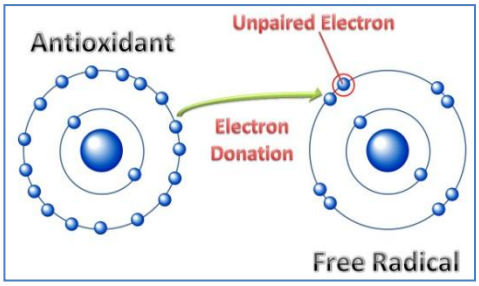
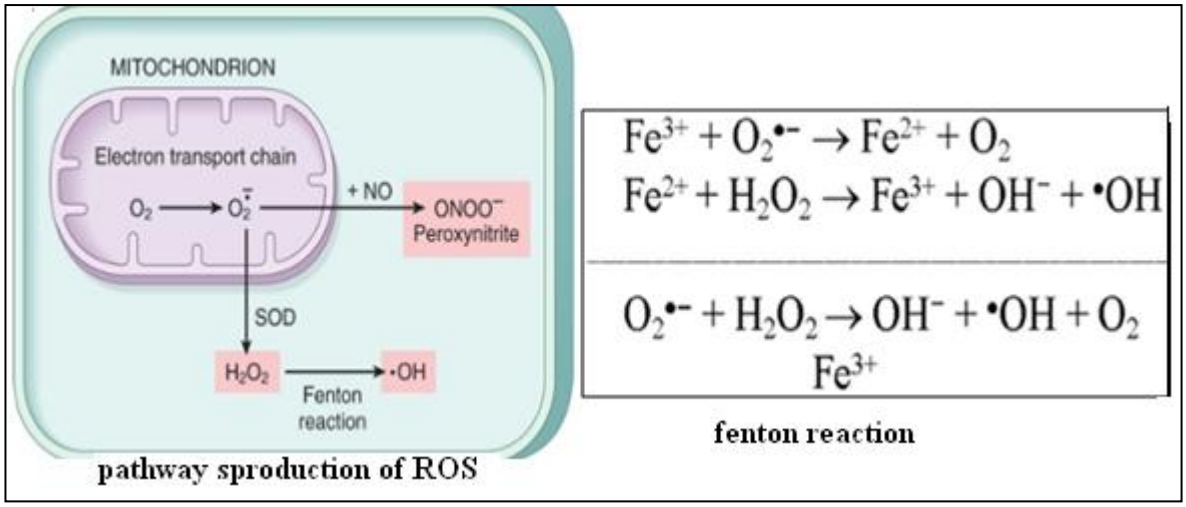
# Accumulation of Oxygen-Derived Free Radicals (Oxidative Stress)

**Free radicals are chemical species with a single unpaired electron in an outer orbital. Free radicals readily react with inorganic and organic chemicals; when generated in cells, they avidly attack nucleic acids as well as a variety of cellular proteins and lipids.**

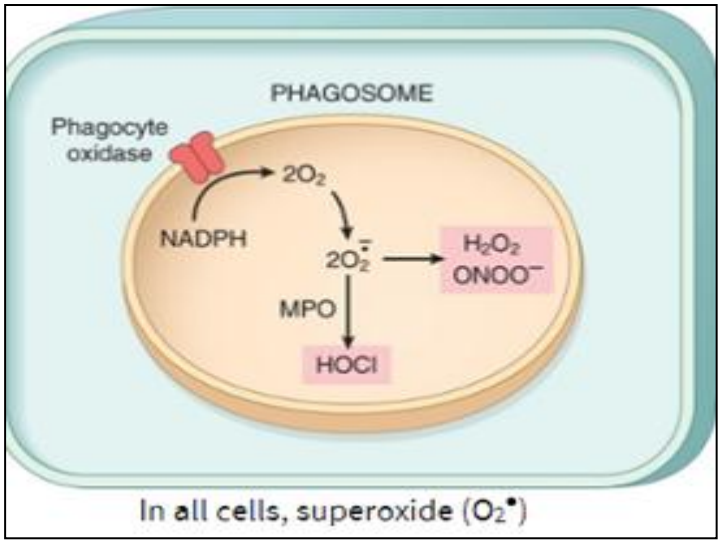



**<u>Reactive oxygen species (ROS)</u> are a type of oxygen-derived free radical whose role in cell injury is well established. Cell injury in many circumstances involves damage by free radicals; these situations include:- 1- ischemia-reperfusion 2-chemical 3-radiation injury4- toxicity from oxygen 5- gases 6-cellular aging microbial killing by phagocytic cells, and tissue injury caused by inflammatory cells.**

