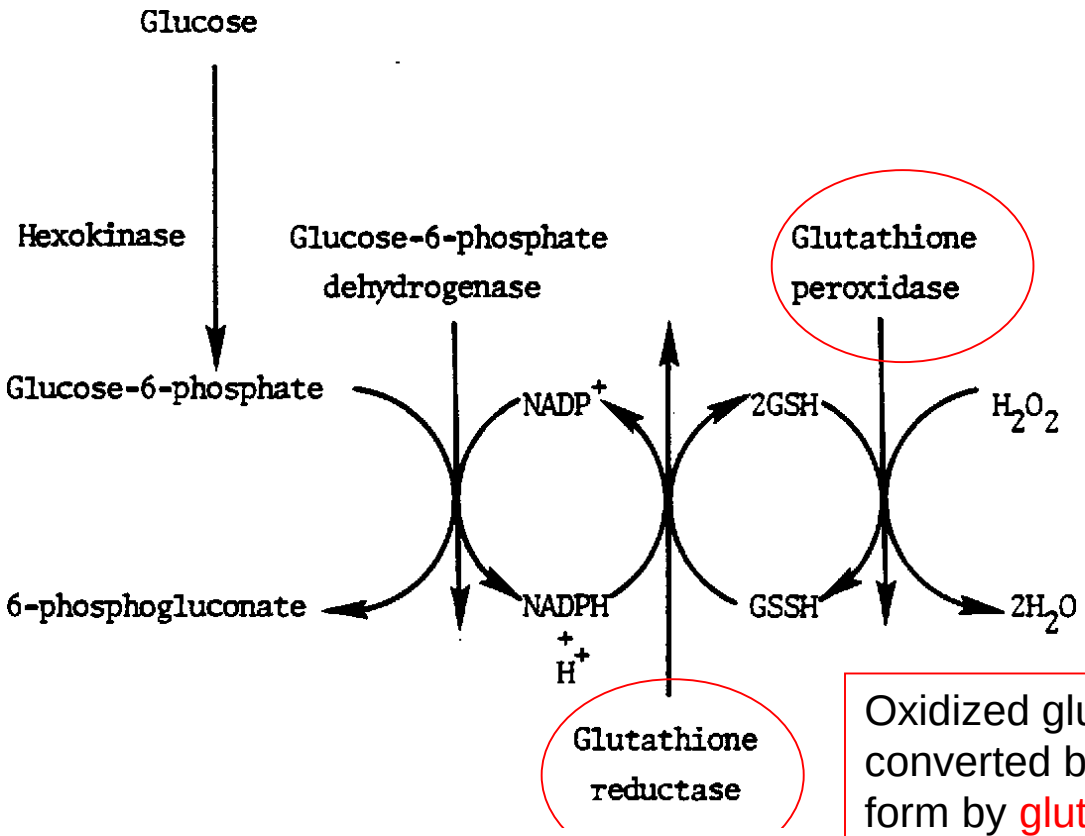
Glutathione (GSH) is a tripeptide that performs various functions, such as antioxidant, reducing the -SH groups of proteins, reducing the ferrous ion of Hb and cofactor of some enzymes

It is synthesized from glutamate



Glucose 6-phosphate dehydrogenase (G6PD) deficiency

In G6PD-deficient individuals, the NADPH production is diminished and detoxification of H_2O_2 is inhibited.



During normal detoxification, H_2O_2 is converted to H_2O in a reaction which glutathione is oxidated by glutathione peroxidase

Oxidized glutathione is converted back to the reduced form by glutathione reductase and NADPH

ROS (Reactive oxygen species)

Oxygen	Superoxide anion	Peroxide
O_2	$\cdot O_2^-$	$\cdot O_2^{-2}$
Hydrogen Peroxide	Hydroxyl radical	Hydroxyl ion
H_2O_2	$\cdot OH$	OH^-

→ Oxidizing molecules

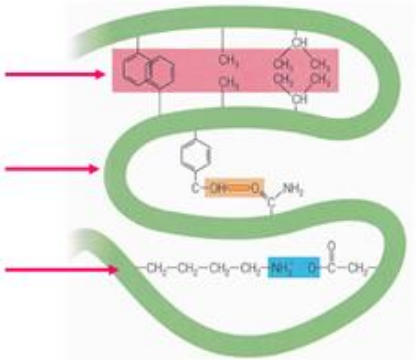
The absence of intra erythrocyte glutathione causes damage to the erythrocytes

①

②

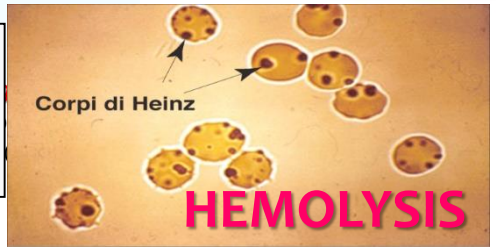
③

② Destruction of the tertiary structure of the Hb



Damage to cellular membrane

Oxidation of heme iron with the formation of methemoglobin



Cellular damage results: lipid peroxidation leading to breakdown of erythrocyte membranes and oxidation of proteins and DNA

It is therefore vital for the integrity of the erythrocytes to have a continuous supply of NADPH + H +.

Glucose 6-phosphate dehydrogenase (G6PD) deficiency

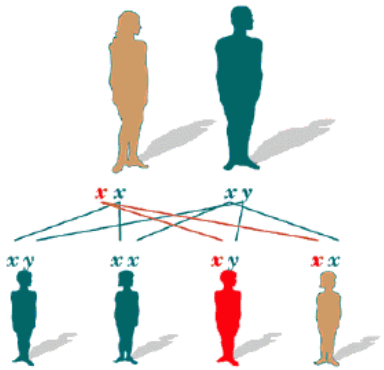
G6PD is expressed in all tissues, but its deficiency occurs essentially in erythrocytes

MUTATION IN THE GENE G6PD

400 variants

- ☞Enzyme is not expressed
- ☞the enzyme is expressed but is labile (half-life of a few days)

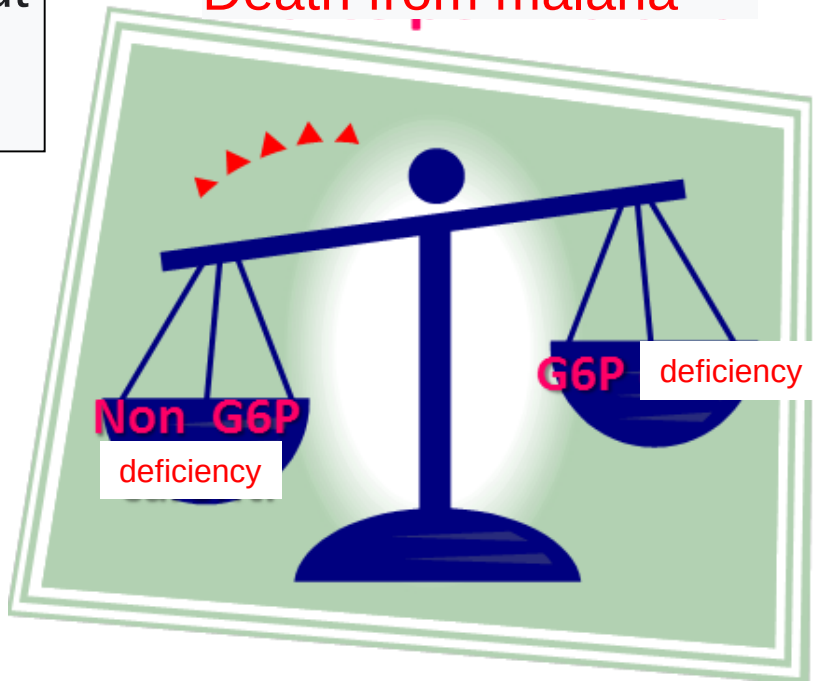
GENE is located on the X chromosome



Glucose 6-phosphate dehydrogenase (G6PD) deficiency

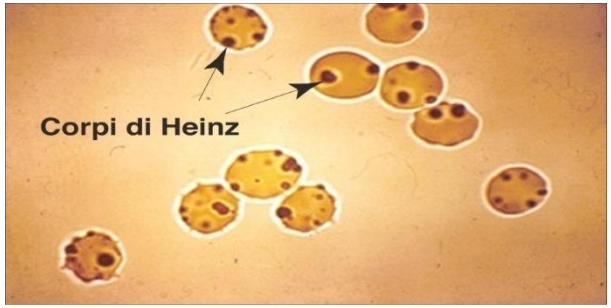
The maximum incidence (about 30%) is observable in areas where malaria was endemic

Death from malaria



Oxidative stress, induced by a condition of G6PD deficiency, has created an unfavorable environment at the erythrocyte level for the establishment of the protozoan *Plasmodium falciparum*

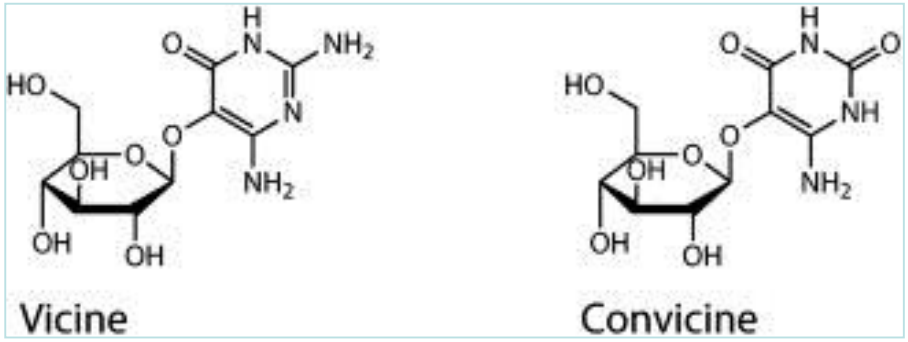
Factors triggering hemolysis in G6PD deficient subjects



Ingestion of fava beans

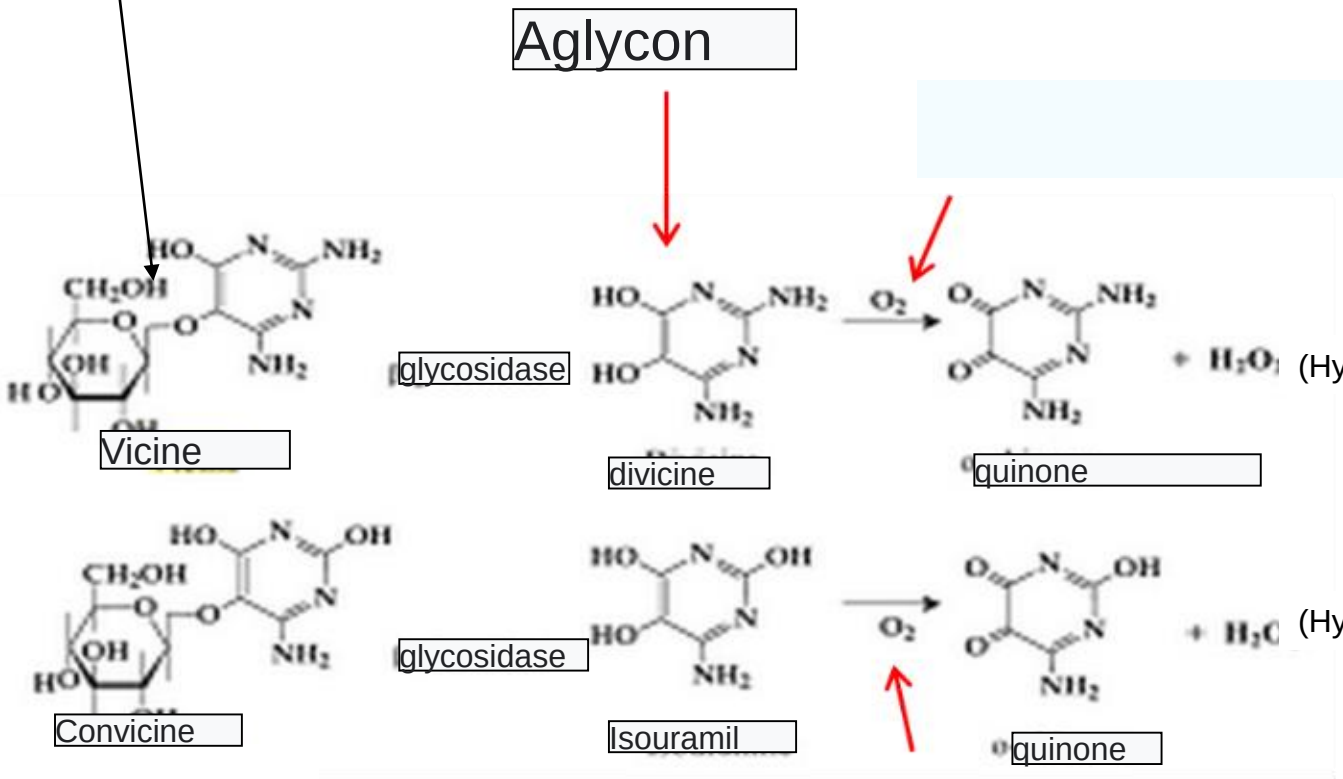
They contain vicine and convicine, able to promote oxidative stress

Vicine and Convicine are pyrimidine glycosides.



