

HEXOSE MONO PHOSPHATE SHUNT

BIOCHEMISTRY

- The pentose phosphate pathway (also called the hexose monophosphate shunt, or 6-phosphogluconate pathway) occurs in the cytosol of the cell.
- It includes two, irreversible oxidative reactions, followed by a series of reversible sugar–phosphate interconversions

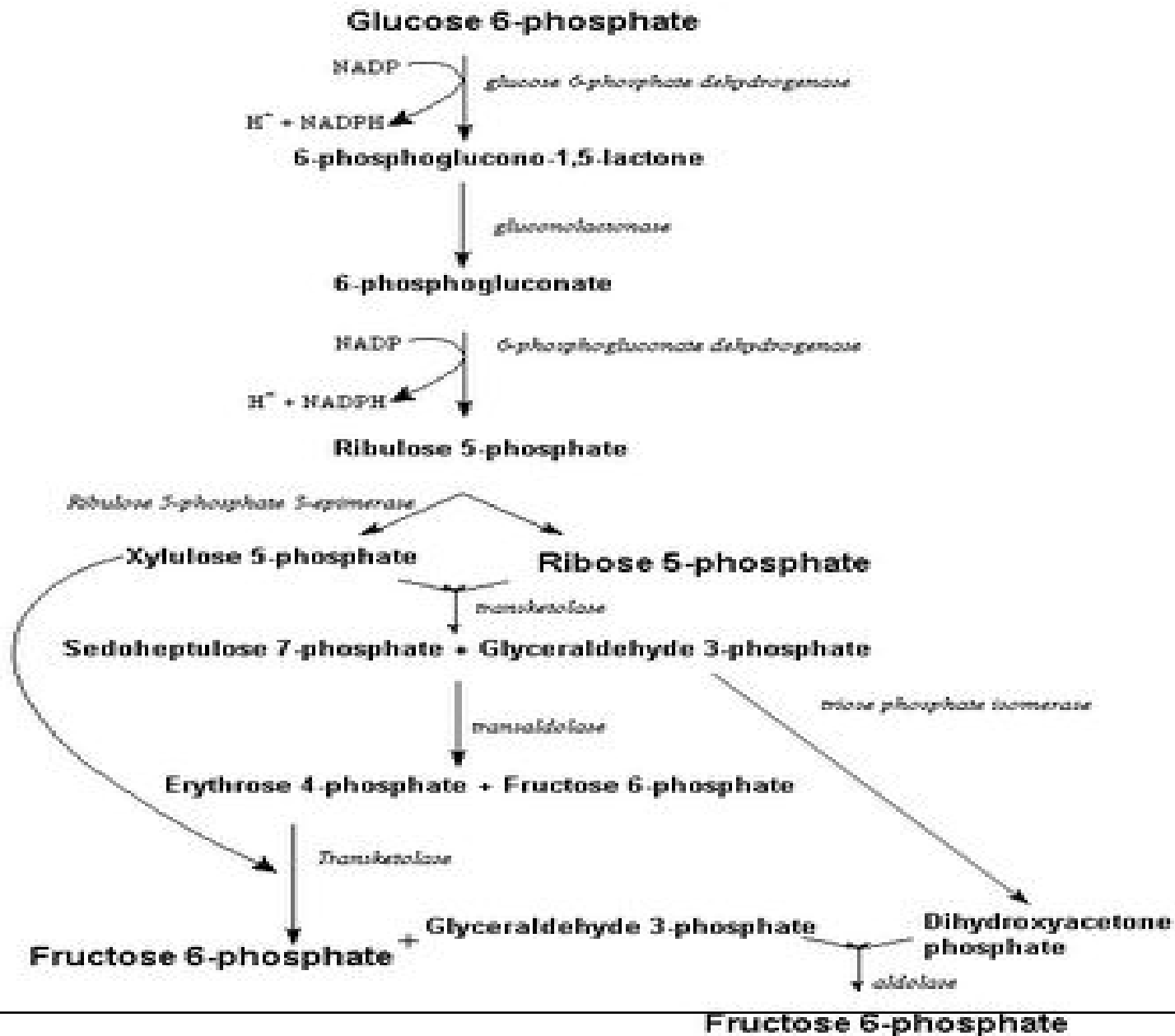
- No ATP is directly consumed or produced in the cycle. Carbon one of glucose 6-phosphate is released as CO_2 , and two NADPH are produced for each glucose 6-phosphate molecule entering the oxidative part of the pathway

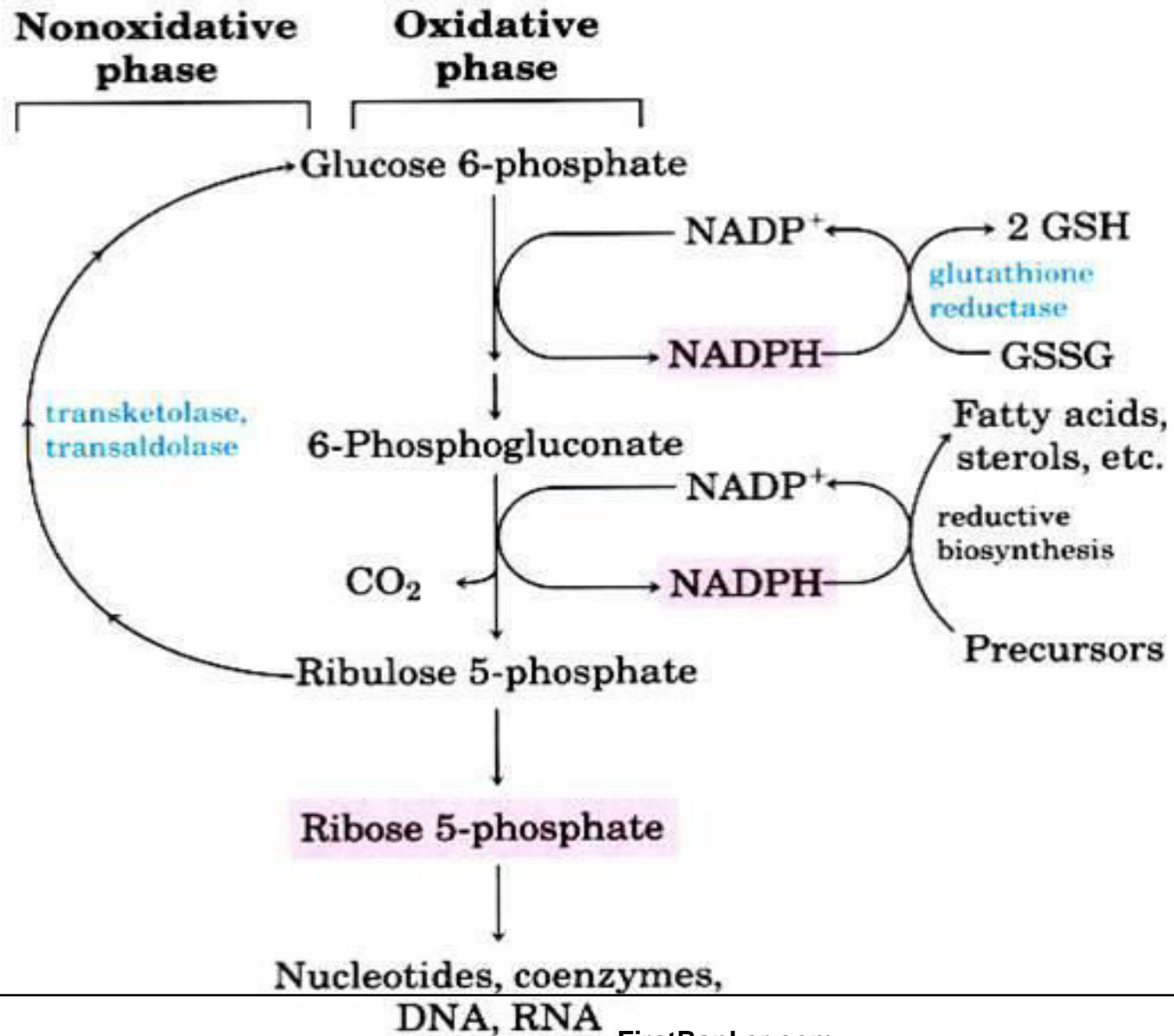
- The pathway provides a major portion of the body's NADPH, which functions as a biochemical reductant.

- It also produces ribose 5-phosphate, required for the biosynthesis of nucleotides and provides a mechanism for the metabolic use of five-carbon sugars obtained from the diet or the degradation of structural carbohydrates in the body.

HMP SHUNT [Hexose Monophosphate Pathway] / PPP [Pentose Phosphate Pathway]

- Principle pathway for oxidation of Glucose-Glycolysis & TCA
- **HMP**-alternative pathway for oxidation of Glucose-not for Energy
- Occours in cytosol
- **Provides: NADPH** [reductive synthesis] & **Pentoses** [for NA synth]
- Most common at the site of synthesis:
Liver [phospholipid, FA synth, Cholesterol], **adipose tissue** [FA synth],
lactating mammary gland [FA synth], **adrenal cortex** [Cholesterol, steroid hormone synth], **testes** & other endocrine glands concerned with steroid synthesis and **RBC**
- **NADPH produced**: required for – **Reductive biosynthesis** of FA, TG, cholesterol, steroids
[NADH – reduction in Catabolic pathways (NADH enters ETC→ ATP);
NADPH – reduction in Synthetic pathways]





- **Three molecules of glucose 6-phosphate give rise to three molecules of CO_2 and three 5-carbon sugars.**
- These are rearranged to regenerate two molecules of glucose 6-phosphate and one molecule of the glycolytic intermediate, glyceraldehyde 3-phosphate.