**Figure 1–18 Pathways of production of reactive oxygen species. A, In all cells, superoxide ($O_2^{\bullet}$) is… ROS are produced normally in small amounts in all cells during the reduction-oxidation (redox) reactions that occur during mitochondrial respiration and energy generation. In this process, molecular oxygen is sequentially reduced in mitochondria by the addition of four electrons to generate water. This reaction is imperfect, however, and small amounts of highly reactive but short-lived toxic intermediates are generated when oxygen is only partially reduced. These intermediates include superoxide , which is converted to hydrogen peroxide ($H_2O_2$) spontaneously and by the action of the enzyme superoxide dismutase. $H_2O_2$ is more stable than and can cross biologic membranes. In the presence of metals, such as $Fe^{2+}$, $H_2O_2$ is converted to the highly reactive hydroxyl radical $^{\bullet}OH$ by the Fenton reaction.**




**ROS are produced in phagocytic leukocytes, mainly neutrophils and macrophages, as a weapon for destroying ingested microbes and other substances during inflammation and host defense .**

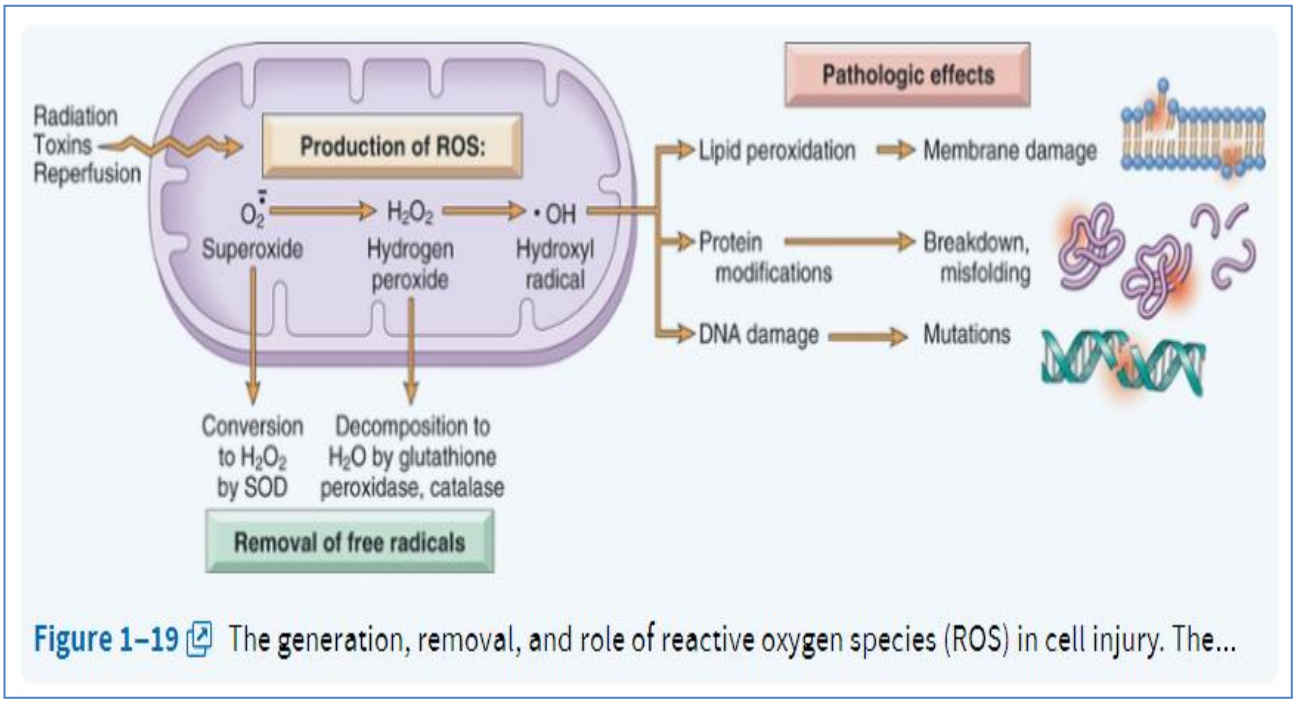


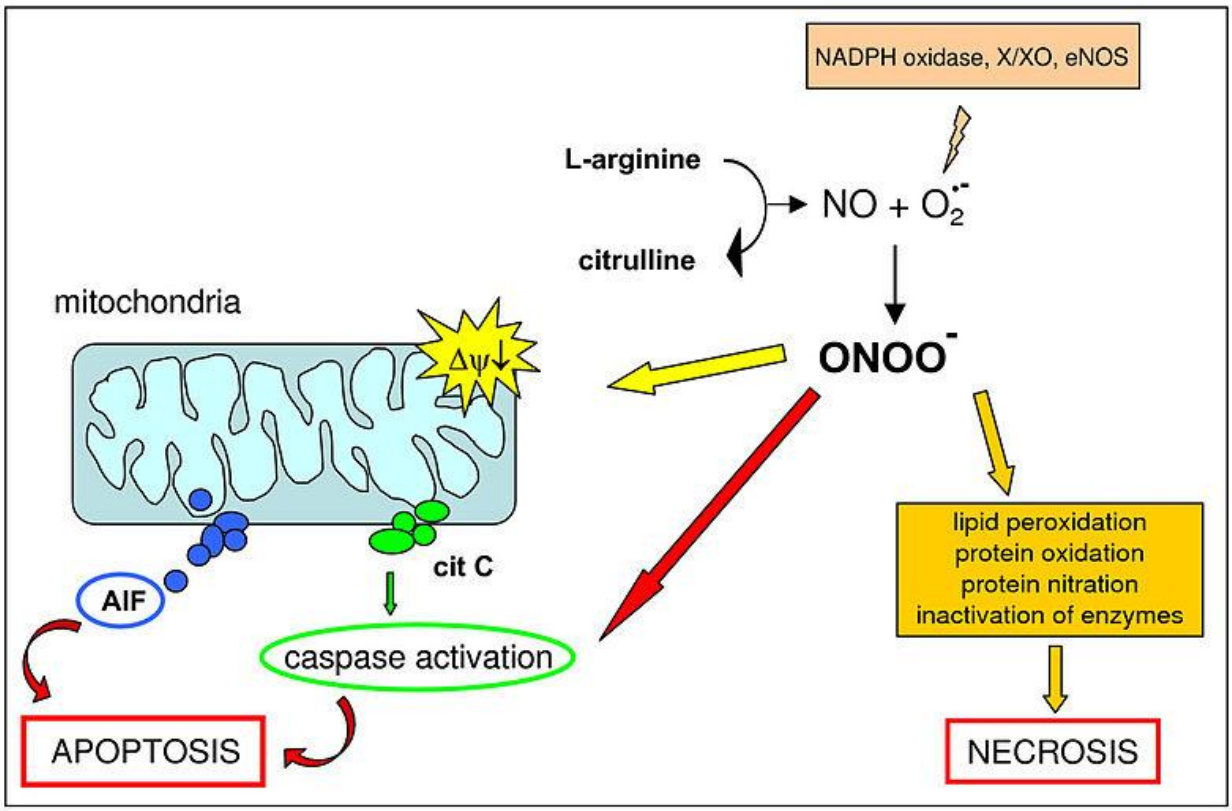