Vicine and Convicine

By hydrolysis of the β-glucosidic bond between glucose and the hydroxyl group at C-5 on the pyrimidine ring, generate the aglycones: divicine (2,6-diamino-4,5-dihydroxypyrimidine) and isouramil (6-amino-2,4,5-trihydroxypyrimidine), respectively.



SCIENTIFIC REPORTS

OPEN

Degradation of vicine, convicine and their aglycones during fermentation of faba bean flour

Carlo Giuseppe Rizzello[1], Ilario Losito[2,3], Laura Facchini[2], Kati Katina[4], Francesco Palmisano[2,3], Marco Gobbetti[1] & Rossana Coda[4]

Received: 07 March 2016
Accepted: 08 August 2016
Published: 31 August 2016

Factors triggering hemolysis in G6PD deficient subjects

Some drugs can promote oxidative stress

DRUGS TO AVOID IN G6PD DEFICIENCY

DEFINITE RISK OF HAEMOLYSIS		POSSIBLE RISK OF HAEMOLYSIS	
Pharmacological Class	**Drugs***	**Pharmacological Class**	**Drugs***
Anthelmintics	• ß-Naphthol • Niridazole • Stibophen	**Analgesics**	• Acetylsalicylic acid (Aspirin) • Acetanilide • Paracetamol (Acetaminophen) • Aminophenazone (Aminopyrine) • Dipyrone (Metamizole) • Phenacetin • Phenazone (Antipyrine) • Phenylbutazone • Tiaprofenic acid
Antibiotics	• Nitrofurans - Nitrofurantoin - Nitrofurazone • Quinolones - Ciprofloxacin - Moxifloxacin - Nalidixic acid - Norfloxacin - Ofloxacin • Chloramphenicol • Sulfonamides - Co-trimoxazole (Sulfamethoxazole + Trimethoprim) - Sulfacetamide - Sulfadiazine - Sulfadimidine - Sulfamethoxazole - Sulfanilamide - Sulfapyridine - Sulfasalazine (Salazosulfapyridine) - Sulfisoxazole (Sulfafurazole)	**Antibiotics**	• Furazolidone • Streptomycin • Sulfonamides - Sulfacytine - Sulfaguanidine - Sulfamerazine - Sulfamethoxypyridazole
		Anticonvulsants	• Phenytoin
		Antidiabetics	• Glibenclamide
		Antidotes	• Dimercaprol (BAL)
		Antihistamines	• Antazoline (Antistine) • Diphenhydramine • Tripelennamine
Antimalarials	• Mepacrine • Pamaquine • Pentaquine • Primaquine	**Antihypertensives**	• Hydralazine • Methyldopa
Antimethemo-globinaemic Agents	• Methylene blue	**Antimalarials**	• Chloroquine & derivatives • Proguanil • Pyrimethamine • Quinidine • Quinine
Antimycobacterials	• Dapsone • Para-aminosalicylic acid • Sulfones - Aldesulfone sodium (Sulfoxone) - Glucosulfone - Thiazosulfone	**Antimycobacterials**	• Isoniazid
		Antiparkinsonism Agents	• Trihexyphenidyl (Benzhexol)
Antineoplastic Adjuncts	• Doxorubicin • Rasburicase	**Cardiovascular Drugs**	• Dopamine (L-dopa) • Procainamide • Quinidine
Genitourinary Analgesics	• Phenazopyridine (Pyridium)	**Diagnostic Agent for Cancer Detection**	• Toluidine blue
Others	• Acetylphenylhydrazine • Phenylhydrazine	**Gout Preparations**	• Colchicine • Probenecid
		Hormonal Contraceptives	• Mestranol
		Nitrates	• Isobutyl nitrite
		Vitamin K Substance	• Menadiol Na sulfate • Menadione • Menadione Na bisulfite • Phytomenadione

Factors triggering hemolysis in G6PD deficient subjects

Some drugs can promote oxidative stress

Controindicato

- Acido nalidissico
- Nitrofurantoina
- Metamizolo sodico
- Rasburicase
- Sulfadiazina (via orale)
- Sulfametossazolo (via orale e parenterale)
- Sulfasalazina
- Trimetoprim (via orale e parenterale)

Sconsigliato (eccetto particolari situazioni) a causa dell'osservazione di casi di emolisi acuta

- Ciprofloxacina (via orale e parenterale)
- Clorochina
- Dimercaprolo
- Fitomenadione (vitamina K_1)
- Glibenclamide
- Levofloxacina (via orale e parenterale)
- Norfloxacina (via orale)
- Spiramicina (via orale e parenterale)
- Sulfadiazina (via orale)

Sconsigliato (eccetto particolari situazioni) a causa dell'appartenenza ad una classe farmacologica a rischio, o a causa di un rischio potenziale di emolisi

- Acido pipemidico
- Chinina
- Enoxacina
- Fenazone (via auricolare)
- Gliclazide
- Glimepiride
- Glipizide
- Idrossiclorochina
- Lomefloxacina
- Moxifloxacina
- Ofloxacina (via orale e parenterale)
- Pefloxacina (via orale e parenterale)
- Prilocaina
- Sulfacetamide

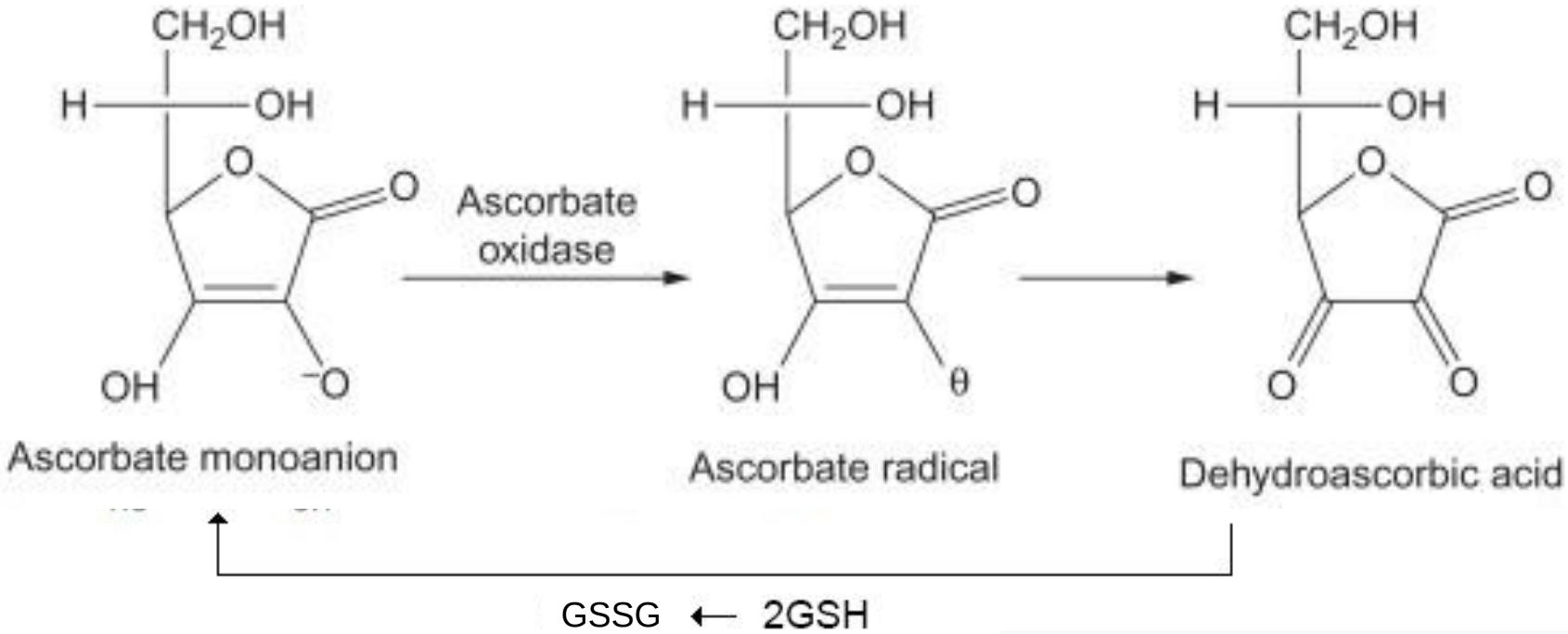
Sconsigliato a dosi elevate

- Acido acetilsalicilico
- Acido ascorbico
- Paracetamolo

Possibile utilizzo dopo l'analisi dei dati disponibili (letteratura e farmacovigilanza)

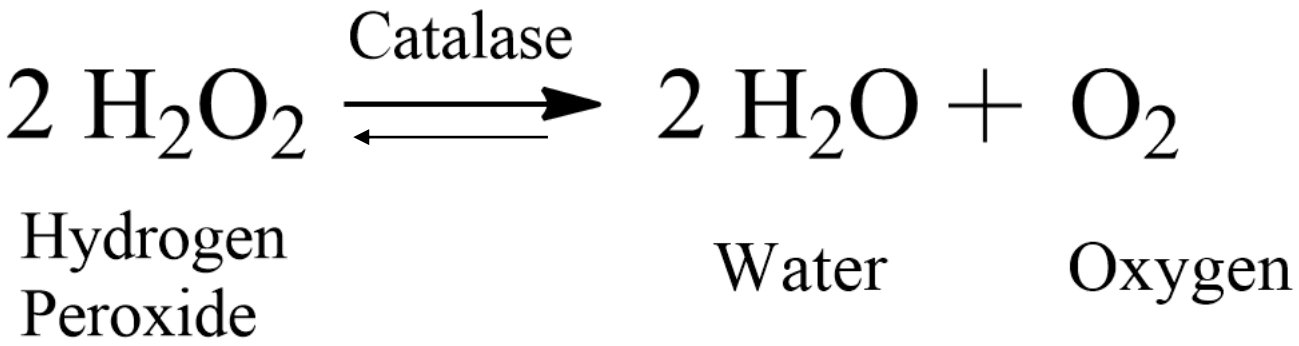
- Blu di metilene (via topica)*
- Bupivacaina*
- Chinidina*
- Ciprofloxacina (via oftalmica e auricolare)*
- Cloramfenicolo (via oftalmica)*
- Colchicina*
- Dietilamina*
- Diidrochinidina*
- Dimenidrinato*
- Doxorubicina*
- Fenazone (via auricolare)*
- Fenilbutazone*
- Fenitoina*
- Glicole propilenico*
- Isoniazide (via orale e parenterale)*
- Levodopa*
- Meflochina*
- Monossido di azoto*
- Nitroprussiato*
- Norfloxacina (via oftalmica)*
- Ofloxacina (via oftalmica e auricolare)*
- Pirimetamina*
- Proguanile*
- Streptomicina*
- Tiamfenicolo*
- Triesilfenidile*

Ascorbic acid → not recommended in high doses



Ascorbic acid is oxidized to dehydroascorbic acid (DHA) and converted back to ascorbic acid at the expence of glutathione.