- Like glycolysis, the enzymes of the pentose phosphate pathway are cytosolic. Unlike glycolysis, oxidation is achieved by dehydrogenation using **NADP⁺**, not **NAD⁺**, as the hydrogen acceptor.

- The sequence of reactions of the pathway may be divided into two phases: an **oxidative nonreversible phase** and a **nonoxidative reversible phase**.

- In the first phase, glucose 6-phosphate undergoes dehydrogenation and decarboxylation to yield a pentose, ribulose 5-phosphate.
- In the second phase, ribulose 5-phosphate is converted back to glucose 6-phosphate by a series of reactions involving mainly two enzymes: **transketolase** and **transaldolase**

Irreversible Oxidative Reactions

- The oxidative portion of the pentose phosphate pathway consists of three reactions that lead to the formation of :
- **Ribulose 5-phosphate**
- **CO₂**
- and **Two molecules of NADPH** for each molecule of glucose 6-phosphate oxidized

- This portion of the pathway is particularly important in the **liver, lactating mammary glands, and adipose, which are active in the biosynthesis of fatty acids**
- **In the adrenal cortex, which is active in the NADPH-dependent synthesis of steroids**
- **In erythrocytes, which require NADPH to keep glutathione reduced.**

- **Glucose 6-phosphate dehydrogenase (G6PD)** catalyzes an irreversible oxidation of **glucose 6-phosphate to 6-phosphogluconolactone** in a reaction that is specific for **NADP⁺** as its coenzyme.

- The pentose phosphate pathway is regulated primarily at the glucose 6-phosphate dehydrogenase reaction. NADPH is a potent competitive inhibitor of the enzyme
- Insulin enhances G6PD gene expression, and flux through the pathway increases in the well fed state.

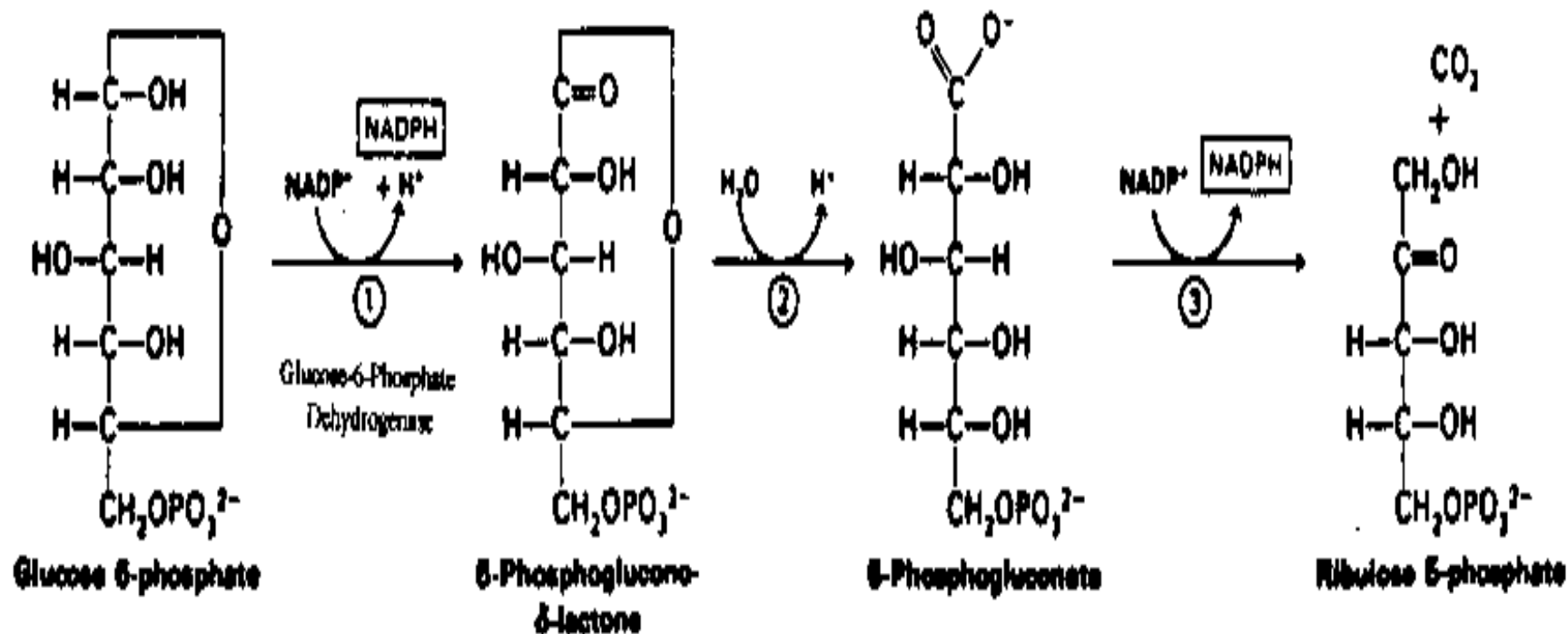
- **6-Phosphogluconolactone** is hydrolyzed by 6-phosphogluconolactone **hydrolase** to **6-phosphogluconate**

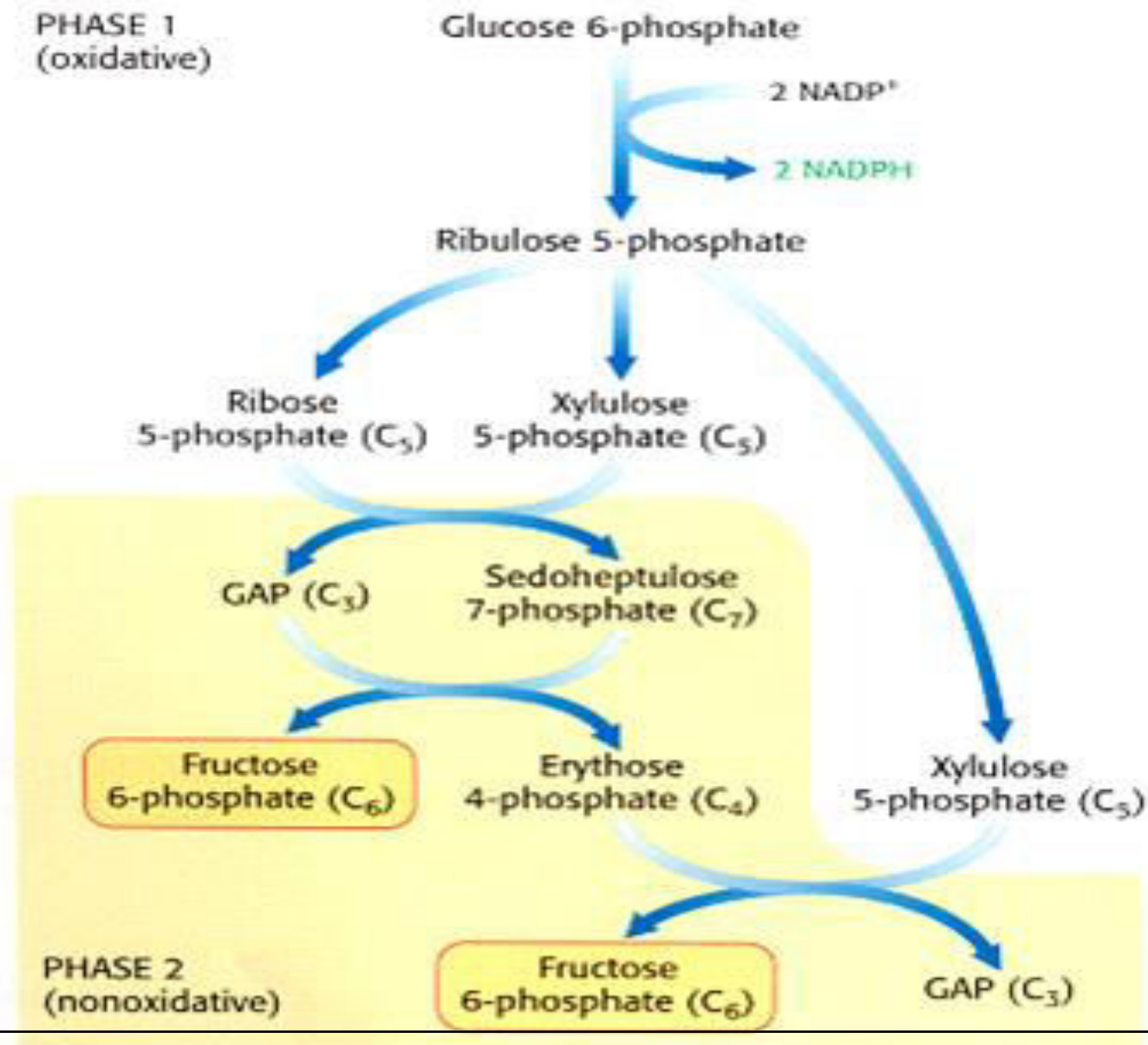
- 6-phosphogluconate is catalyzed by 6-phosphogluconate **dehydrogenase**
- This irreversible reaction produces a pentose sugar–phosphate (**ribulose 5-phosphate**), **CO₂** (from carbon 1 of glucose), and a second molecule of **NADPH**