**The ROS are generated in the phagosomes and phagolysosomes of leukocytes by a process that is similar to mitochondrial respiration and is called the respiratory burst (or oxidative burst). In this process, a phagosome membrane enzyme catalyzes the generation of superoxide, which is converted to $H_2O_2$. $H_2O_2$ is in turn converted to a highly reactive compound hypochlorite (the major component of household bleach) by the enzyme myeloperoxidase, which is present in leukocytes.**

**Nitric oxide (NO):-** **is another reactive free radical produced in leukocytes and other cells. It can react with to form a highly reactive compound, peroxynitrite, which also participates in cell injury.**

**The damage caused by free radicals is determined by their rates of production and removal (Fig. 1–19). When the production of ROS increases or the scavenging systems are ineffective, the result is an excess of these free radicals, leading to a condition called oxidative stress.**

**Figure 1–19 The generation, removal, and role of reactive oxygen species (ROS) in cell injury.**






**Reactions of peroxynitrite leading to either apoptotic or necrotic cell death.**

**The generation of free radicals is increased under several condition :**

**1-The absorption of radiant energy (e.g., ultraviolet light, x-rays). Ionizing radiation can hydrolyze water into hydroxyl ($^{\bullet}$OH) and hydrogen (H$^{\bullet}$) free radicals.**