H_2O_2 is also broken down to H_2O and O_2 by catalase, which also requires NADPH.



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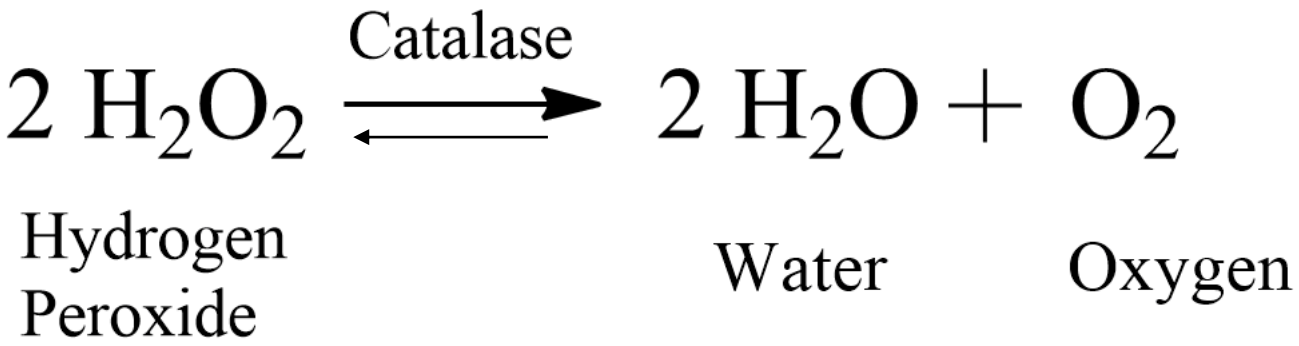


Wernicke-Korsakoff Syndrome Is Exacerbated by a Defect in Transketolase

Transketolase requires thiaminepyrophosphate (TPP) as coenzyme

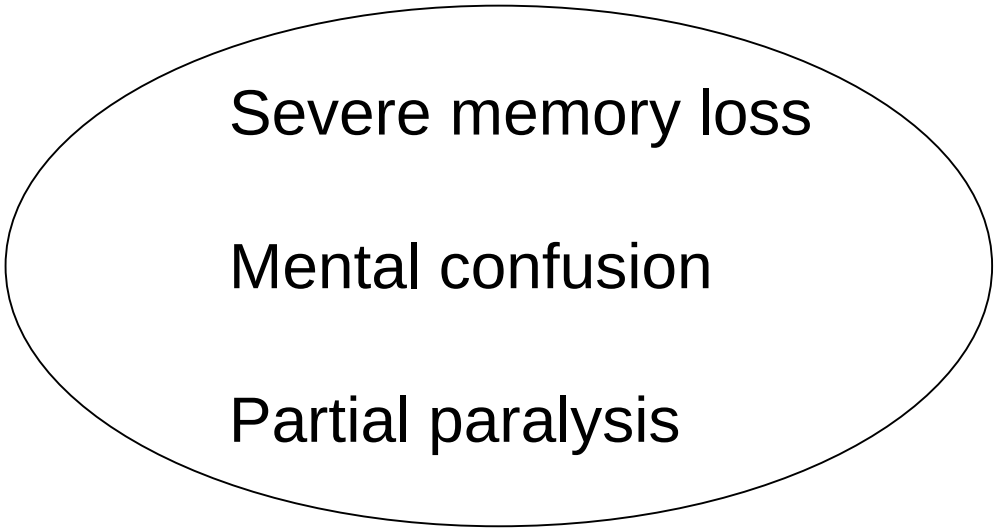
Wernicke-Korsakoff syndrome is a disorder caused by a severe deficiency of thiamine, a component of TPP.

The syndrome is more common among people with alcoholism than in the general population, because chronic, heavy alcohol consumption interferes with the intestinal absorption of thiamine.

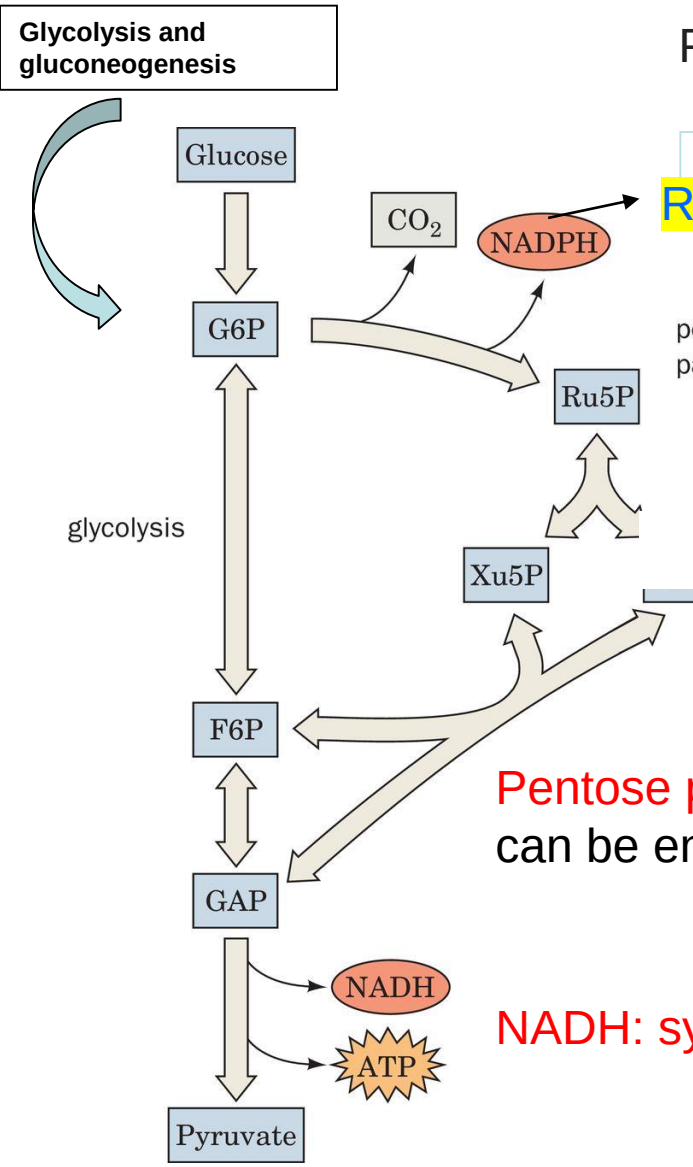
The syndrome can be exacerbated by a mutation in the gene for transketolase that results in an enzyme with a lowered affinity for TPP an affinity one-tenth that of the normal enzyme.

The result is a slowing down of the whole pentose phosphate pathway.

Wernicke-Korsakoff syndrome symptoms:



Relationship Between Glycolysis & Pentose Phosphate Pathway



Principal function:

Reducing power used for reductive biosynthesis

ribose-5 phosphate (R5P)

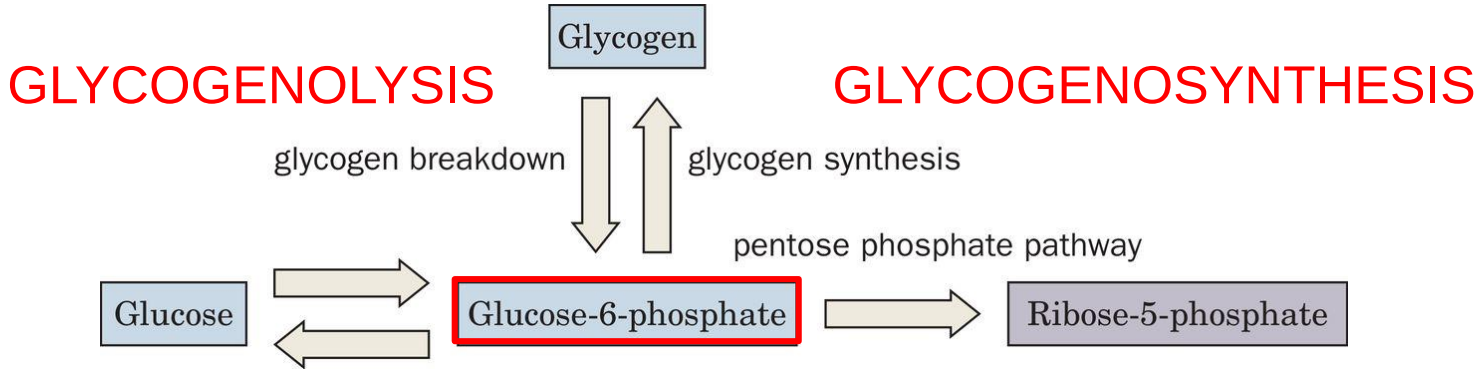
Nucleotid synthesis in cells

Pentose phosphate pathway produts can be entered to glycolysis

NADH: synthesis ATP

Overview of Glucose Metabolism

CYTOSOL

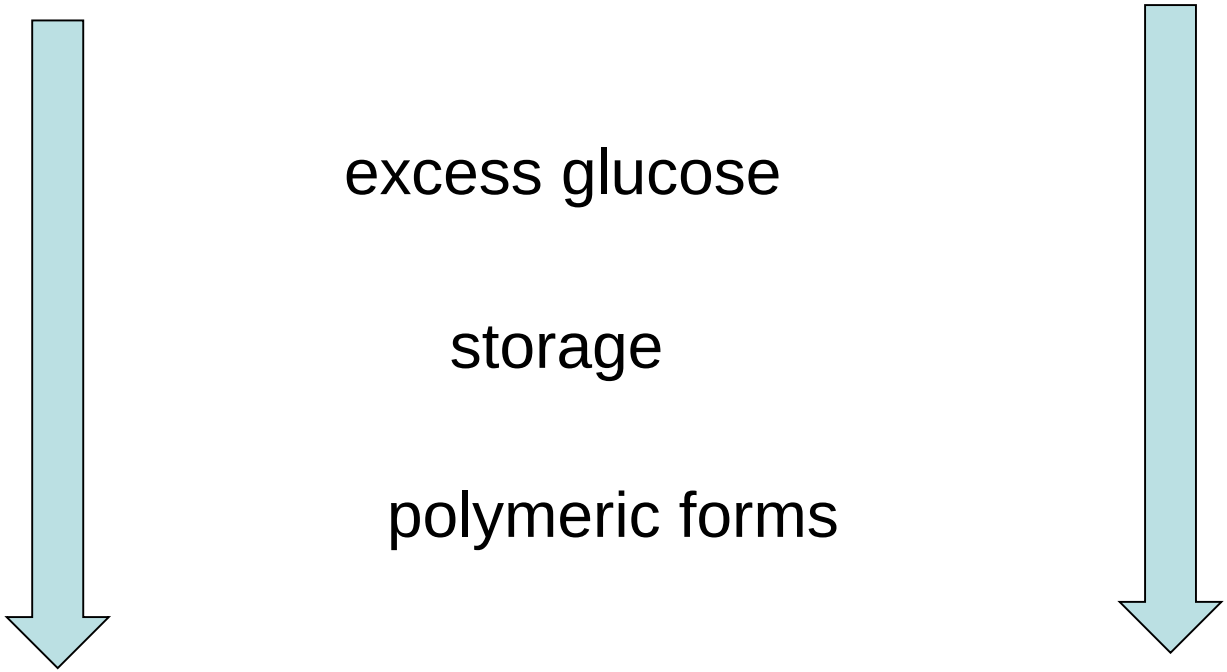


The two glycogen storage tissues, liver and muscle, store and then metabolize this glucose polymer for different purposes

Plants

Bacteria

Vertebrates



excess glucose

storage

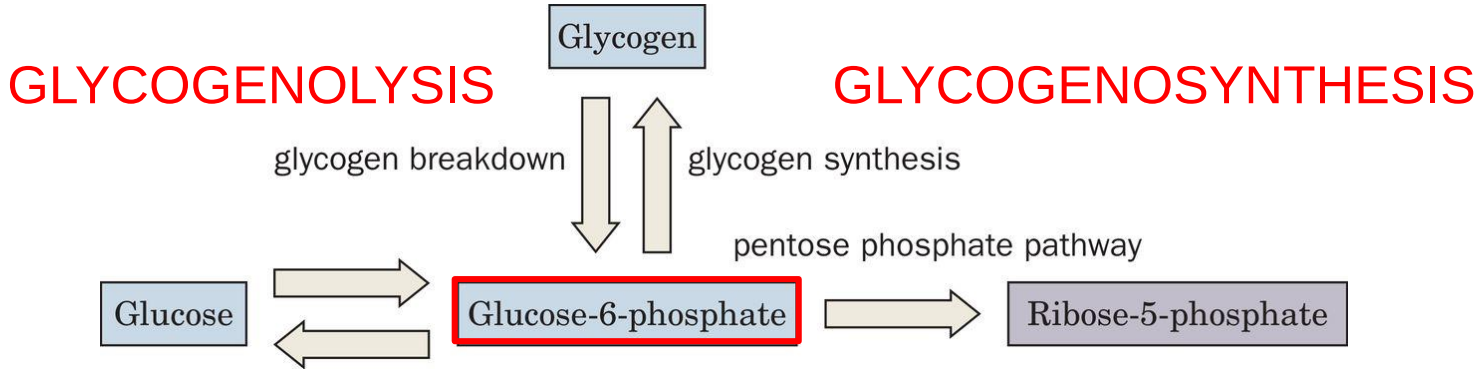
polymeric forms

Starch in plants

Glycogen

Overview of Glucose Metabolism

CYTOSOL



TTwo glycogen storage tissues, liver and muscle, store and then metabolize this glucose polymer for different purposes and purposes

Glycogen Breakdown

- Glycogen, the storage form of glucose, is a branched polymer.