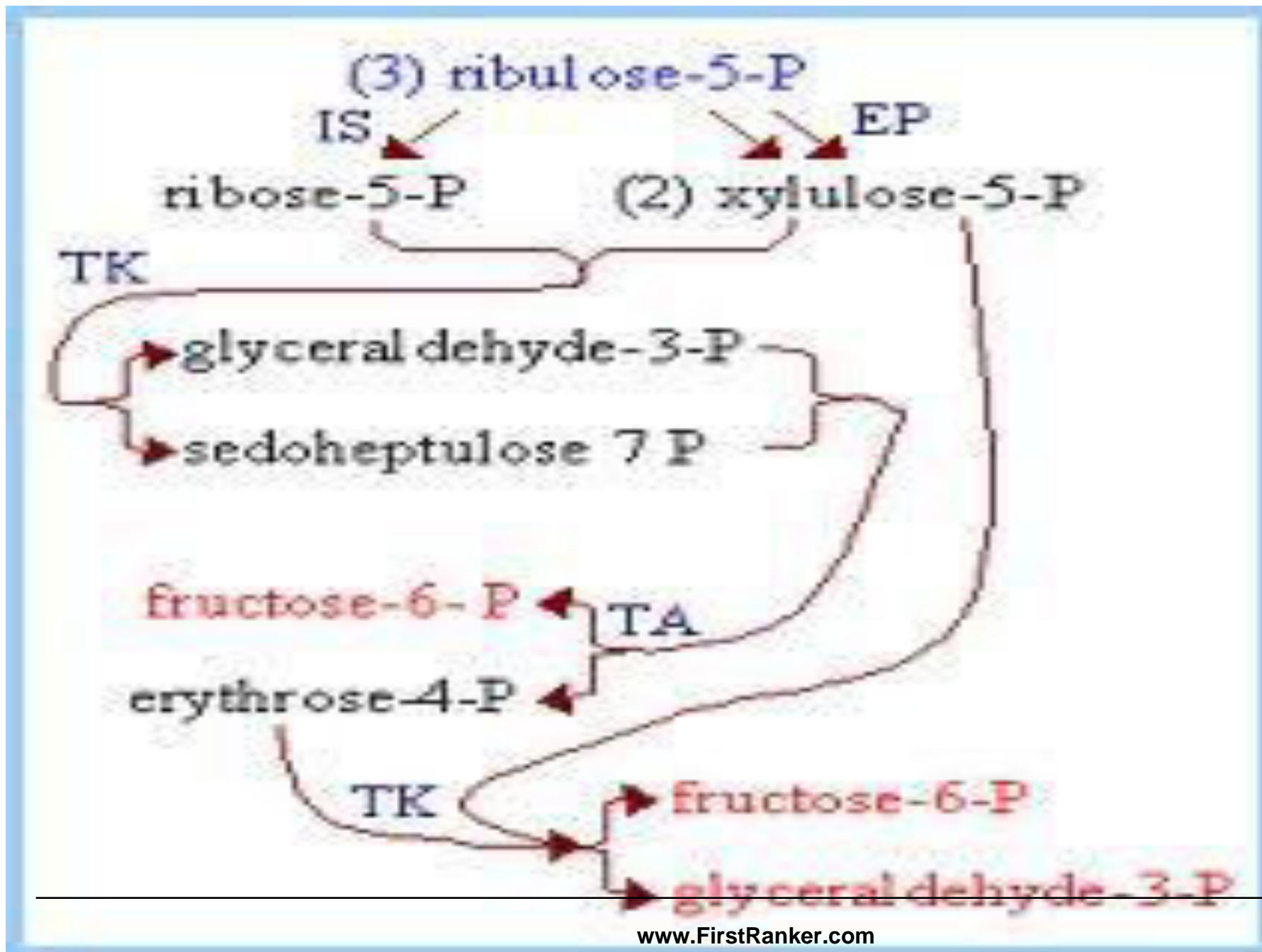
Reversible Nonoxidative Reactions

- **The nonoxidative reactions of the pentose phosphate pathway occur in all cell types** synthesizing nucleotides and nucleic acids. These reactions catalyze the interconversion of three-, four-, five-, six-, and seven-carbon sugars

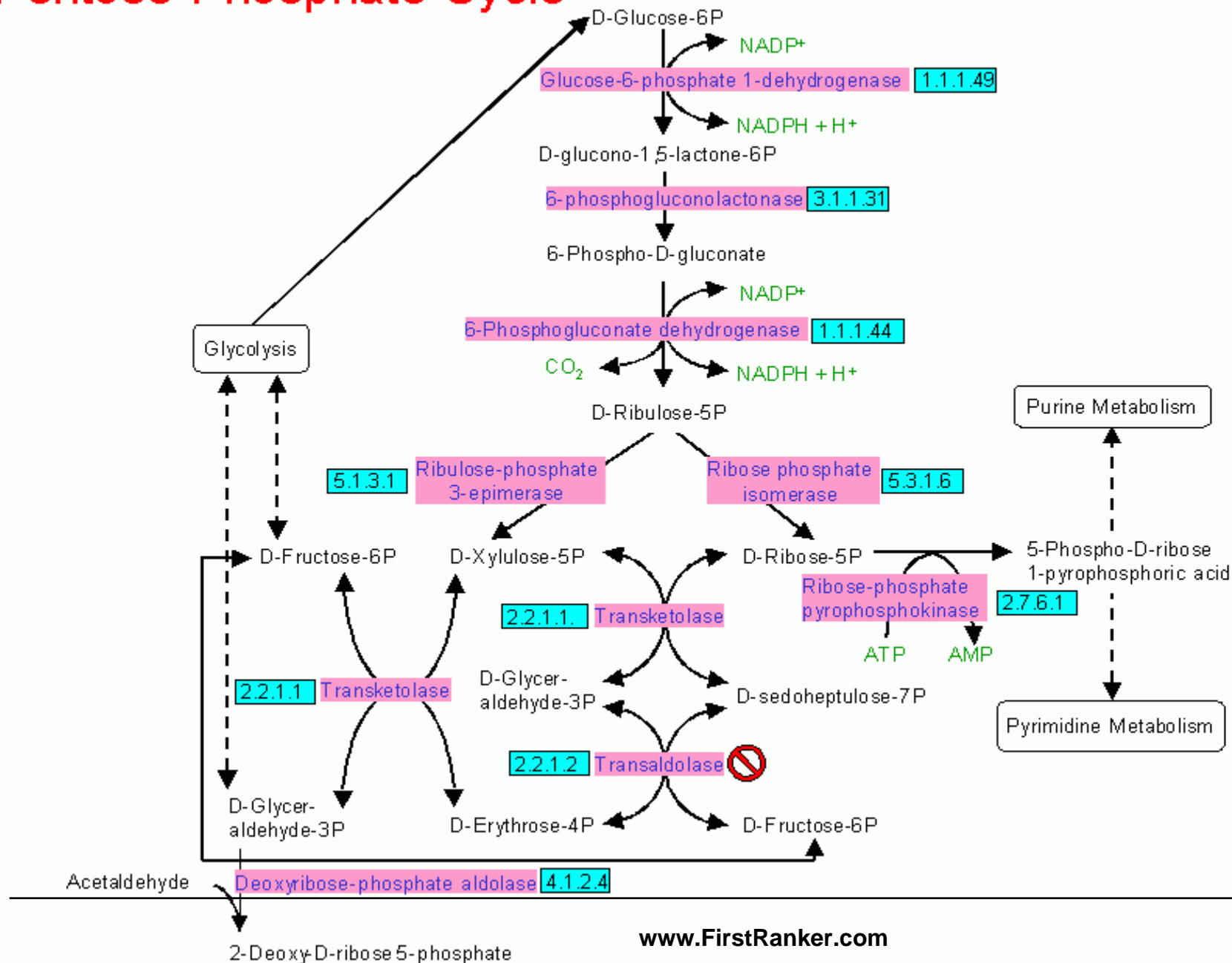
Pentose Phosphate Pathway







Pentose Phosphate Cycle



Uses of NADPH

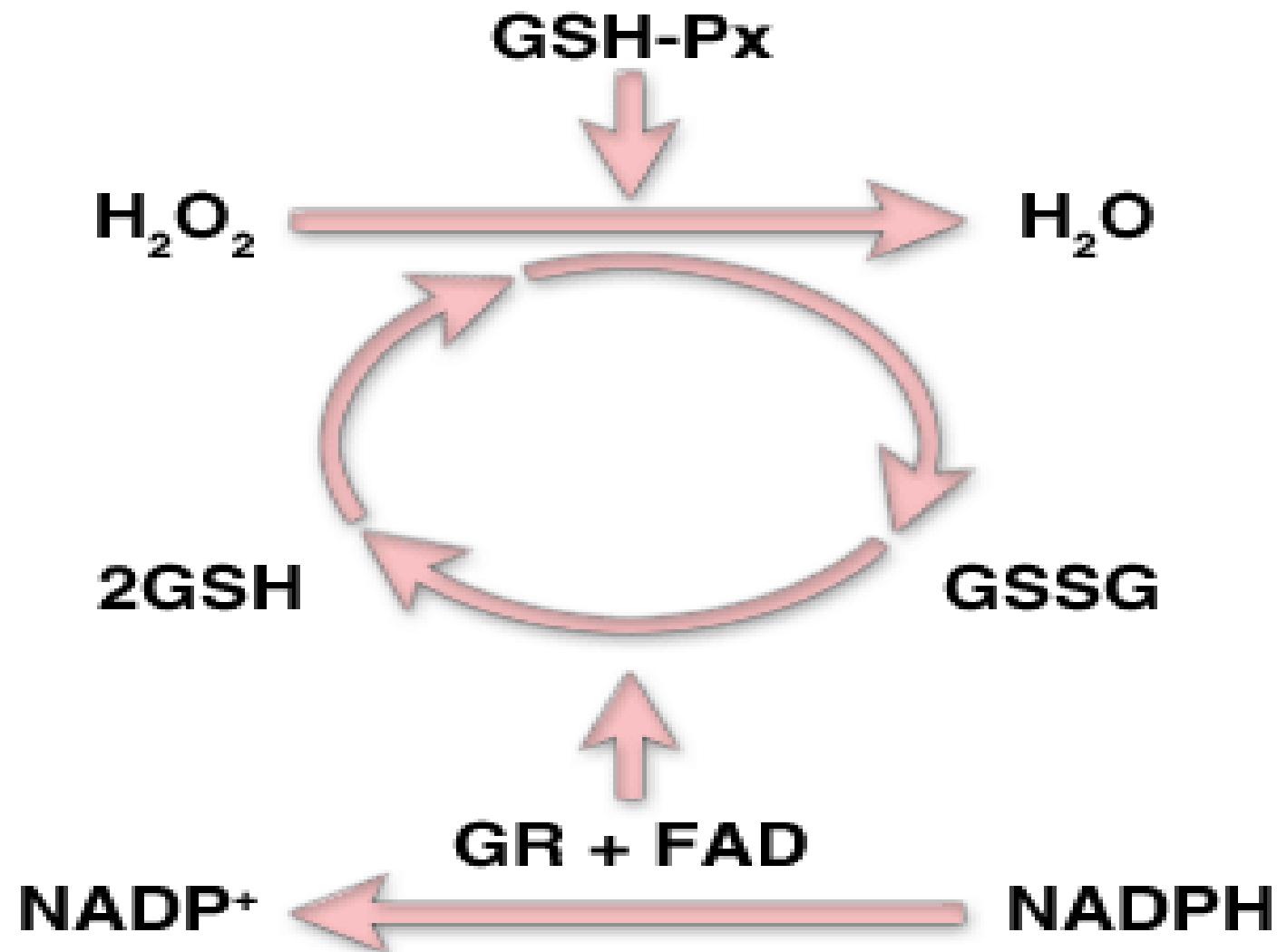
1. Reduction of hydrogen peroxide
2. Cytochrome P450 monooxygenase system
3. Phagocytosis by white blood cells
4. Synthesis of nitric oxide