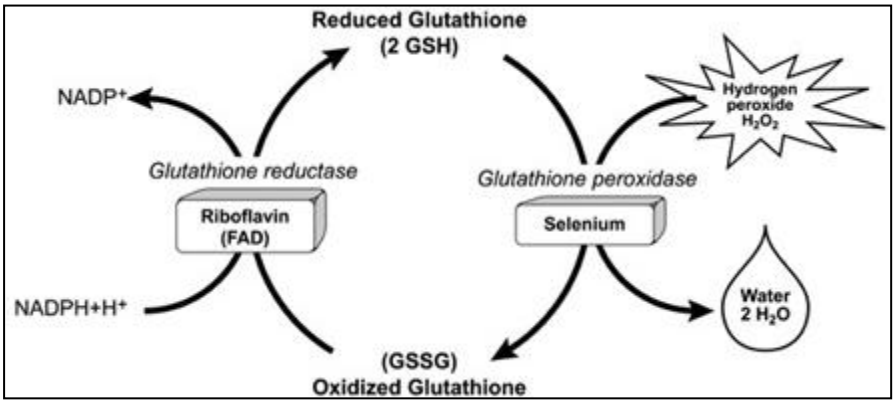
**2-The enzymatic metabolism of exogenous chemicals (e.g., carbon tetrachloride)**

**3-Inflammation, in which free radicals are produced by leukocytes**

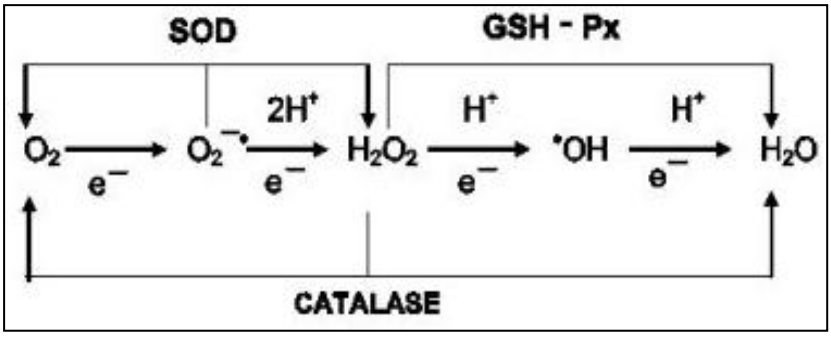
**Cells have developed many mechanisms to remove free radicals and thereby minimize injury. Free radicals are inherently unstable and decay spontaneously. There are also non enzymatic and enzymatic systems that contribute to inactivation of free radicals .**

**1-The rate of decay of superoxide is significantly increased by the action of superoxide dismutases (SODs) found in many cell types.**

**2-Glutathione (GSH) peroxidases are a family of enzymes whose major function is to protect cells from oxidative damage. The most abundant member of this family, glutathione peroxidase 1, is found in the cytoplasm of all cells. It catalyzes the breakdown of $H_2O_2$ by the reaction 2 GSH (glutathione) + $H_2O_2$ → GS-SG + 2 $H_2O$. The intracellular ratio of oxidized glutathione (GSSG) to reduced glutathione (GSH) is a reflection of this enzyme's activity and thus of the cell's ability to catabolize free radicals.**