- Glucose mobilization in the liver involves a series of conversions from glycogen to glucose-1-phosphate to glucose-6-phosphate and finally to glucose.

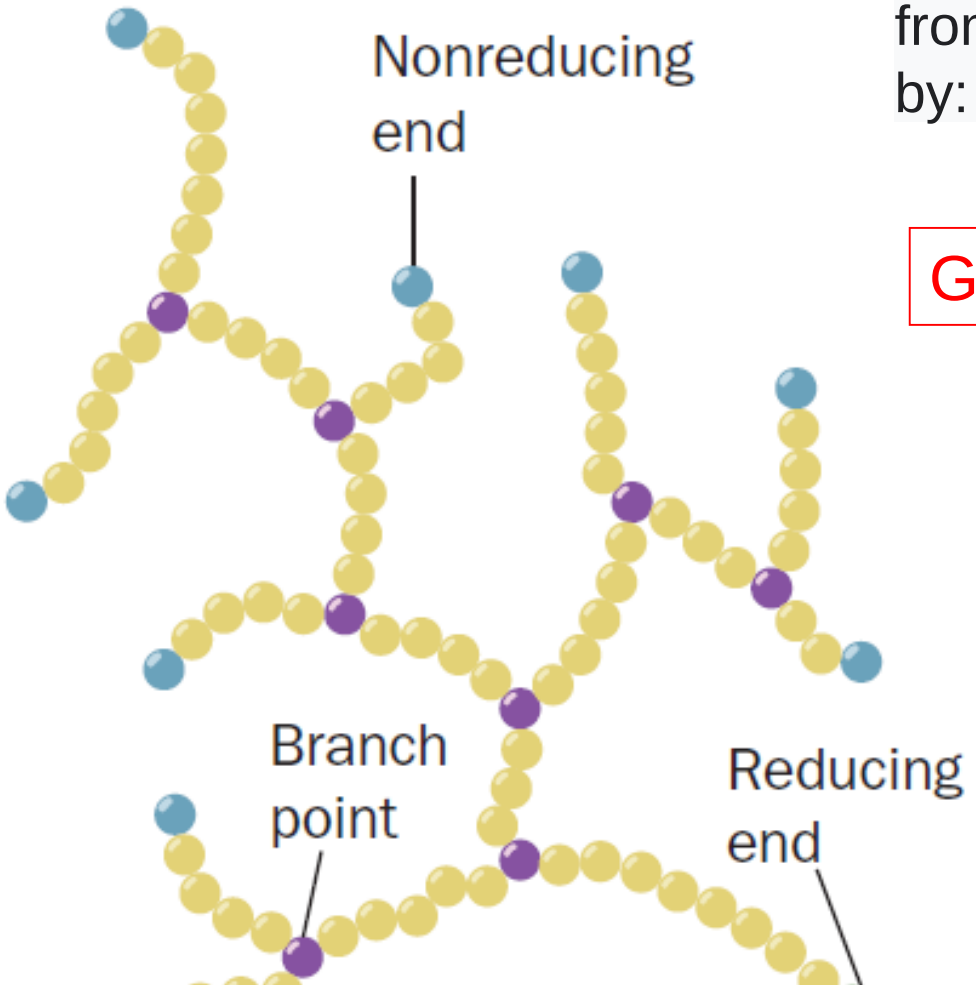
Glycogenolysis is guaranteed by the intervention of three enzymatic activities:

1)Glycogen phosphorylase

2) Deramifying enzyme

3) Phosphoglucomutase

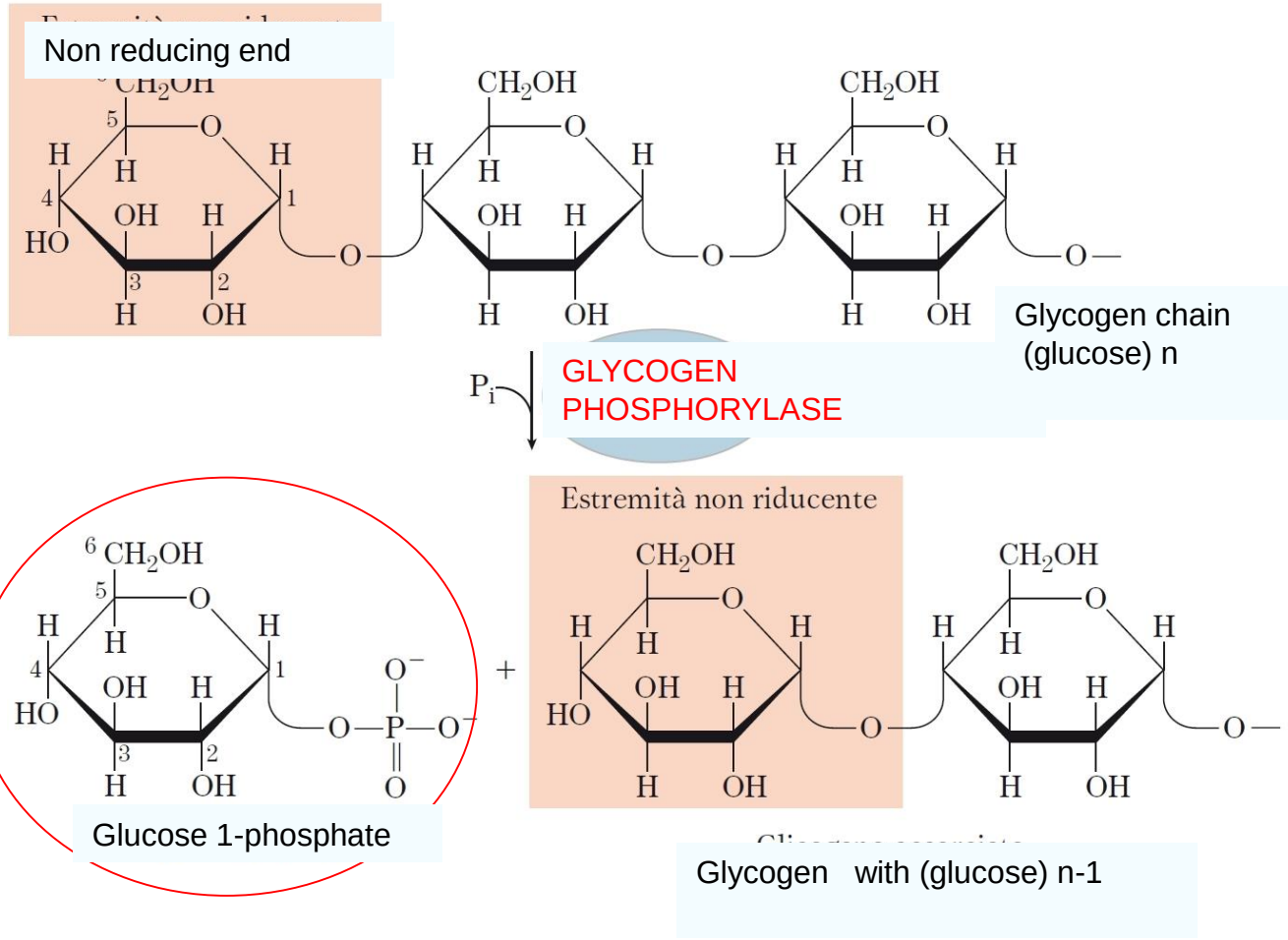
Branched Structure of Glycogen



The demolition of glycogen starts from the NON-REDUCING ends by:

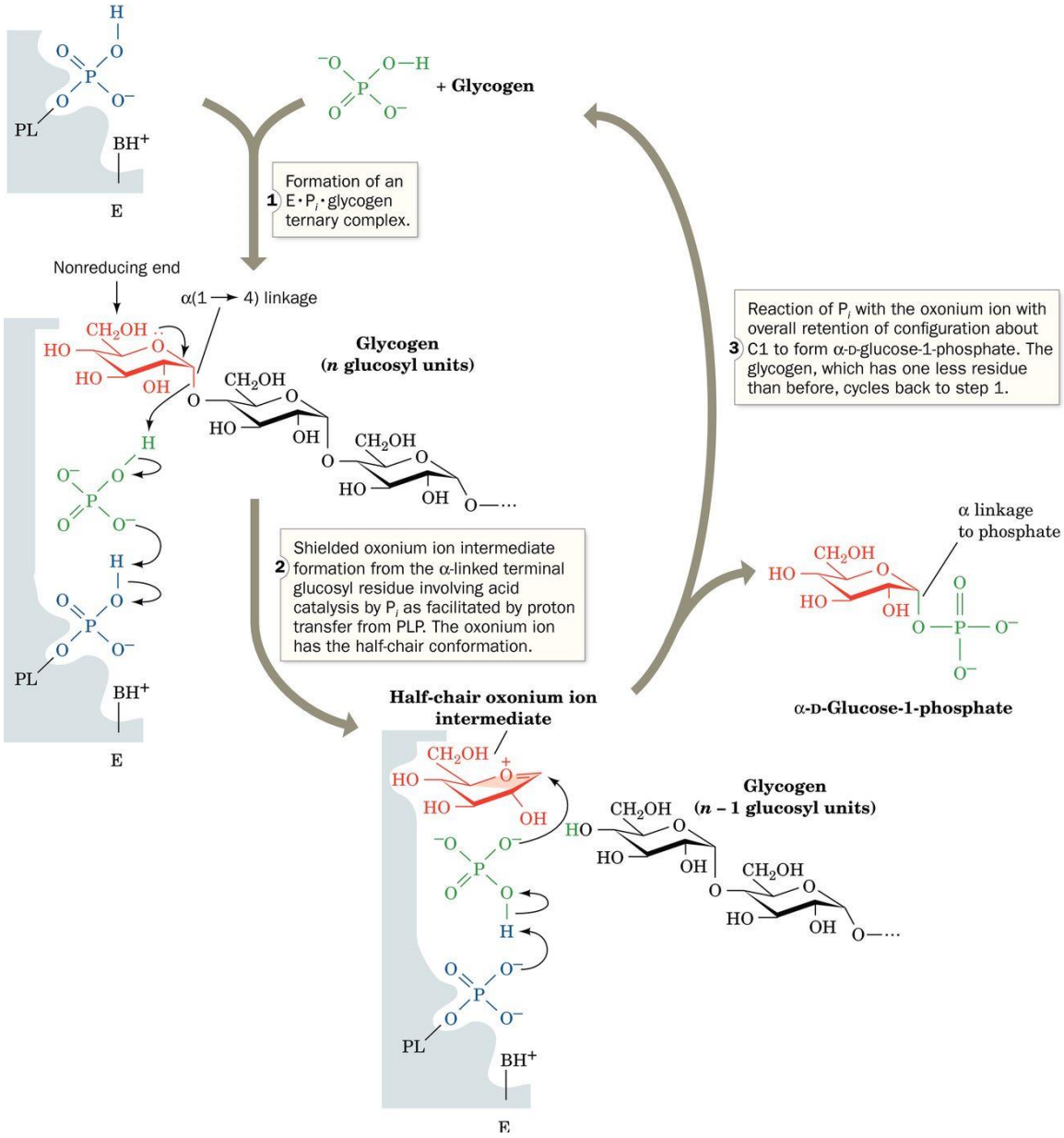
Glycogen phosphorylase

GLYCOGEN PHOSPHORYLASE catalyzes the PHOSPHOROLYSIS of glycogen by detaching G1P only if it is at least 5 units away from the branching