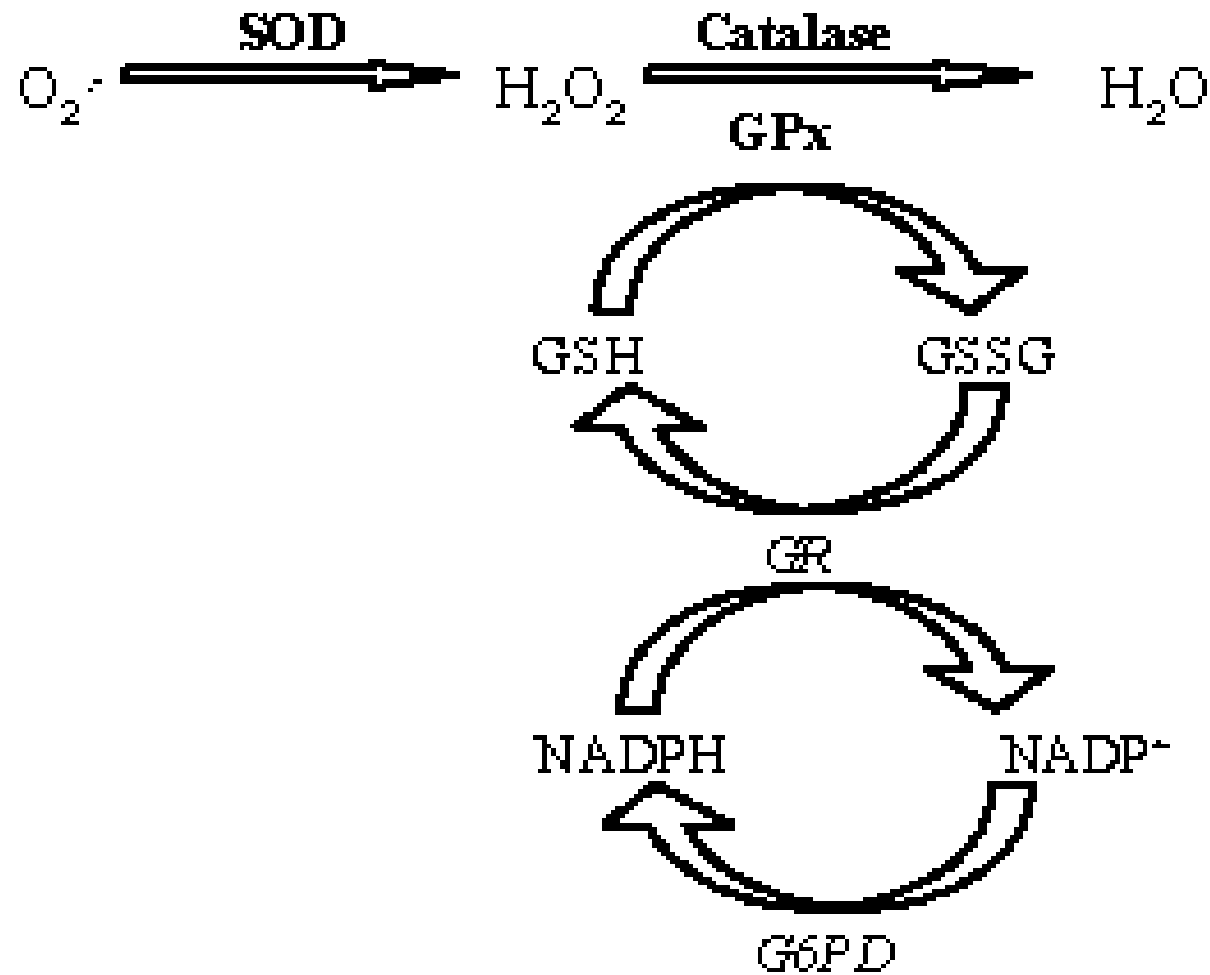
Reduction of Hydrogen Peroxide

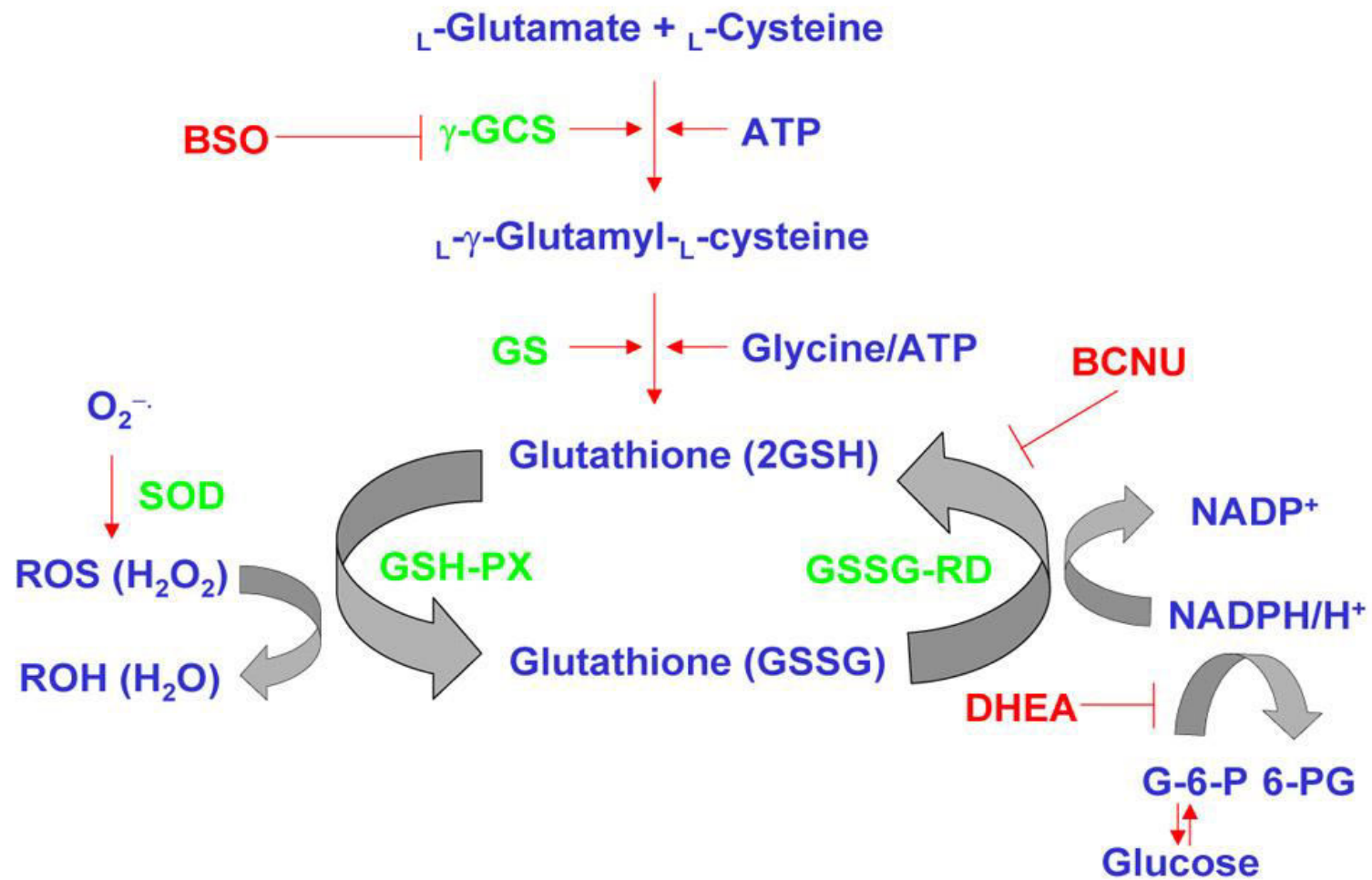
- Reduced glutathione, a tripeptide-thiol (γ -glutamylcysteinylglycine) present in most cells, can chemically detoxify hydrogen peroxide

- This reaction catalyzed by the selenium-requiring glutathione peroxidase, forms oxidized glutathione, which no longer has protective properties.
- The cell regenerates reduced glutathione in a reaction catalyzed by glutathione reductase, using NADPH as a source of reducing electrons.