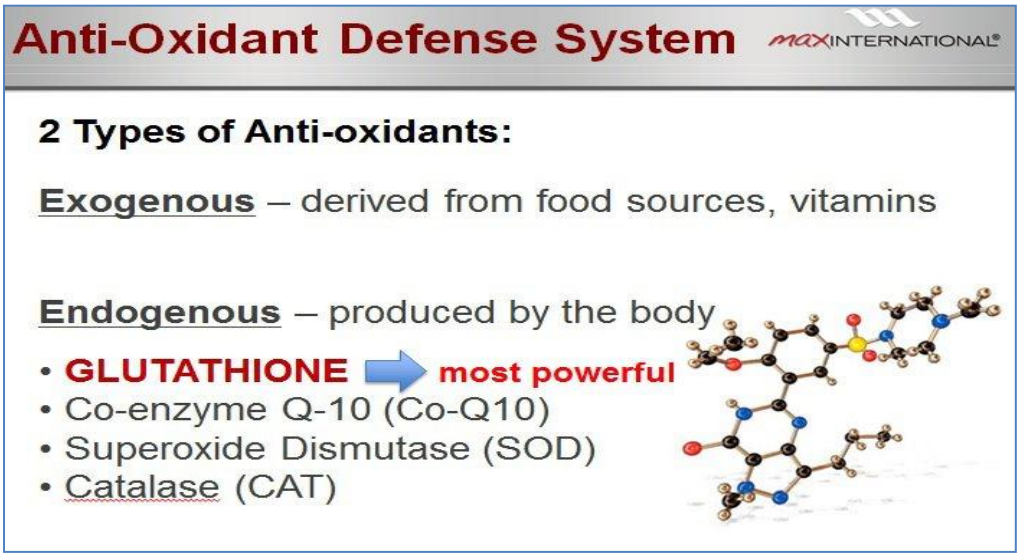



**3- Catalase, present in peroxisomes, catalyzes the decomposition of hydrogen peroxide ($2H_2O_2 \rightarrow O_2 + 2H_2O$). It is one of the most active enzymes known, capable of degrading millions of molecules of $H_2O_2$ per second.**



