Chemical and Biochemical Principles of Toxicology

Role of metals in toxicity

- Some metals interfere with the biological function of some electrolytes that have the same electronic valence, such as :

- Li + mimic K+ , thus it interferes with cellular K+ homeostasis and alters neuronal repolarization

- Pb ++ mimic Ca++ , thus it interferes with Ca++ deposition in bone forming lead phosphate instead of calcium phosphate

- Ti + mimic K+

- As +5 mimic phosphor and interferes with formation of ATP

Transition metals and toxicity

- Transition metals are metals present in the body acting as elements forming a positive ion in their oxidative states

- They can accept electrons from any donors, so they have an important role in redox reaction (oxidation – reduction reaction)

- They participate in the generation of free radicals through Fenton and Haber – Weiss reactions

Oxidation – Reduction Reaction (Redox Reaction)

It involves the movement of electrons from one atom or molecule to another, and actually comprise two dependent reactions : reduction and oxidation

Redox chemistry is important in medical toxicology :

Ferrous (Fe 2+) to ferric (Fe 3+) iron in hemoglobin molecule converting it to methemoglobin molecule

Lead and mercury after oxidization are converted from elemental harmless state to ionic toxic state

Conversion of ethanol to acetaldehyde by alcohol dehydrogenase enzyme

(The electron delivered to oxidized nicotinamide adenine dinucleotide (NAD +), reducing it to NADH

Reactive Species *(Free Radicals)*

- Reactive molecules that contain one or more unpaired electrons
- Reactive species can be anions , cations , or neutrals
- Reactive species can receive electron from other molecules to fill their empty orbital in order to be stable
- Reactive species can be :

- Reactive oxygen species (ROS) *(the most important)*
- Reactive nitrogen species (RNS)

Important reactive species

ROS		RNS	
Super oxide radical	$O_2^{\bullet -}$	Nitric oxide	$NO^{\bullet}$
Hydroxyl radical	$OH^{\bullet}$	Nitrogen dioxide	$NO_2^{\bullet}$
Alcoxyl radical	$RO^{\bullet}$	Peroxynitrite anion	$ONOO^-$
Peroxyl radical	$ROO^{\bullet}$	Nitronium ion	NO_2^+
Hydrogen peroxide	H_2O_2		
Ozone	O_3		

Hypochlorous acid HOCl

Singlet oxygen [O]

- ROS generated by two main ways :

- Endogenously in the body by different mechanisms
- Exogenously by xenobiotics

Formation of hydroxyl radical (OH•)

Steps include :

The addition of an electron to O2 to create the superoxide ion

Superoxide combines with hydrogen and another electron to produce hydrogen peroxide

Hydrogen peroxide undergoes various reactions to produce the hydroxyl radical in the presence of metal ion such as iron

Steps of OH• generation

- $O_2 + e^-$ $O_2\bullet^-$
- $O_2\bullet^- + 2H^+ + e^-$ H_2O_2
- H_2O_2 / Fe 2+ OH^- + OH • *(Fenton reaction)*
- H_2O_2 + $O_2\bullet^-$ / Fe 2+ O_2 + OH^- + OH • *(Haber – Weiss)*