- Erythrocytes are totally dependent on the pentose phosphate pathway for their supply of NADPH because, unlike other cell types, erythrocytes do not have an alternate source for this essential coenzyme

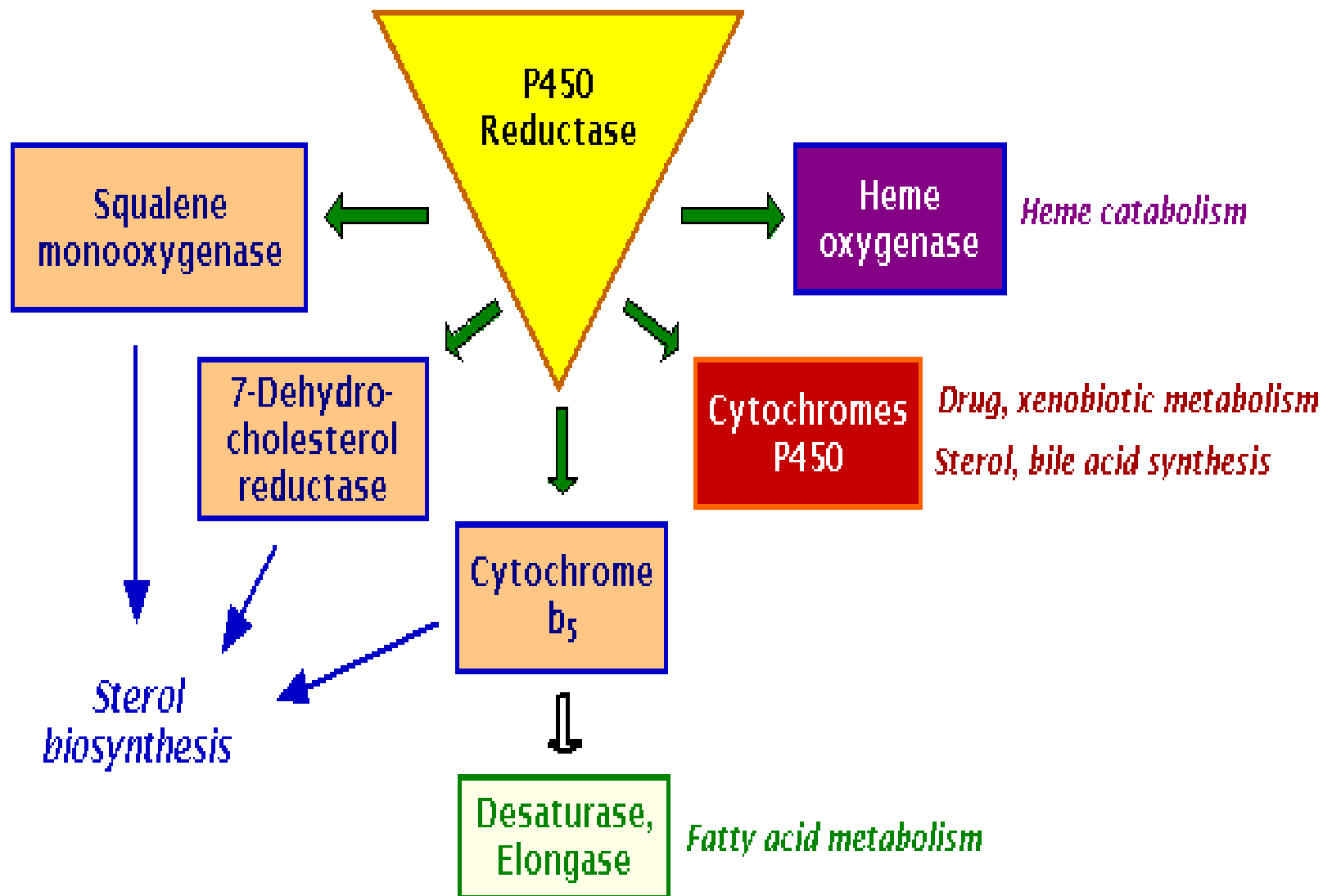






Cytochrome P450 monooxygenase system

- Monooxygenases (mixed function oxidases) incorporate one atom from molecular oxygen into a substrate (creating a hydroxyl group), with the other atom being reduced to water.
- In the cytochrome P450 monooxygenase system, NADPH provides the reducing equivalents required by this series of reactions



Action of mixed function oxidases (cytochrome P450)

- Involve **insertion of a single oxygen** into drug/chemical
- Require **cytochrome P450/NADPH-cytochrome P450 reductases/lipid**
- Found in endoplasmic reticulum of **liver, kidney, lung and intestine**

Phagocytosis by white blood cells

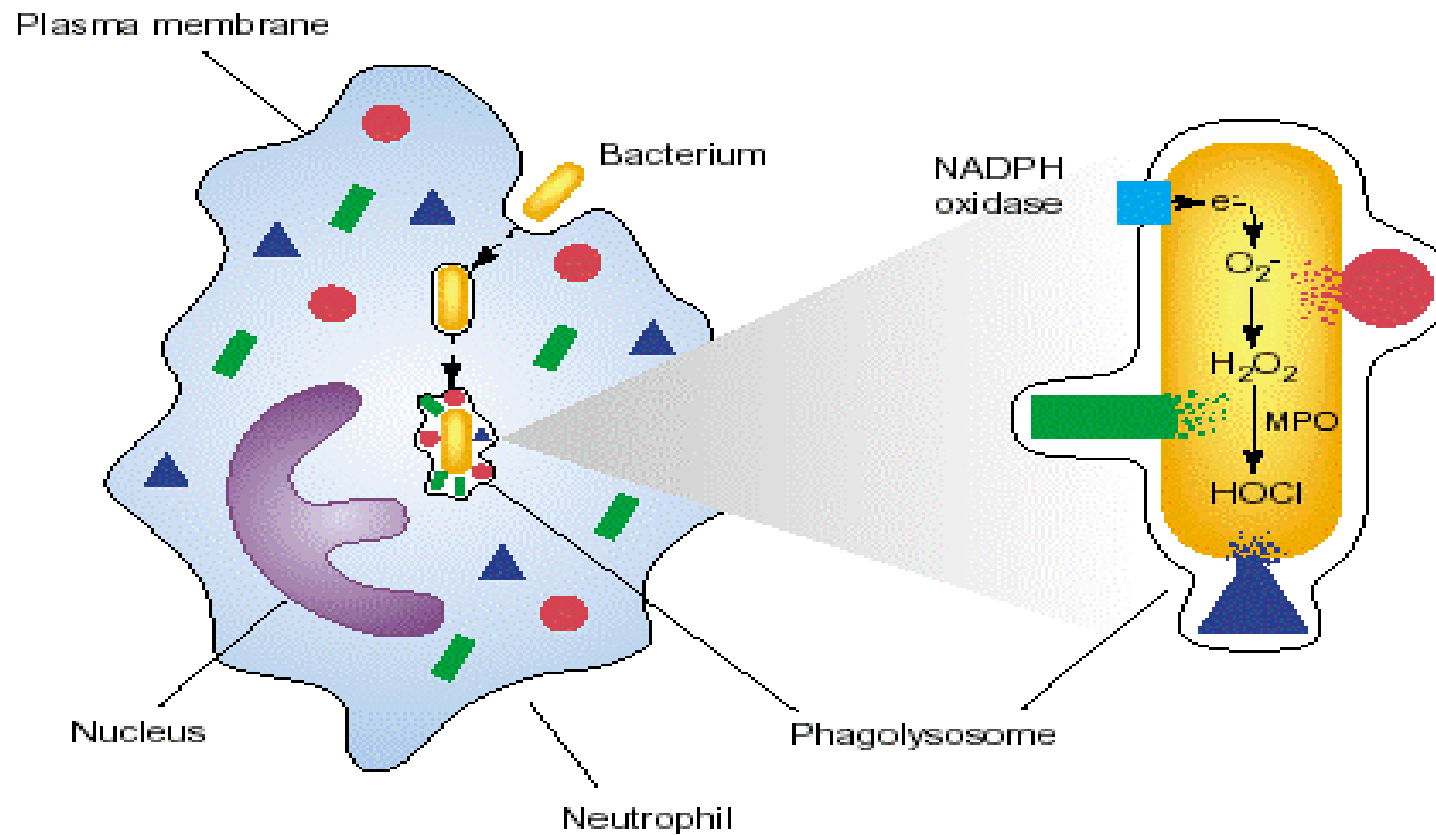
- Neutrophils and monocytes are armed with both **oxygen-independent** and **oxygen-dependent** mechanisms for killing bacteria.