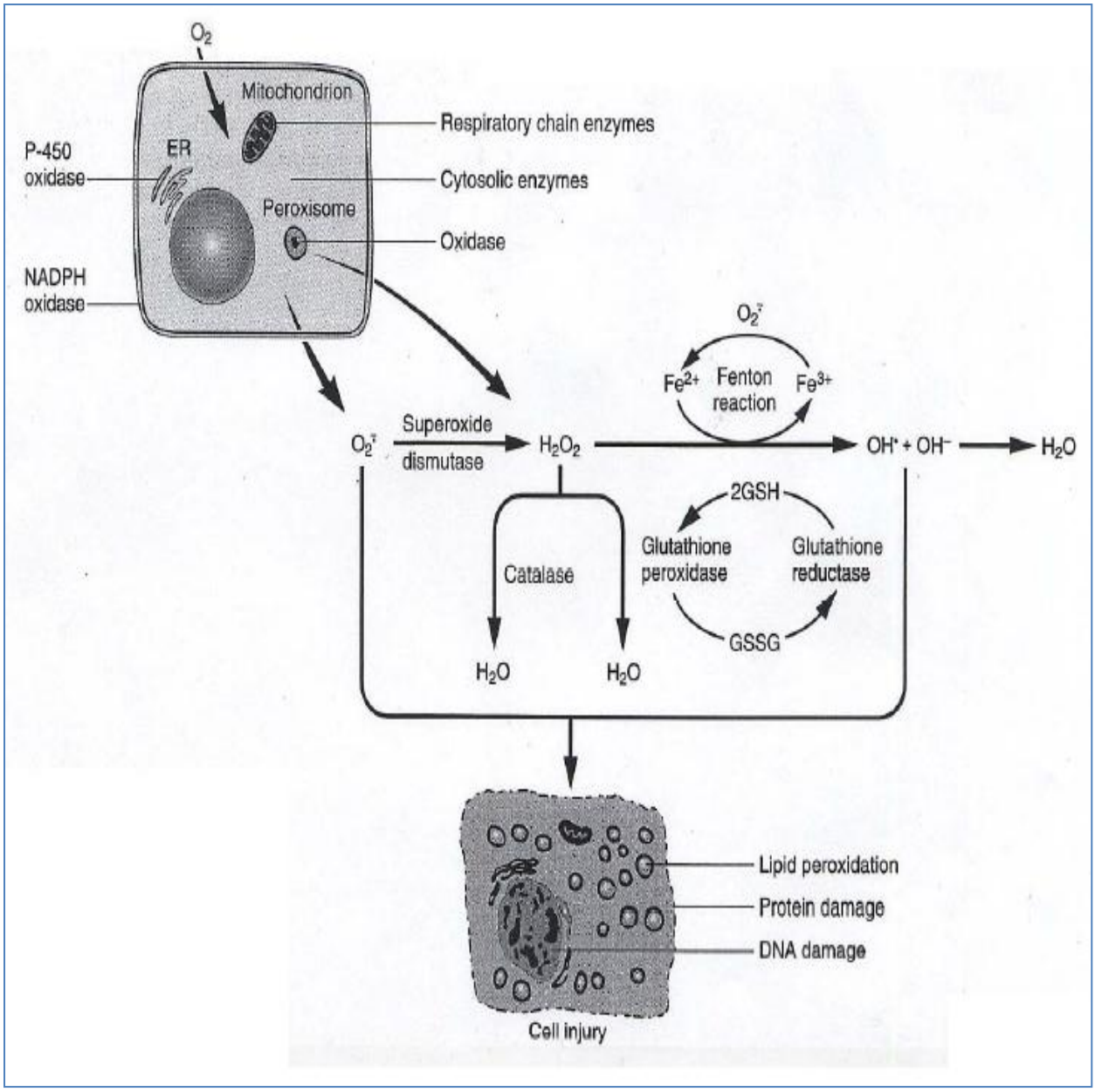

**Catalase, present in peroxisomes,**

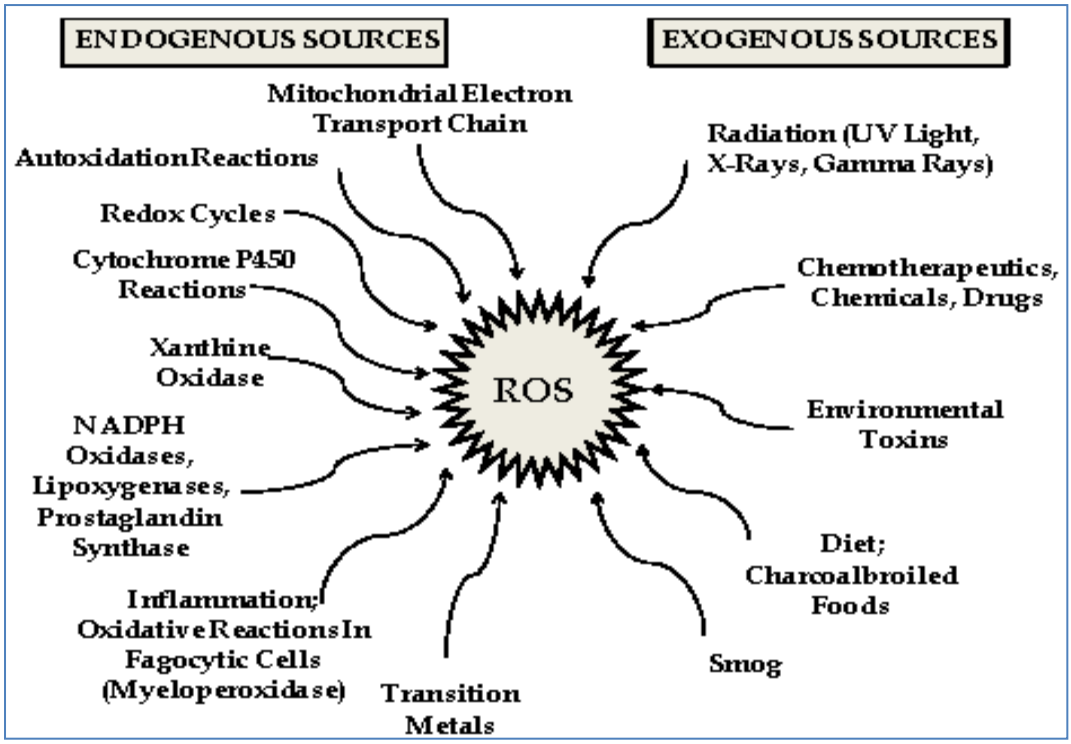
**4-Endogenous or exogenous antioxidants (e.g., vitamins E, A, and C and β-carotene) may either block the formation of free radicals or scavenge them once they have formed.**




**Reactive oxygen species cause cell injury by three main reactions (Fig. 1–19):**

- **Lipid peroxidation of membranes. Double bonds in membrane polyunsaturated lipids are vulnerable to attack by oxygen-derived free radicals. The lipid–radical interactions yield peroxides, which are themselves unstable and reactive, and an autocatalytic chain reaction ensues.**
- **Cross-linking and other changes in proteins. Free radicals promote sulfhydryl-mediated protein cross-linking, resulting in enhanced degradation or loss of enzymatic activity. Free radical reactions may also directly cause polypeptide fragmentation.**
- **DNA damage. Free radical reactions with thymine in nuclear and mitochondrial DNA produce single-strand breaks. Such DNA damage has been implicated in cell death, aging, and malignant transformation of cells.**