Enzyme removes other glucose residues until it reaches the five residue from the branch

Glycogen Phosphorylase Mechanism

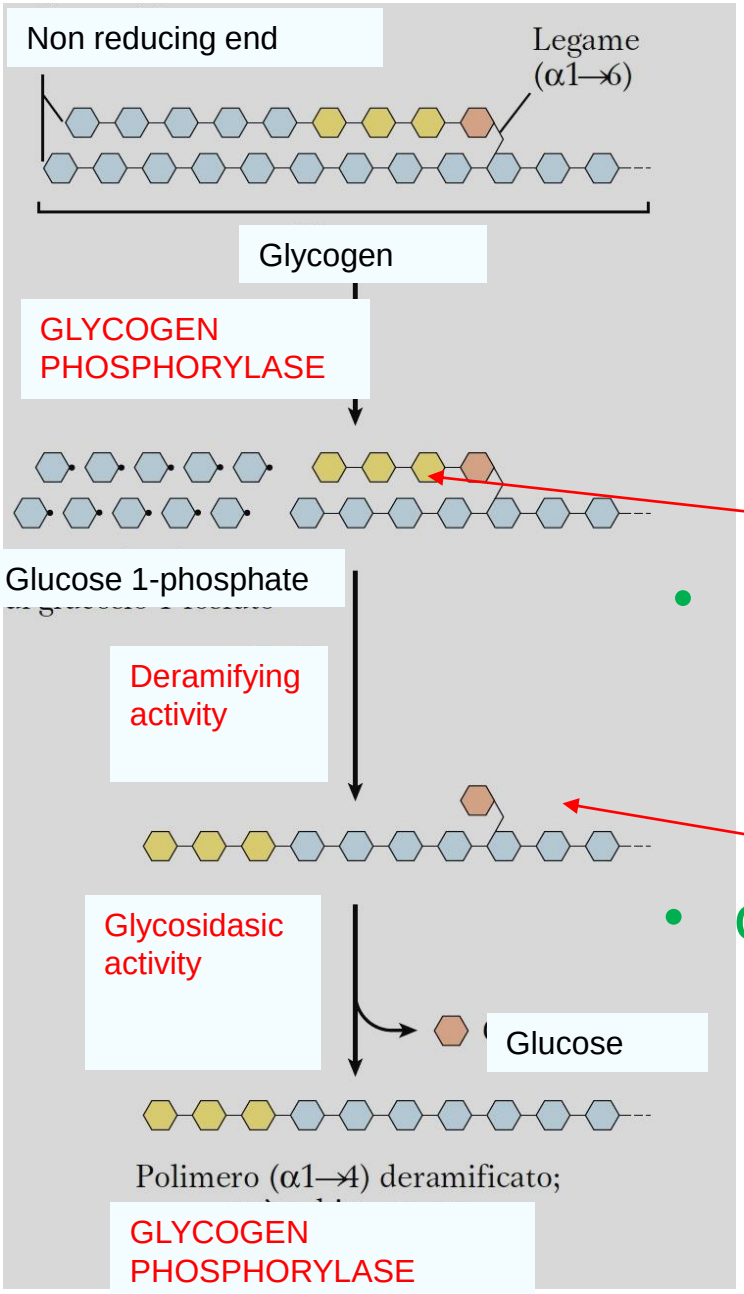


Glycogen phosphorylase uses PLP as a prosthetic group which catalyzes phosphorolysis with its phosphoric group:

A ternary complex

ENZYME + Pi + Glycogen

is formed



The glycogen DERAMIFYING enzyme removes the ramifications making new saccharide units accessible to the action of glycogen phosphorylase - DOUBLE ACTION!

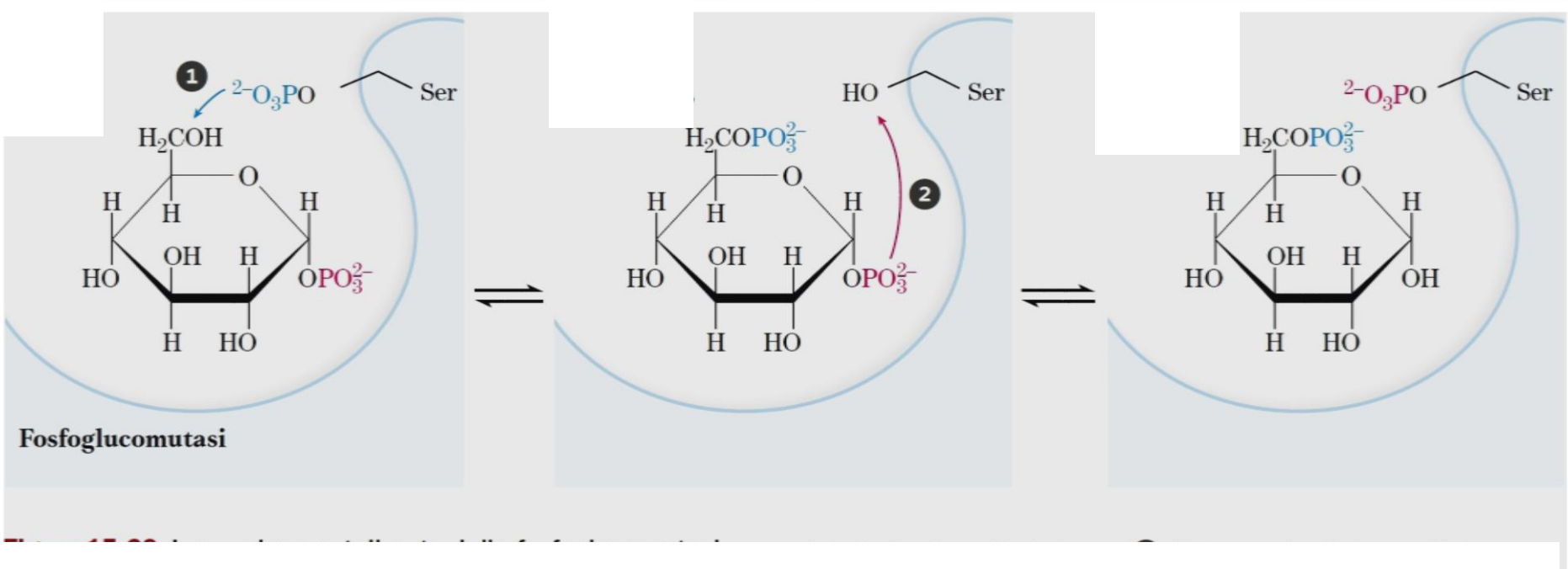
- α(1→4) transglycosylase >> transfers a trisaccharide unit from a branch to a reducing end
- α(1→6) glycosidase >> removes glucose (not G1P) from the branching by HYDROLYSIS (non-phosphorolysis)

Phosphoglucomutase Mechanism

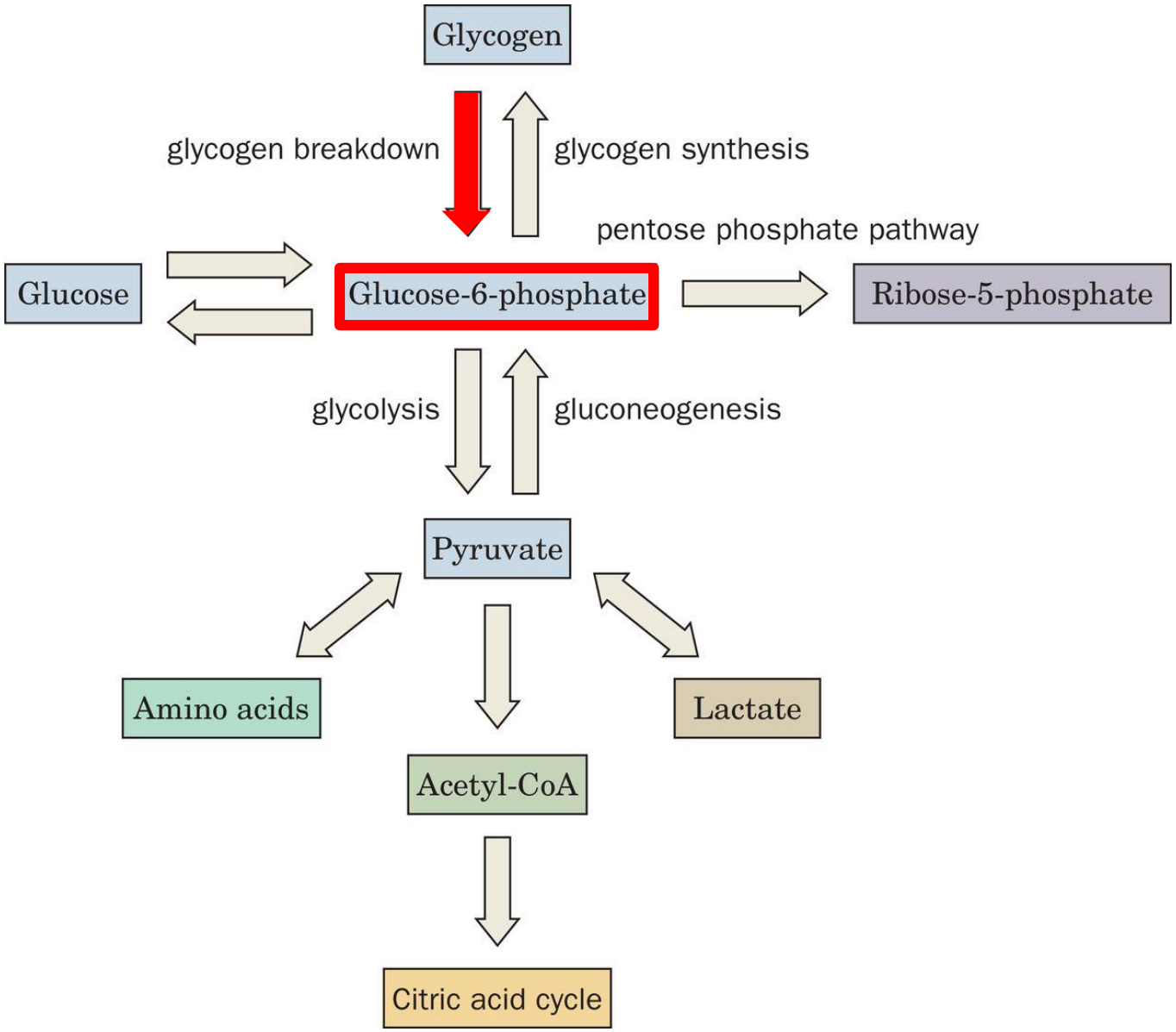
Glucose1-P

[Glucose-1,6-P]

Glucose-6-P



Glucose 6-phosphate can follow different metabolic fates…..