- **oxygen-independent** - lysosomes
- **oxygen-dependent- NADPH OXIDASE & MYELOPEROXIDASE**

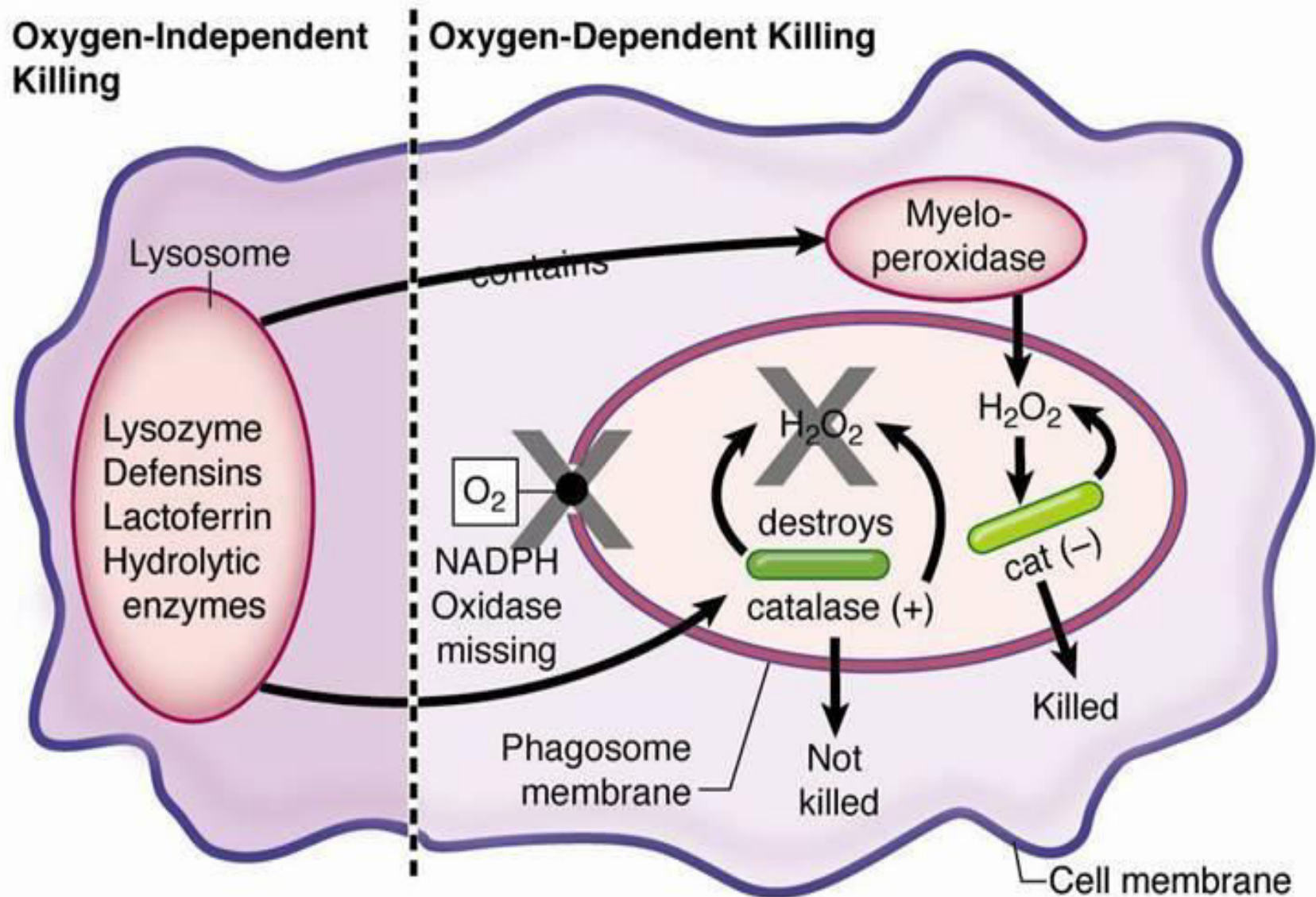
- After internalization of the microorganism has occurred, NADPH oxidase, located in the leukocyte cell membrane, converts molecular oxygen from the surrounding tissue into superoxide.

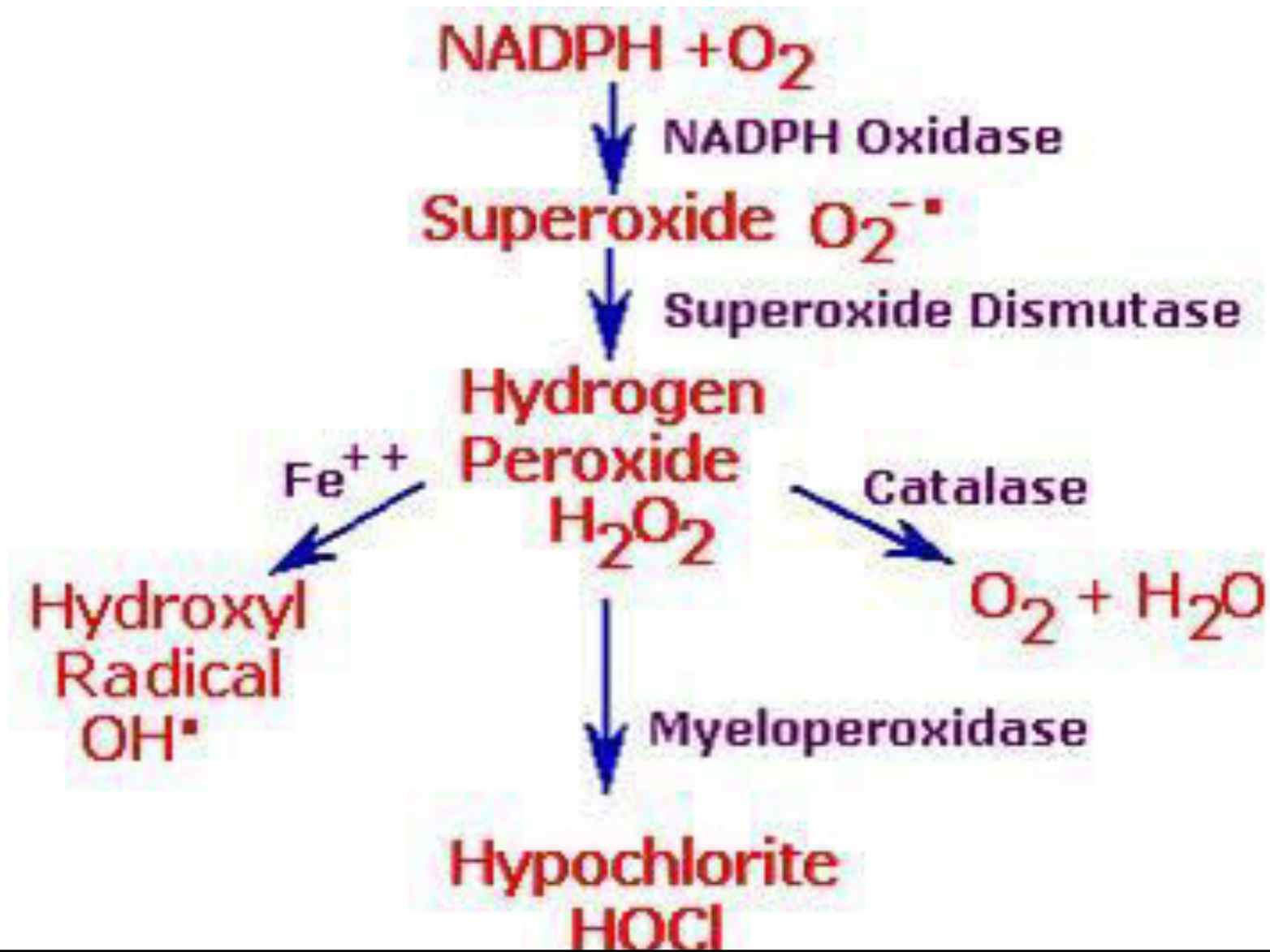
- The rapid consumption of molecular oxygen that accompanies formation of superoxide is referred to as the respiratory burst

- Genetic deficiencies in NADPH oxidase cause chronic granulomatous disease characterized by severe, persistent infections. and the formation of granulomas (nodular areas of inflammation) that sequester the bacteria that were not destroyed.



- Azurophilic granules
e.g.: CG, NE, defensins, MPO, BPI, lysozyme
- Specific granules
e.g.: hCAP-18, lactoferrin, lysozyme
- ▲ Gelatinase granules
e.g.: lysozyme, acetyltransferase, gelatinase





Synthesis of nitric oxide

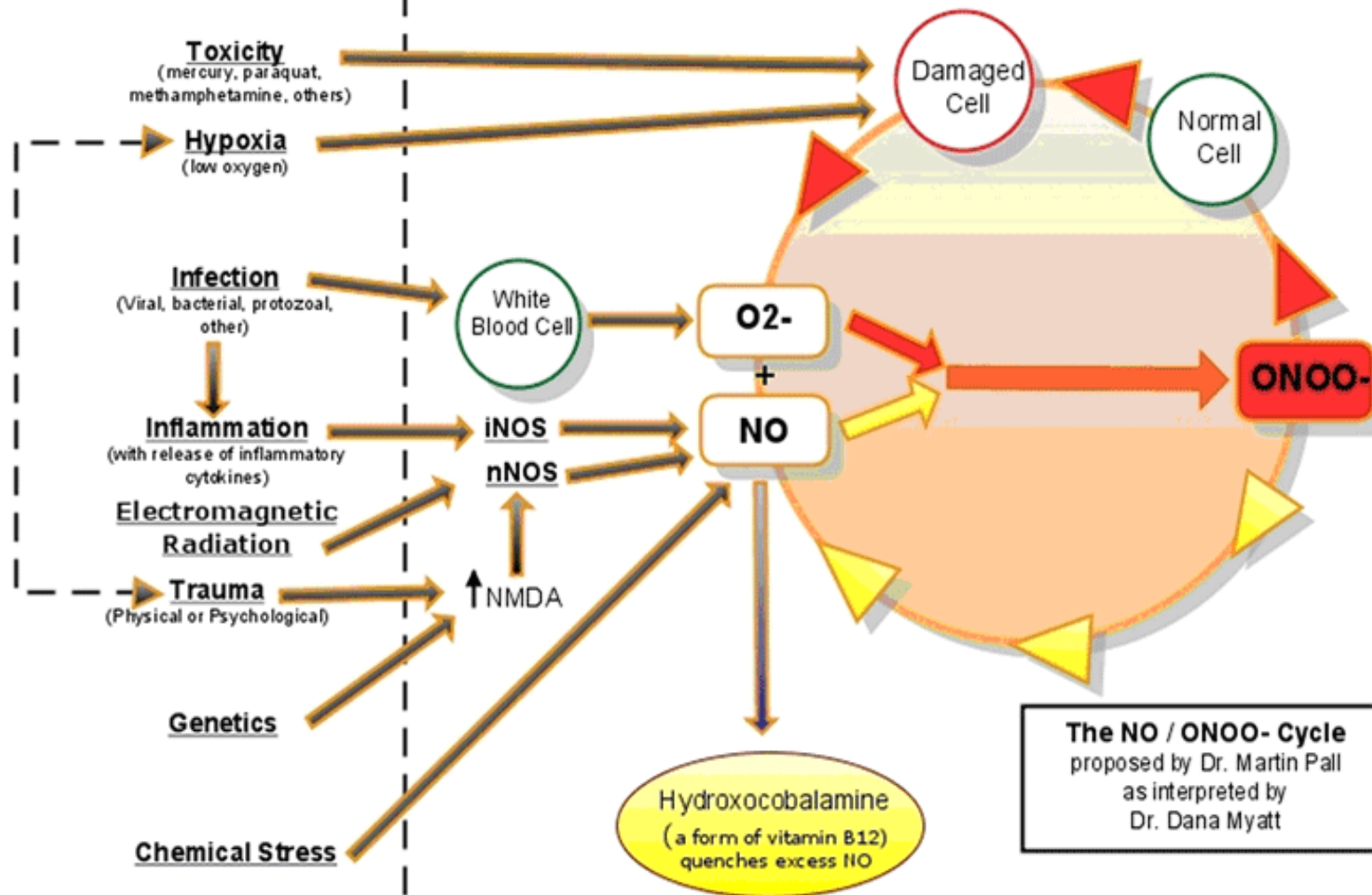
- Synthesis of NO
- Actions of NO on vascular endothelium
- Role of NO in mediating macrophage bactericidal activity