$O_2$
Mitochondrion
Respiratory chain enzymes
P-450 oxidase
ER
Cytosolic enzymes
Peroxisome
Oxidase
NADPH oxidase
$O_2^-$
$Fe^{2+}$
Fenton reaction
$Fe^{3+}$
$O_2^-$
Superoxide dismutase
$H_2O_2$
$OH^\bullet + OH^-$
$H_2O$
2GSH
Glutathione peroxidase
Glutathione reductase
Catalase
GSSG
$H_2O$
$H_2O$
Lipid peroxidation
Protein damage
DNA damage
Cell injury




**<u>Defects in Membrane Permeability:-</u>** **Figure 1–20 Mechanisms of membrane damage in cell injury. Decreased $O_2$ and increased cytosolic…**

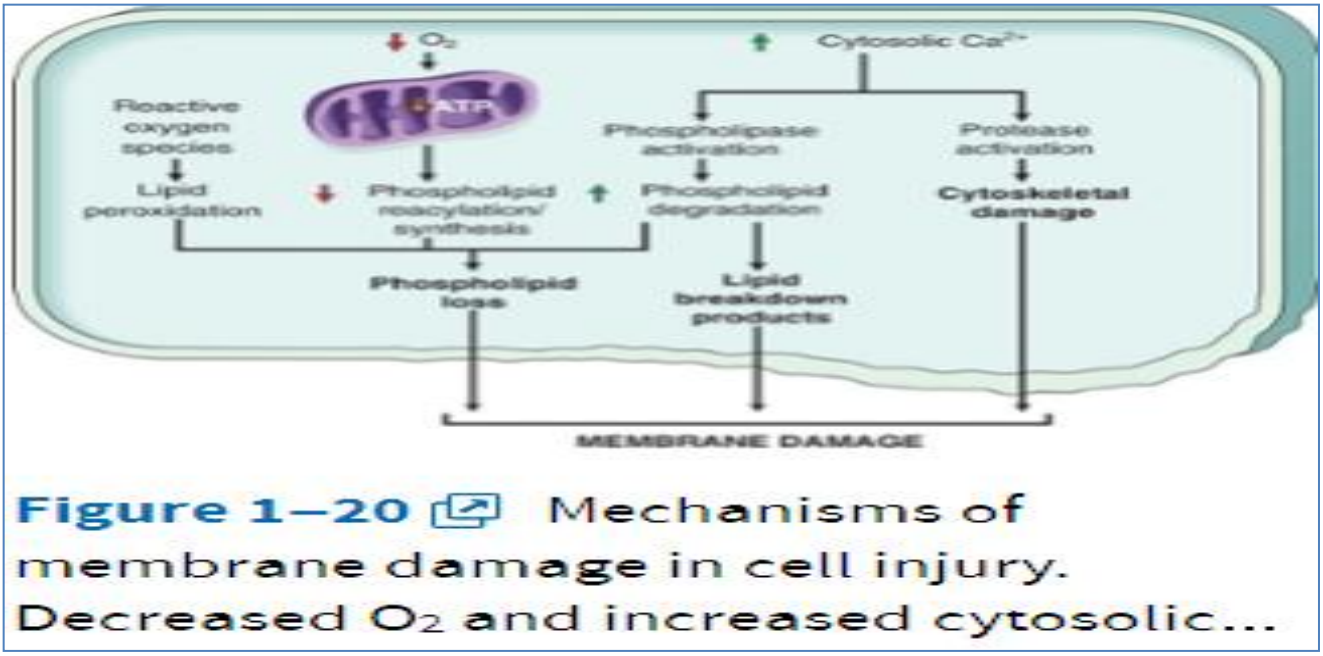



**Figure 1–20** Mechanisms of membrane damage in cell injury. Decreased $O_2$ and increased cytosolic…

**Summary:- Mechanisms of Cell Injury**

**1-ATP depletion: failure of energy-dependent functions → reversible injury → necrosis**

**2-Mitochondrial damage: ATP depletion → failure of energy-dependent cellular functions → ultimately, necrosis; under some conditions, leakage of mitochondrial proteins that cause apoptosis**

**3-Influx of calcium: activation of enzymes that damage cellular components and may also trigger apoptosis**

**4-Accumulation of reactive oxygen species: covalent modification of cellular proteins, lipids, nucleic acids**

**5-Increased permeability of cellular membranes: may affect plasma membrane, lysosomal membranes, mitochondrial membranes; typically culminates in necrosis**

**6-Accumulation of damaged DNA and misfolded proteins: triggers apoptosis**