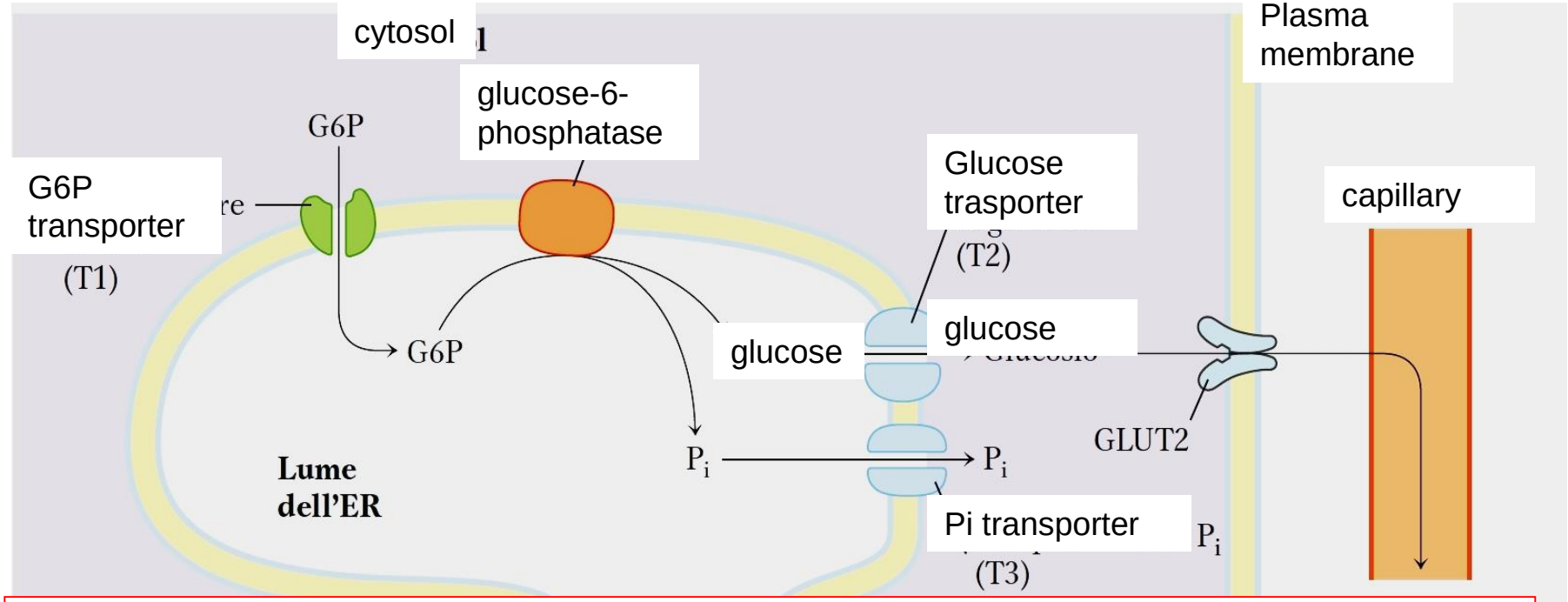


Glucose-6-phosphate----------→ Glucose

only in the liver

Glucose-6-phosphatase (G6Pase)

Produce free glucose in the endoplasmic reticulum (ER)



Muscle does not has glucose-6-phosphatase (G6Pase)

Von Gierke Disease (GSD-I): glucose 6-phosphatase deficiency (incidenza ca.1/100.000)

The deficiency impairs the liver's ability to produce glucose free from glycogen and via gluconeogenesis. Causes severe hypoglycemia during fasting and increased glycogen in the liver and kidneys with their enlargement, convulsions. Other metabolic problems include lactic acidosis and hyperlipidemia.
Therapy: Frequent or continuous feeding with carbohydrates.

McArdle Disease (GSD-V): deficiency of myophosphorylase.
Its incidence is reported as 1 in 100,000, approximately the same as glycogen storage disease type I. Symptoms include exercise intolerance with myalgia, early fatigue, painful cramps, weakness of exercising muscles and myoglobinuria.

Pompe Disease (GSD-II) caused by an accumulation of glycogen in the lysosome due to deficiency of the lysosomal acid α-1,4-glucosidase enzyme. Damages to muscle and nerve cells throughout the body. The build-up of glycogen causes progressive muscle weakness (myopathy) throughout the body and affects various body tissues, particularly heart, skeletal muscles, liver and nervous system.

Glycogen Synthesis

Liver glycogen synthesis involves a series of conversions from

glucose -> glucose-6-phosphate -> UDP–glucose -> glycogen

- UDP–glucose is an activated molecule.

- Glycogen is extended from a primer and by the protein glycogenin.

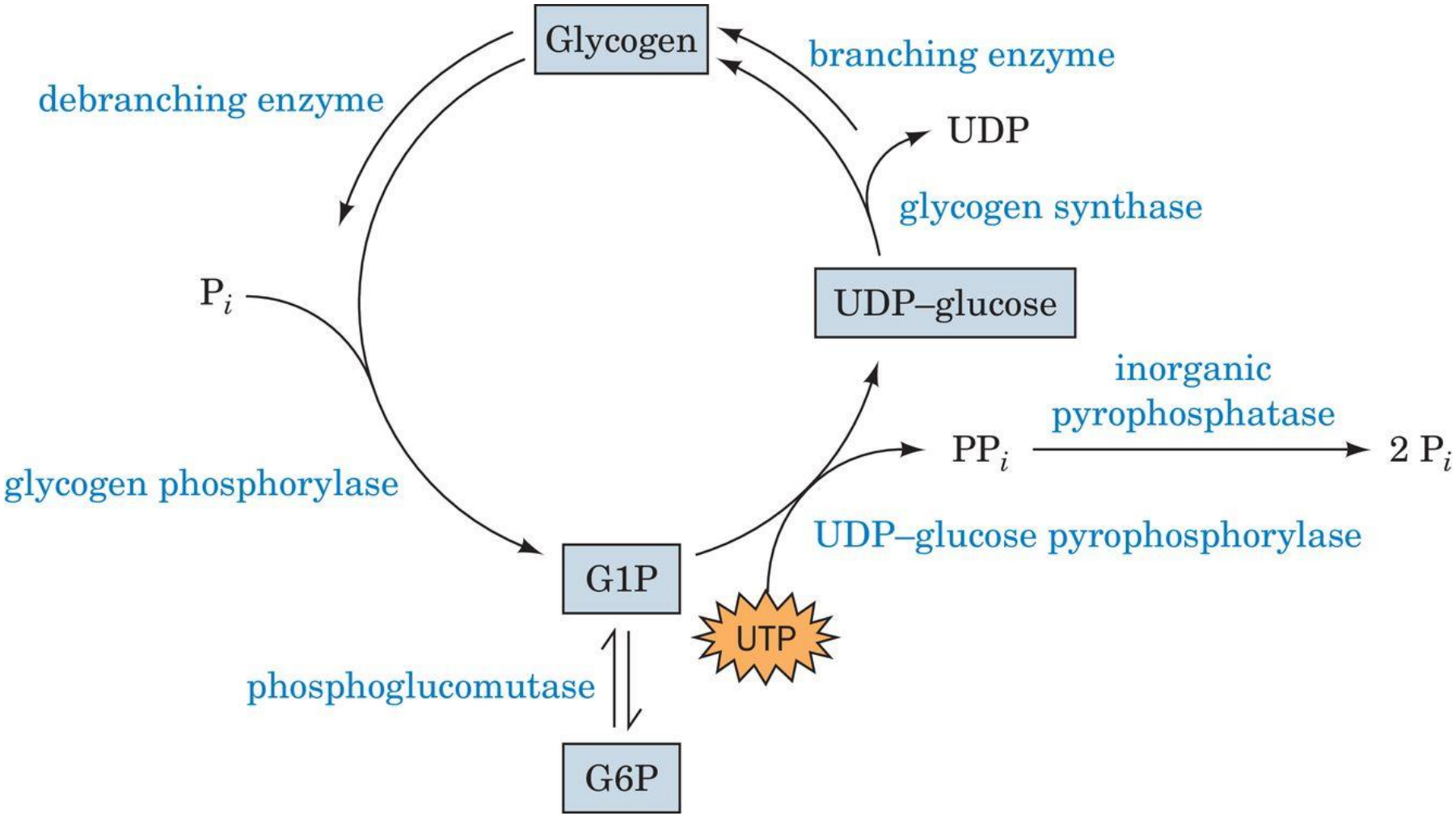
Glycogenosynthesis is ensured by the intervention of three enzymatic activities:

1) UDP-glucose pyrophosphorylase

2) Glycogen synthetase (or synthase)

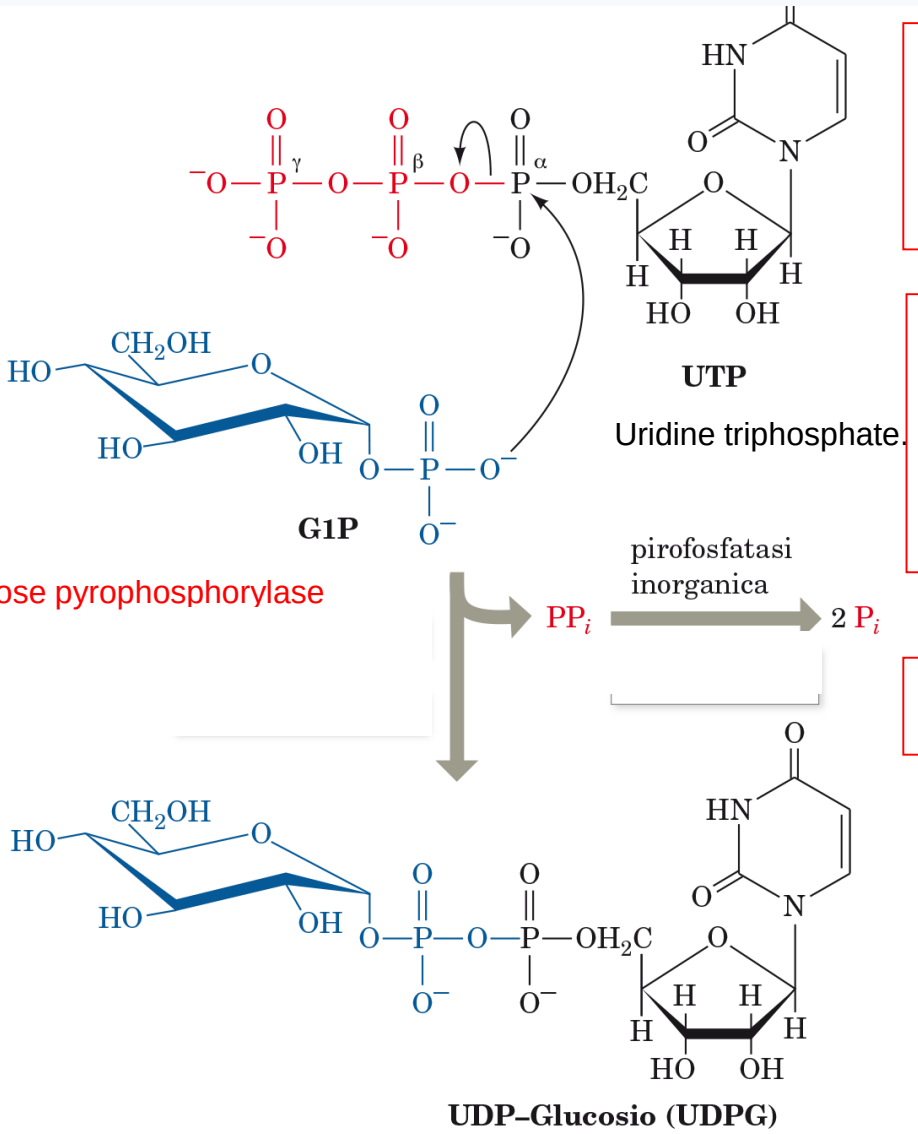
3) Glycogen branching enzyme

Opposing Glycogen Pathways: Synthesis & Degradation



1) UDP-glucose pyrophosphorylase

The biosynthesis of glycogen goes through the formation of a sugar bound to a nucleotide



Glucose 1 phosphate is converted to UDP-glucose by the action of UDP-glucose pyrophosphorylase, in a key step of glycogen biosynthesis.