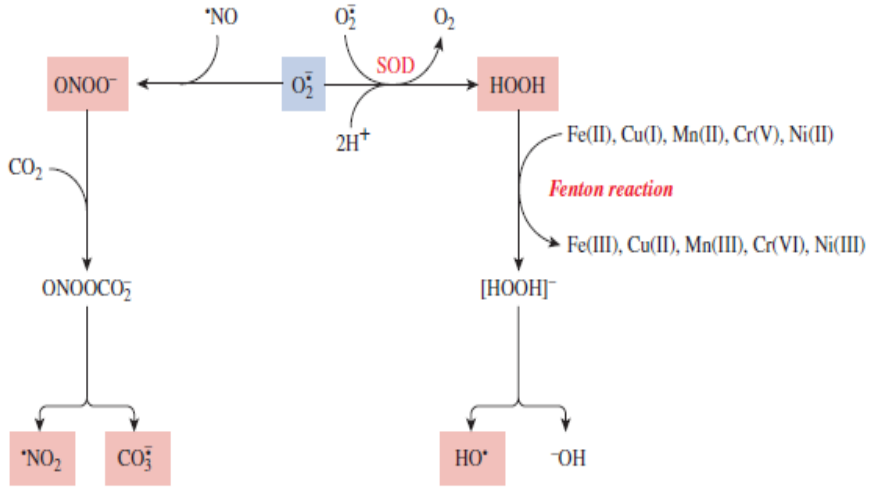


Figure 3-4. *Two pathways for toxicity of superoxide anion radical $O_2^{\bullet-}$ from conversion of ONOO⁻ and HOOH (nonradical products) to $•NO_2$, $CO_3^{\bullet-}$, and HO• (radicals).* Conversion of $O_2^{\bullet-}$ to HOOH is spontaneous or is catalyzed by SOD. Homolytic cleavage of HOOH to hydroxyl radical and hydroxyl ion (Fenton reaction) is catalyzed by the transition metal ions shown. Hydroxyl radical is the ultimate toxicant for xenobiotics that form $O_2^{\bullet-}$. In the other pathway, $O_2^{\bullet-}$ reacts avidly with nitric oxide (•NO), the product of •NO synthase (NOS), forming peroxynitrite ($ONOO^-$). Spontaneous reaction of $ONOO^-$ with carbon dioxide (CO_2) yields nitrosoperoxy carbonate that with homolytic cleavage forms nitrogen dioxide ($•NO_2$) and carbonate anion radical ($CO_3^{\bullet-}$). All three radical products indicated in this figure are oxidants, whereas $•NO_2$ is also a nitrating agent.

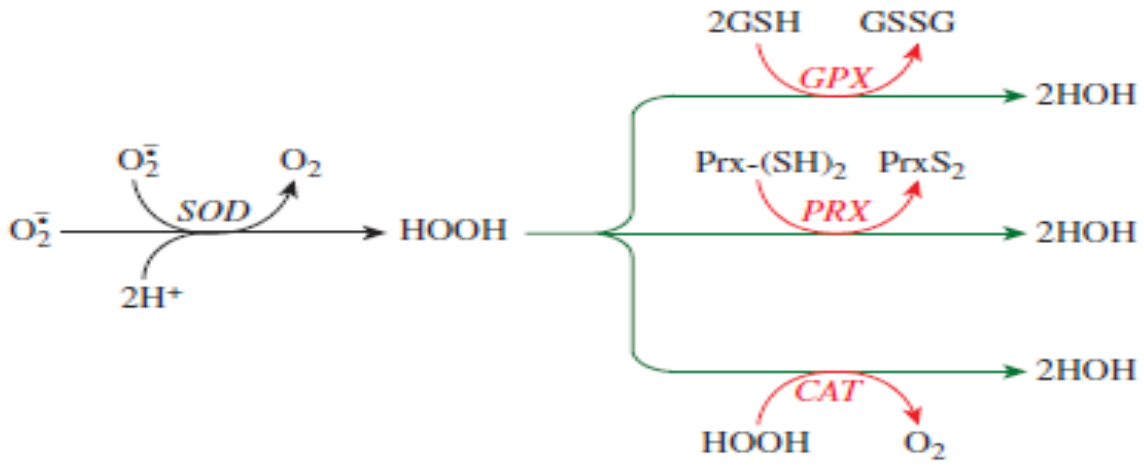


Figure 3-5. *Detoxification of superoxide anion radical (O_2^-) and HOOH.* Superoxide dismutase (SOD) converts O_2^- to HOOH, which is further detoxified by glutathione peroxidase (GPX), peroxiredoxin (Prx-$(SH)_2$), and catalase (CAT). When GPX reduces HOOH, it forms glutathione disulfide (GSSG), which is reduced back to GSH by glutathione reductase (requires NADPH; see Fig. 3-6). When Prx-$(SH)_2$ reduces HOOH, its catalytic thiol group (–R–S–H) is oxidized to a sulfenic acid group (–R–S–OH), which in turn reacts with another SH group of Prx, forming HOH and Prx disulfide ($PrxS_2$). Finally, catalase converts HOOH into two moles of water. Catalase is a high-capacity system, whereas Prx and GPX can be saturated at high concentrations of HOOH.

Effect of free radicals

Xenobiotic [R]

Free radical [R●]

Attack lipid membranes [LH]

Forming lipid peroxy radical [L●]