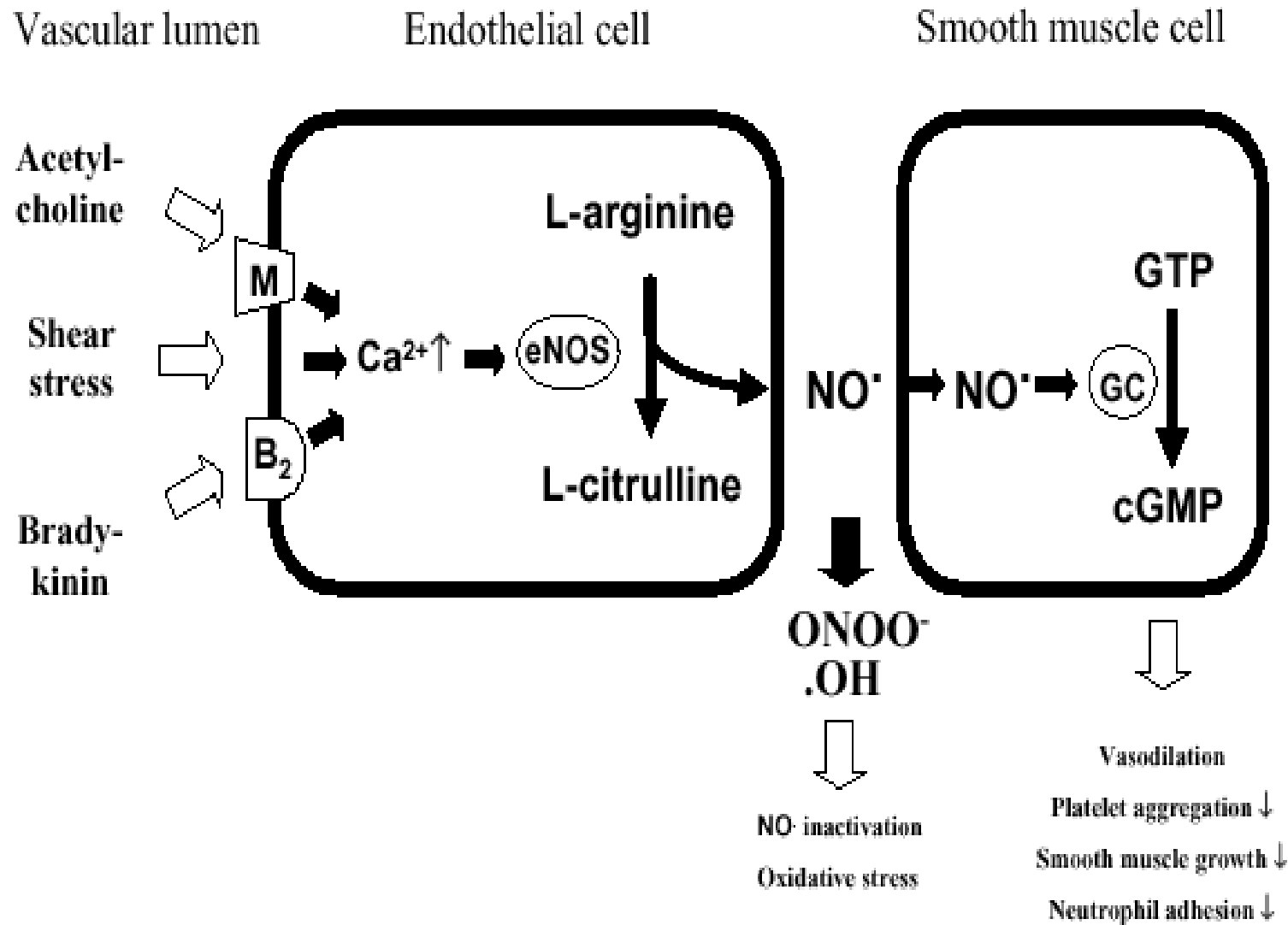
- NO is synthesized by eNOS in endothelial cells, and diffuses to vascular smooth muscle, where it activates the cytosolic form of guanylate cyclase (also known as guanylyl cyclase). [Note: This reaction is analogous to the formation of cAMP by adenylate cyclase (see p. 94), except that this guanylate cyclase is not membrane-associated.]

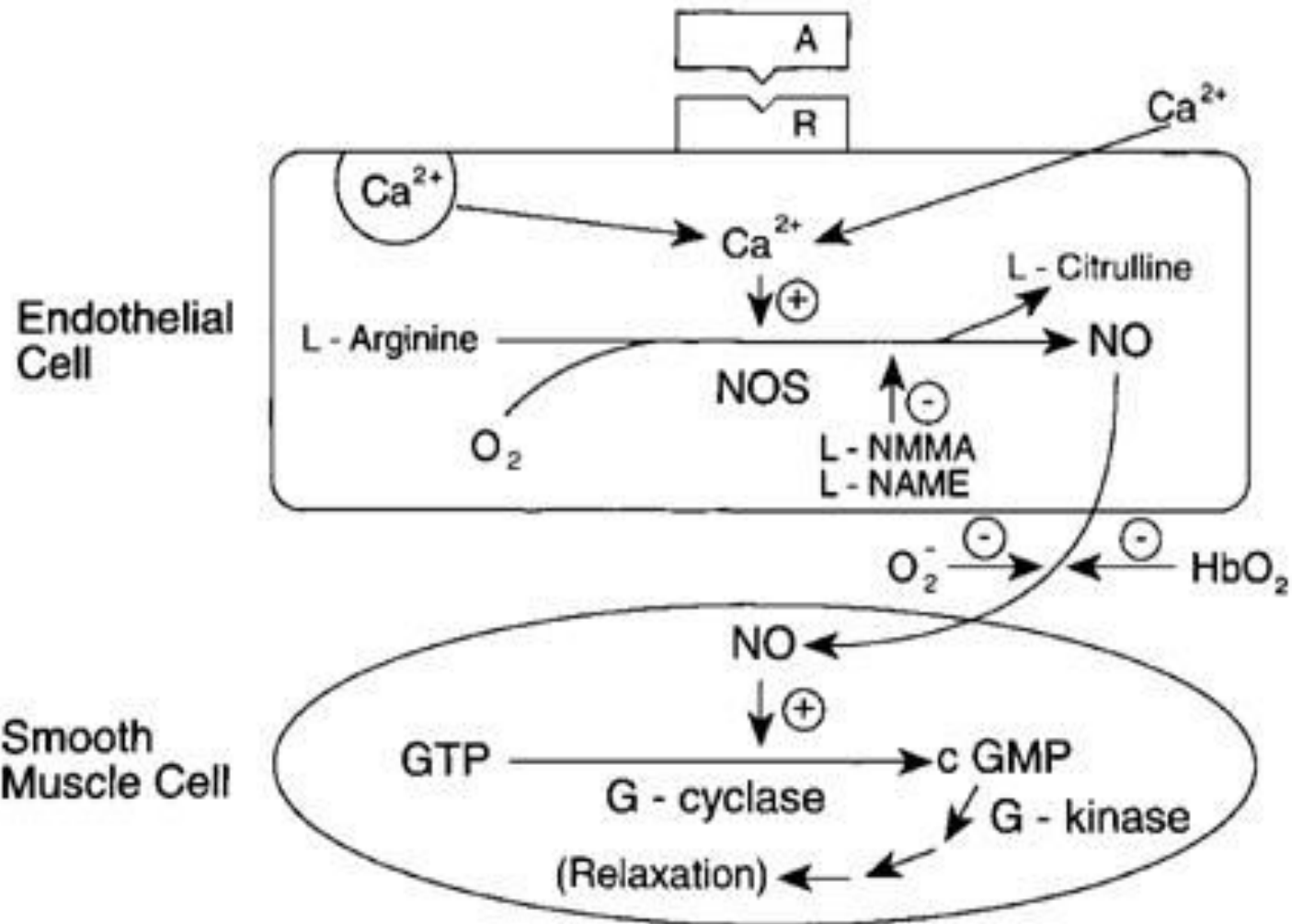
- The resultant rise in cGMP causes activation of protein kinase G, which phosphorylates Ca^{2+} channels, causing decreased entry of Ca^{2+} into smooth muscle cells. This decreases the calcium-calmodulin activation of myosin light-chain kinase, thereby decreasing smooth muscle contraction and favoring relaxation.