The negatively charged oxygen on the sugar phosphate acts as a nucleophile, attacking the phosphate of the nucleoside triphosphate and displacing pyrophosphate.

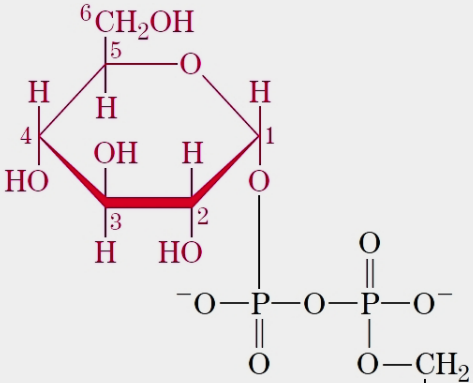
Pyrophosphate is rapidly hydrolyzed by inorganic pyrophosphatase

The hydrolysis of pyrophosphate to Pi makes the EXERGONIC reaction (-19kJ / mol) therefore spontaneous

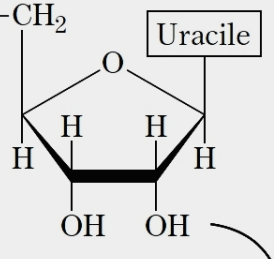
2) Glycogen synthetase (or synthase)

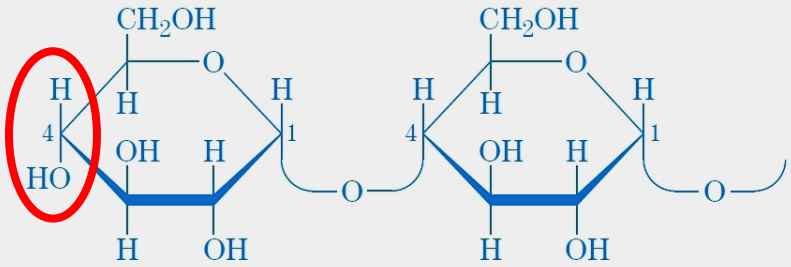
UDP-glucose is the immediate donor of glucose residues in the reaction catalyzed by glycogen synthase,

Glycogen synthase transfers a glucose residue from UDP-glucose to a nonreducing end of a branched glycogen molecule



UDP-glucose.



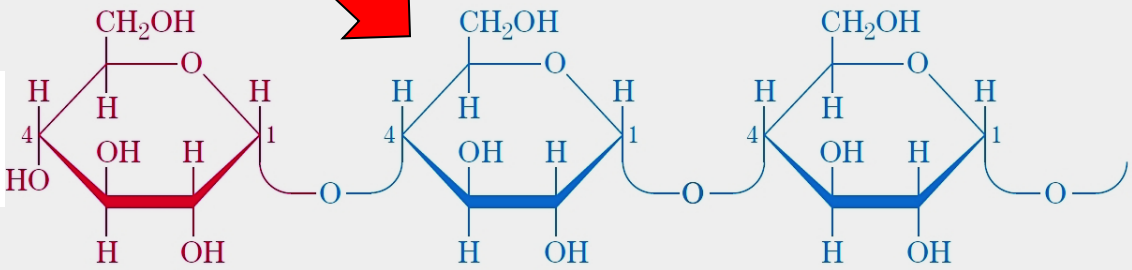


Nonreducing end of a glycogen chain with n residues (n > 4)

Glycogen sinthase

UDP

New nonreducing end



Elongated glycogen with n + 1 residues

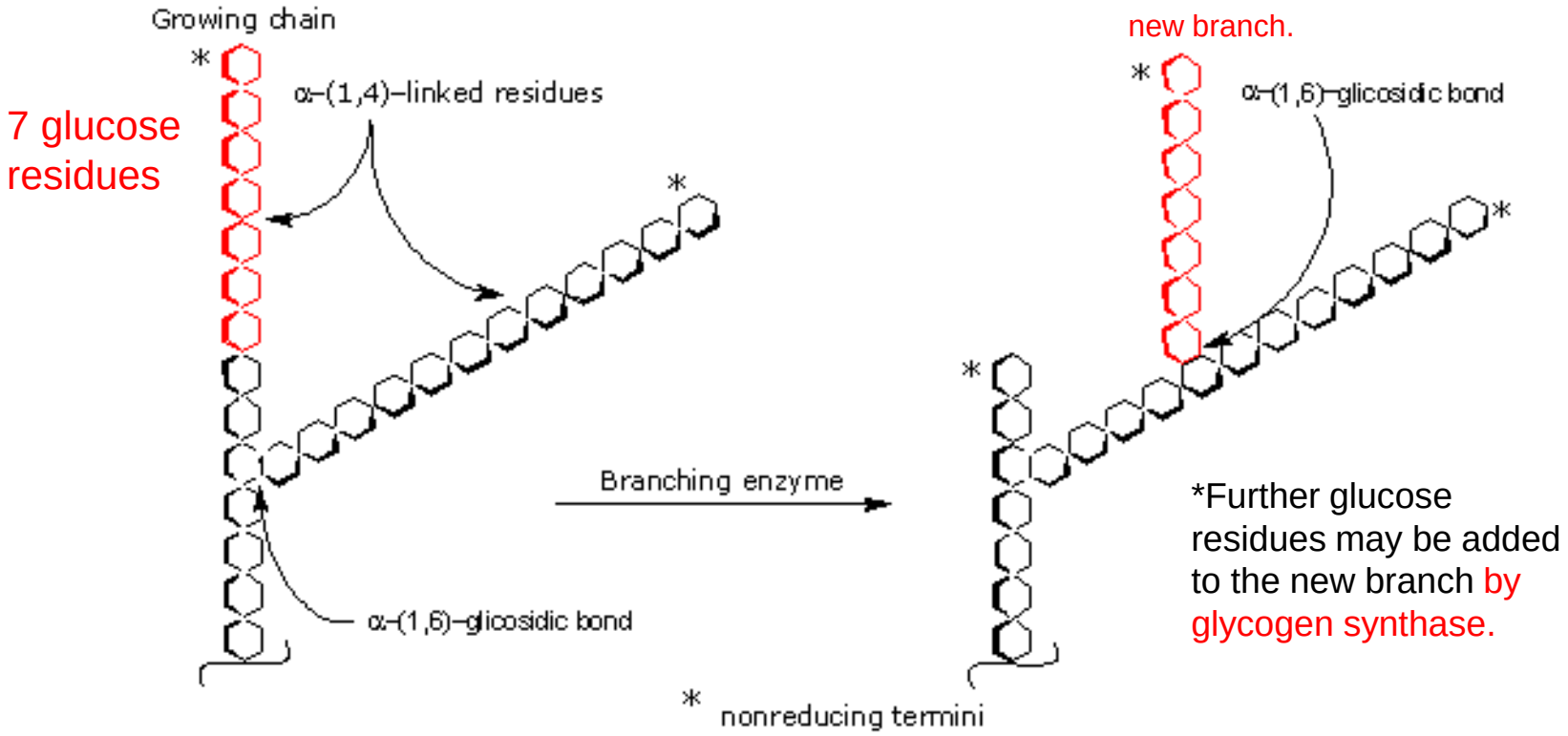
UDP + ATP → UTP + ADP

Glycogen synthase can not make (1-> 6) bonds at the branch points of glycogen.

3) Glycogen branching enzyme

also called amylo (1->4) to (1->6) transglycosylase, or glycosyl- (4->6) transferase.

Glycogen-branching enzyme catalyzes transfer of a terminal fragment of 6 or 7 glucose residues from the nonreducing end of a glycogen branch having at least 11 residues to the C-6 hydroxyl group of a glucose residue at a more interior position of the same or another glycogen chain, thus creating a new branch.

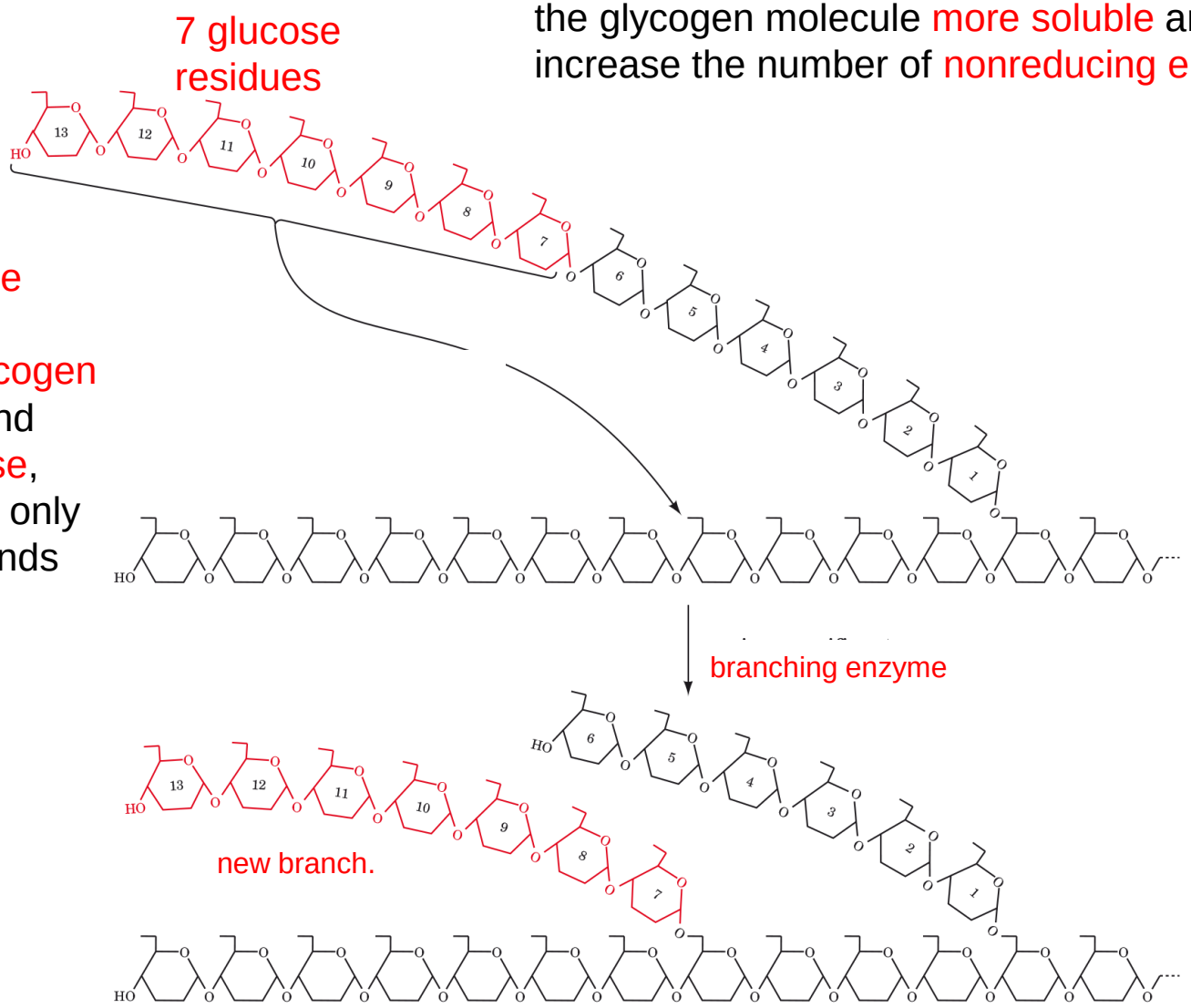


*Further glucose residues may be added to the new branch by glycogen synthase.

Glycogen-branching enzyme,

The biological effect of branching is to make the glycogen molecule more soluble and to increase the number of nonreducing ends.