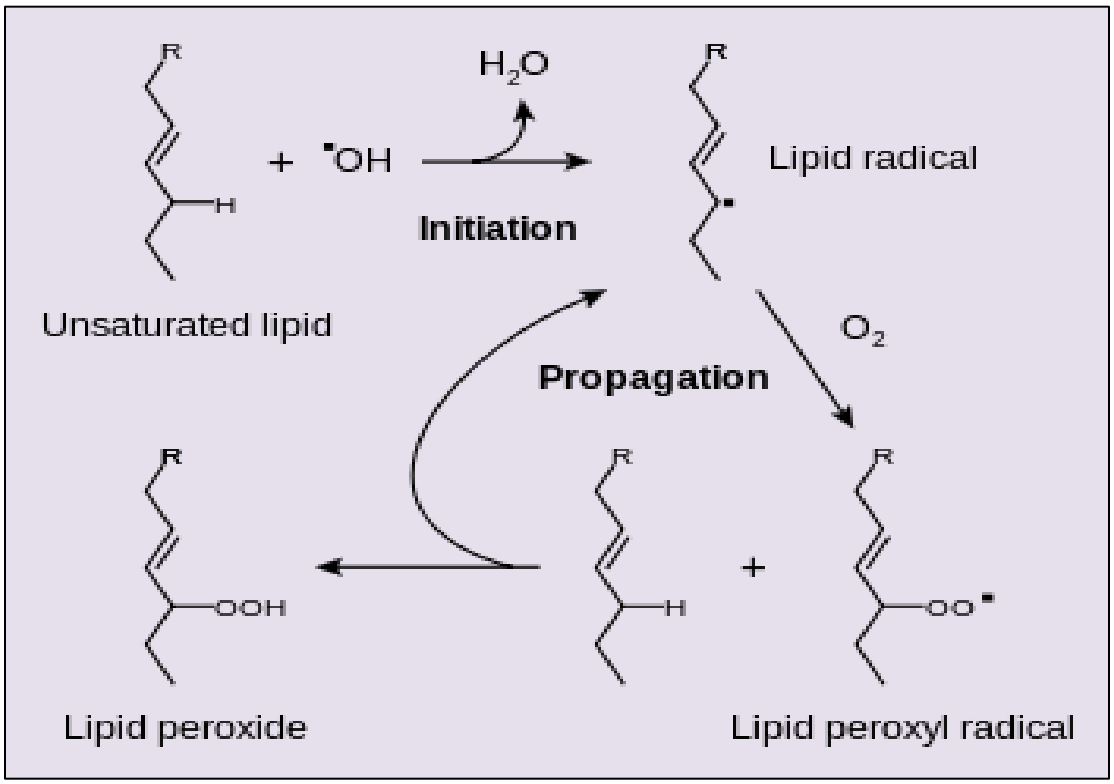
(Lipid peroxidation)

Oxidative Stress (lipid peroxidation)

- It occurs when formation of reactive oxidizing species overwhelm the defense mechanism

- Oxidative stress may result in oxidative damage to nucleic acids, proteins, and polyunsaturated fatty acids *(PUFA)* in cellular membranes, resulting in lipid peroxidation (the oxidative destruction of lipids).
- Attack of ROS to *PUFA* removes the particularly reactive hydrogen atom, with its lone electron, from a methylene carbon of a *PUFA;* leaving an unpaired electron and causing the formation of a lipid radical. This lipid radical attacks other *PUFA,* causing a chain reaction that destroys the cellular membrane. Membrane degradation products initiate inflammatory reactions in the cells, resulting in further damage

Lipid Peroxidation



Xenobiotics generating free radicals

- Many xenobiotics can be toxic by generation of free radicals (Redox Cycling) :

- Paraquat leading to pulmonary toxicity

- Doxorubicin leading to cardiotoxicity
- Zinc phosphide leading to multiorgan damage

Fate / Detoxification of free radicals

- Free radicals can be detoxified by many mechanisms :
- Superoxide radical converted to hydrogen peroxide via superoxide dismutase (SOD) which is rapidly followed by the transformation of hydrogen peroxide to water by glutathione peroxidase or catalase
- Transition metal ions can be sequestered and rendered harmless, such as :
- Iron binding with ferritin
- Copper binding with ceruloplasmin
- Cadmium binding with metallothionein

- Antioxidants such as glutathione act as reducing agent and nucleophile to prevent exogenous oxidants from producing hemolysis and the acetaminophen metabolite N – acetyl – p – benzoquinoneimine (NAPQI) from damaging the hepatocyte

Effect of xenobiotics on ATP production

- Effect of arsenic on glycolysis :
- Arsenic inhibits acetyl Co – A synthesis
- It replaces phosphate in many reactions
- It attacks the enzyme glyceraldehyde 3 – P dehydrogenase leading to formation of 1 – Arseno – 3 – Phosphoglycerate (it is unstable compound that hydrolyzed rapidly preventing ATP synthesis)
- Effect of xenobiotics on citric acid cycle :

- Sodium fluoroacetate and fluoroacetamide, are combined with coenzyme A to create fluoroacetyl CoA (FAcCoA) . The FAcCoA substitutes for acetyl CoA, entering the TCA cycle by condensation with oxaloacetate to form fluorocitrate, which inhibits citrate metabolism, resulting in inhibition of the cycle and termination of oxidative metabolism
- Arsenite inhibits thiamine - dependent enzymes within the citric acid cycle
- Effect of xenobiotics on oxidative phosphorylation
- Cyanide, carbon monoxide, and hydrogen sulfide *block the cytochrome a - a3 – mediated reduction of O 2 to H 2 O*
- Xenobiotics that uncouple oxidative phosphorylation, like inhibitors of the electron transport chain, stop ATP synthesis. These substances like dinitrophenol