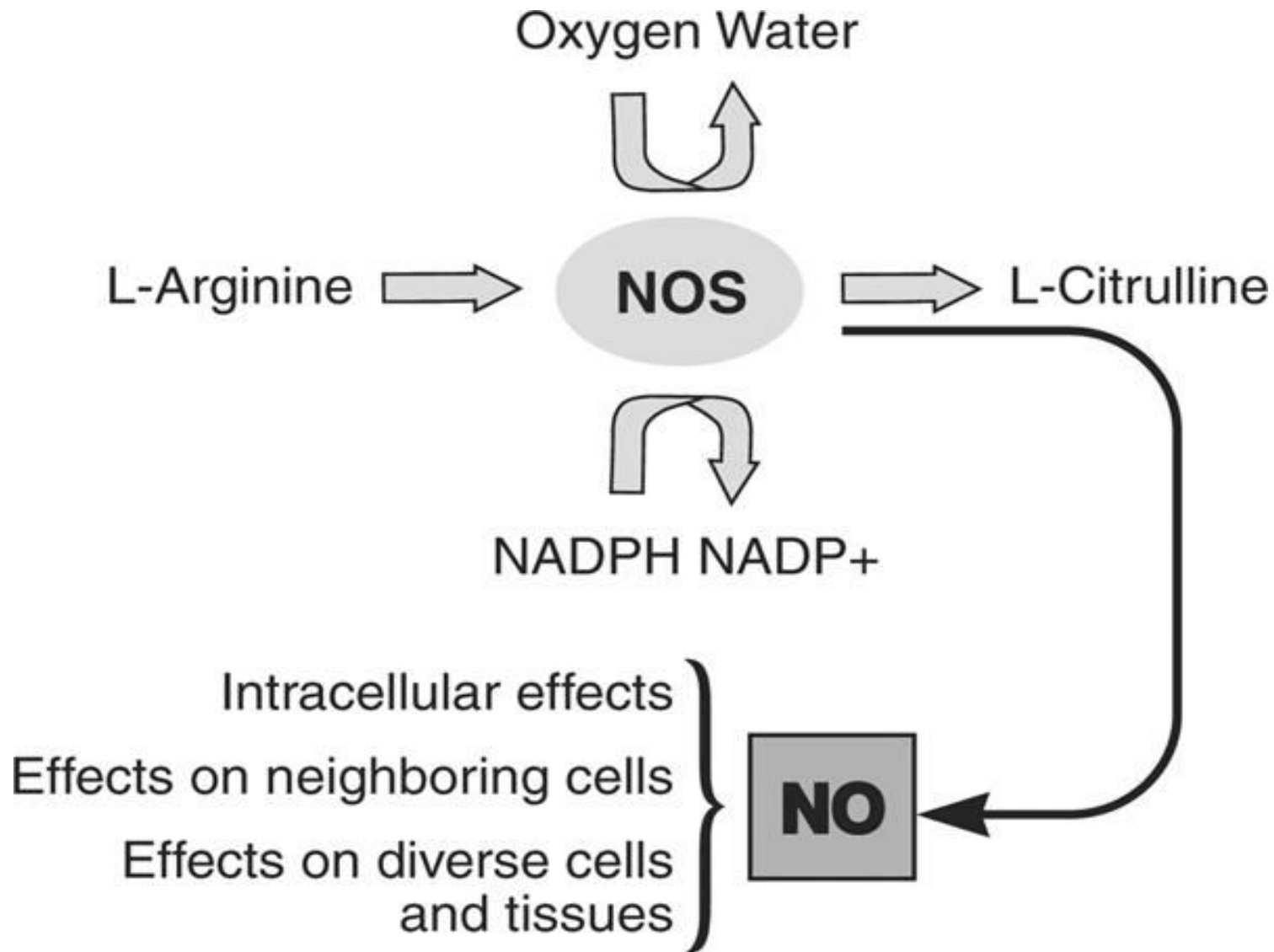
- Vasodilator nitrates, such as nitroglycerin and nitroprusside, are metabolized to nitric oxide, which causes relaxation of vascular smooth muscle and, therefore, lowers blood pressure.

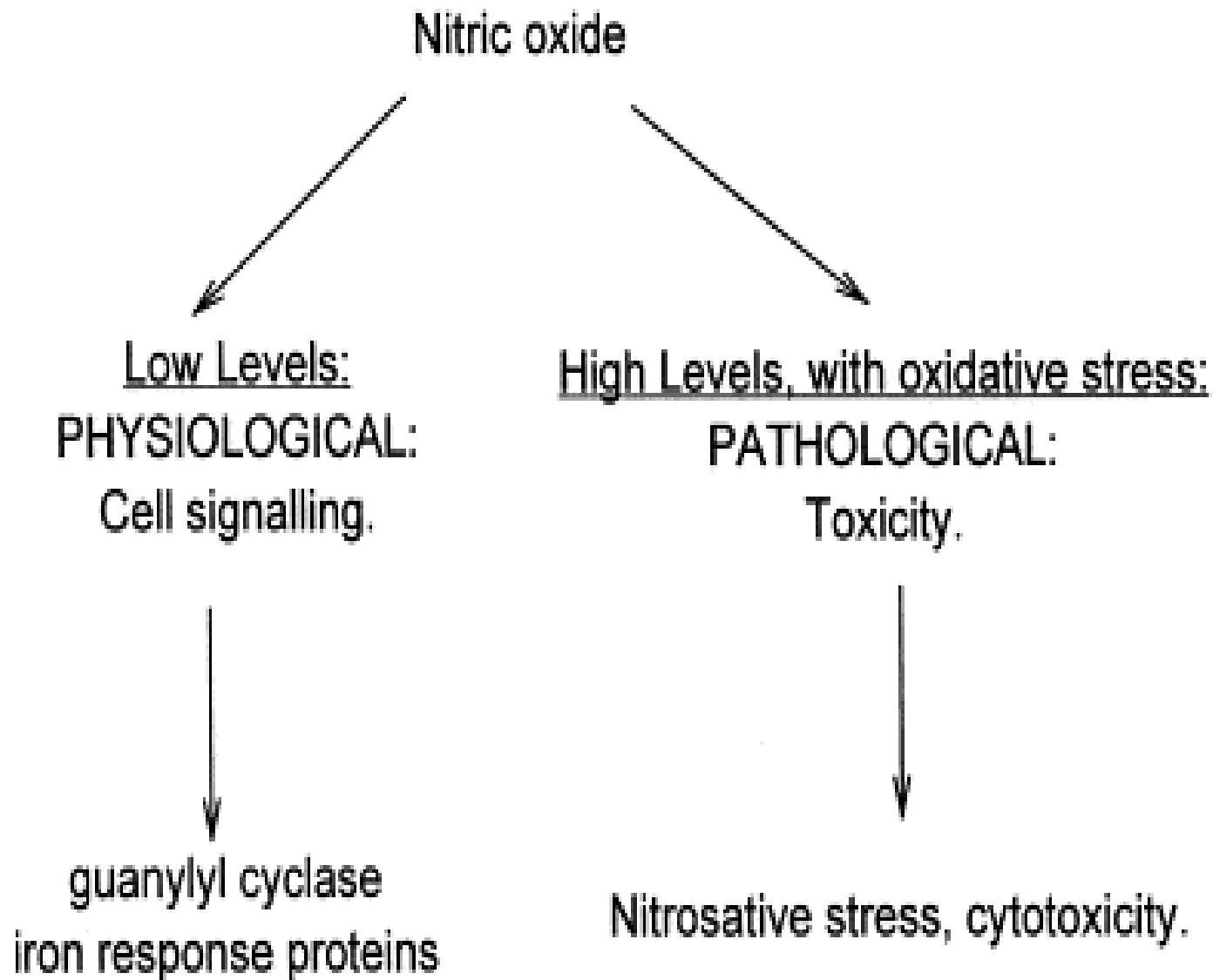
Observed "Initiators" of ME/CFS/FM and Related Multi-System Diseases











Glucose 6-P Dehydrogenase Deficiency

- Diminished G6PD activity impairs the ability of the cell to form the NADPH that is essential for the maintenance of the reduced glutathione pool. This results in a decrease in the cellular detoxification of free radicals and peroxides formed within the cell

- Glutathione also helps maintain the **reduced states of sulfhydryl groups in proteins**, including hemoglobin.
- **Oxidation** of those sulfhydryl groups leads to the formation of **denatured proteins** that form insoluble masses (called **Heinz bodies**) that attach to the red cell membranes

Precipitating factors in G6PD deficiency

- Oxidant drugs (Antibiotics (for example, sulfamethoxazole and chloramphenicol), Antimalarials (for example, primaquine but not quinine), and Antipyretics (for example, acetanilid but not acetaminophen)).
- Favism
- Infection
- Neonatal jaundice

- Class I- very severe- $<10\%$ (residual enzyme activity)
- Class II – severe - $< 10\%$
- Class III – moderate – 10- 60%
- Class IV – none - $> 60\%$

- **G6PD A⁻** is the prototype of the moderate (**Class III**) form of the disease. The red cells contain an **unstable but kinetically normal** G6PD, with most of the enzyme activity present in the reticulocytes and younger erythrocytes

- **G6PD Mediterranean** is the prototype of a more severe (**Class II**) deficiency in which the enzyme shows **normal stability** but **scarcely detectable activity** in all red blood cells

Molecular biology of G6PD

- Most of these DNA changes are **missense, point mutations**
- Both G6PD A⁻ and G6PD Mediterranean represent mutant enzymes that differ from the respective normal variants by a single amino acid.

- Large deletions or frameshift mutations have not been identified, suggesting that complete absence of G6PD activity is probably lethal.

Learning Resources

- Lippincott's Biochemistry
- Harper's Biochemistry
- Teacher Notes