This increases the number of sites accessible to glycogen phosphorylase and glycogen synthase, both of which act only at nonreducing ends

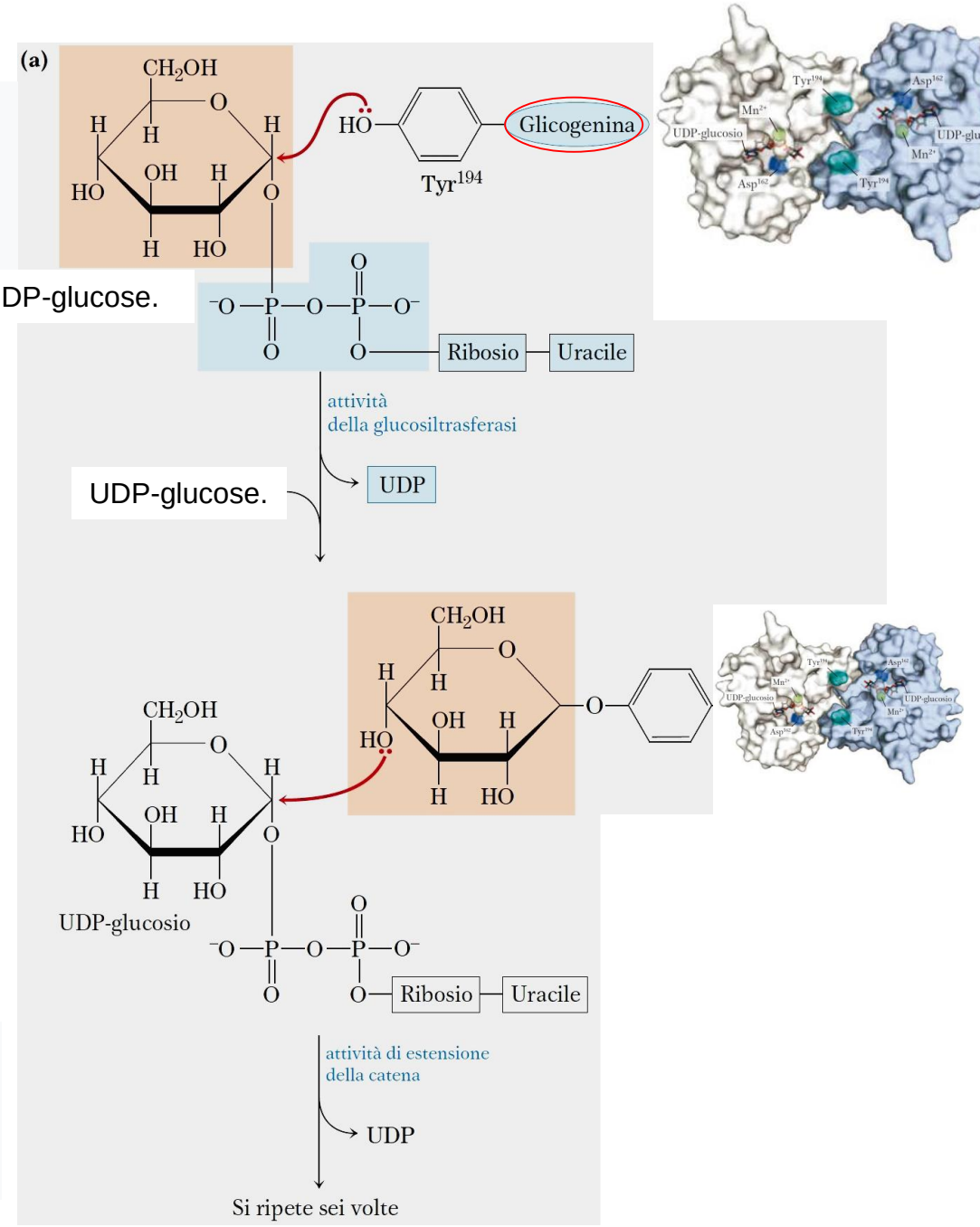


GLYCOGEN SYNTHASE cannot de novo create a glycogen molecule, but only stretch a pre-existing molecule.

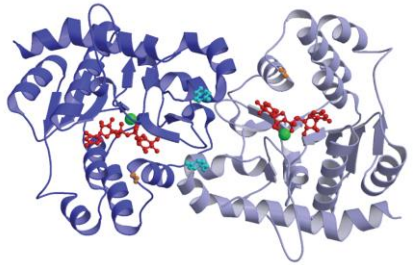
GLYCOGENIN

The GLYCOGENIN protein attaches 8 glucose units donated by UDP-G to the OH group of its tyrosine residue forming a PRIMER for glycogen synthase.

There is one molecule of glycogenin per molecule of glycogen.



GLYCOGENIN



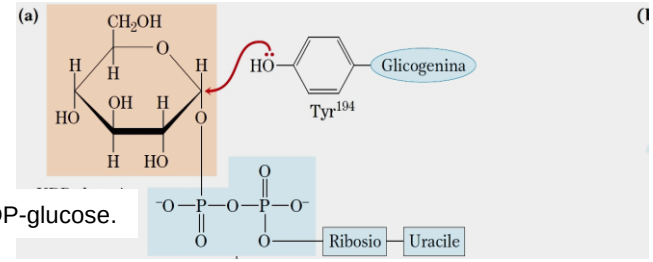
It is a protein of 332 amino acids that acts as a trigger for the attack of the first glucose molecule, but is also endowed with enzymatic activity

1) Glycosyl transferase activity (transfers the glucose residue from UDP-glucose to the OH group of a tyrosine residue (Tyr 194)

2) Chain extension activity (adds 7 glucose residues provided by UDP-glucose)

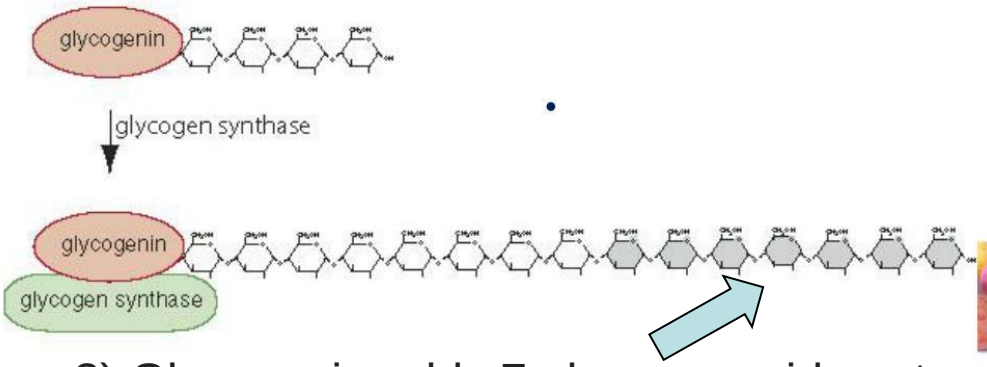
In solution it has a dimeric form and each monomer has catalytic activity mediated by a cofactor, Mn2 +

1) Binding of UDP-glucose to tyrosine 194 of glycogenin (glycosyl transferase activity)



UDP-glucose.

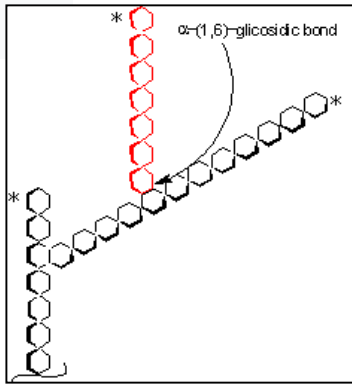
2) Binding of glycogenin to glycogen synthase



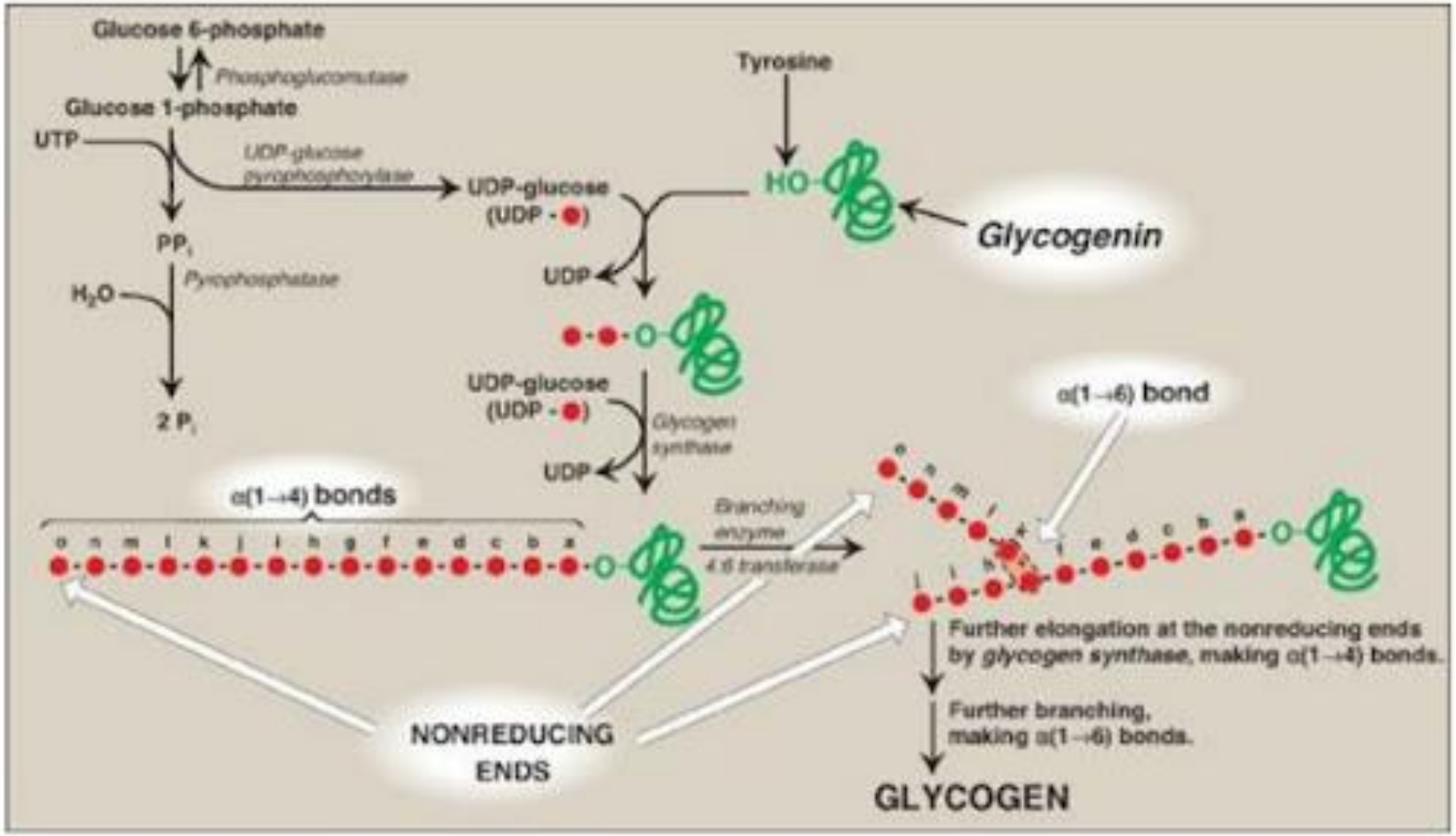
3) Glycogenin adds 7 glucose residues to the nascent chain

4) Dissociation of glycogenin from glycogen synthase

5) Glycogen synthase lengthens the chain



6) Glycogen branching enzyme creates the branching


Glucose 6-phosphate
Phosphoglucomutase
Glucose 1-phosphate
UTP
UDP-glucose pyrophosphorylase
PPi
H2O
Pyrophosphatase
2 Pi
UDP-glucose (UDP - ●)
UDP
Tyrosine
HO
Glycogenin
UDP-glucose (UDP - ●)
Glycogen synthase
UDP
α(1→6) bond
α(1→4) bonds
o n m l k j i h g f e d c b a
Branching enzyme
4:6 transferase
Further elongation at the nonreducing ends by glycogen synthase, making α(1→4) bonds.
Further branching, making α(1→6) bonds.
NONREDUCING ENDS
GLYCOGEN