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# The Anti-Oxidant Effects of Nitric Oxide

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Abbreviation:
PUFA: polyunsaturated fatty acids ROS: reactive oxygen species
RNOS: reactive nitrogen oxide species

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***Abstract***

There is increasing evidence that nitric oxide (NO˙), a free radical that is involved in the both the Pro- and Anti-Oxidant mechanisms *in vivo*. The direct antioxidant mechanism of nitric oxide is mainly through three different pathways: The NO˙ can interact with other biologically relevant free radicals rapidly; the NO˙ also can interact with some redox metal complexes; in addition, NO˙ can interact with nitrogen oxide species (RNOS). Together with the direct antioxidant chemistry of nitric oxide, antioxidant properties of NO˙ can be greatly amplified by the activation of signal transduction pathways that lead to the increased synthesis of endogenous antioxidants or down regulate responses to some stresses.

***1. Introduction***

Nitric oxide is commonly known as an endothelial-derived relaxation factor, but has multiple functions *in vivo*, including modulation of blood pressure, platelet activation and aggregation, neurotransmission, pathogen killing, and triggering of mitochondria biogenesis [1,2,3,4,5,6]. Both antioxidant and pro-oxidant roles for NO· have been reported [7,8,9,10]. General speaking, where, when, and how much NO· is present or is being produced under a given circumstance determines the biological response and whether the result is beneficial or deleterious [10,11,12]. The chemistry of NO· may be separated into two basic categories, direct effects or indirect effects, chiefly based on the concentration of NO·. Direct effects are reactions in which NO· reacts directly with the biological target. Indirect effects are those in which NO· initially combines with another reactant, such as $O_2$ or $O_2^{\cdot -}$ prior to participating in chemical modification of biological targets [10]. Recently, studies of the molecular mechanisms involved in combating oxidative or nitrosative stress have shown that in addition of direct chemical reaction with a free radical or oxidant, the roles as modulators of cell signaling pathways being suggested [3,4].

**2.** ***Reaction between nitric oxide and metal complexes***

There are three major types of NO· reactions with metals: (1) the direct reaction of NO· with the metal center, (2) NO· redox reaction with dioxygen metal complexes, and (3) high valent oxo-complexes (Fig.1).

NO· may react with a variety of metal complexes to form metal nitrosyls. NO· in the presence aquated ferrous ion forms a ferrous-nitrosyl complex, whereas aquated ferric ion does not (Reaction 1). Strong oxidants are produced through Fenton-type reactions of $H_2O_2$ with transition

metal complexes. Direct reaction of NO˙ with metals form relatively stable metal-nitrosyl complexes thereby inhibiting their availability for reaction with $H_2O_2$ [10,12].

$Fe_{aq}(II) + NO˙ \rightarrow Fe_{aq}(II)—NO$ Reaction 1

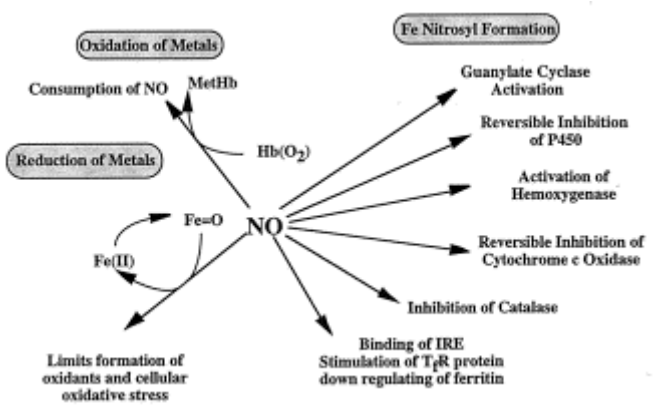


Fig. 1. Direct effects of nitric oxide on metal complexes [12].

The reactivity of NO˙ with metals is not limited to just covalent interactions with metal ions. Various metal-oxygen complexes and metallo-oxo complexes rapidly react with NO˙. An important direct effect of NO˙ is the reaction between NO˙ and oxyhemoglobin to form met-hemoglobin and nitrate (Reaction. 2) [12]

$Hb(Fe---O_2) + NO˙ \rightarrow$ met Hb (Fe(III)) + $NO_3^-$ Reaction 2

One other set of rapid reactions of NO with a biological metal is the reaction of NO˙ with high valent metal complexes. Metallo-oxo species are formed from oxidation of metal species or metal-oxygen complexes by agents such as hydrogen peroxide. Such hypervalent metal complexes are powerful oxidants that can lead to cellular damage such as lipid peroxidation. NO˙ rapidly reacts with these hypervalent complexes to abate oxidative chemistry mediated by metallo-oxo species (Reaction 3) [12].

$Fe^{(2,3)+} + H_2O_2 \rightarrow Fe^{(4,5)+}=O + H_2O$ (3)

Addition of NO˙ results in the reduction of the hypervalent complex to a lower valent state (Reaction 4) [10,11,12].

$Fe^{4+}=O + NO˙ \rightarrow Fe^{3+}+NO_2^-$ (4)

The scavenging of deleterious metallo-oxo species is a potentially important reaction by which NO˙ protects tissue from peroxide-mediated damage [12].

***3. Reaction between nitric oxide and other free radicals***

Kelly *et al*. found that Nitric Oxide can terminate PUFA chain oxidation reaction and works as a great antioxidant [2].

$LOO˙ + NO˙ \rightarrow LOON=O \quad (k = 2 \times 10^9\ M^{-1}\ s^{-1})$ (5)

$LO˙ + NO˙ \rightarrow LON=O \quad (k = 7 \times 10^4\ M^{-1}\ s^{-1})$ (6)

O'Donnell VB. *et al*. researched mechanisms of this reaction, including kinetic parameters and nature of termination products .The reaction between nitric oxide and lipid peroxyl radicals has been proposed to account for the potent inhibitory properties of NO˙ toward lipid peroxidation processes; however, the, have not been defined. Kinetic analysis revealed that a simple radical-radical termination reaction (NO˙:ROO˙ = 1:1) does not account for the inhibition of lipid oxidation by NO˙, and at least two molecules of NO˙ are consumed per termination reaction. A mechanism is proposed whereby NO˙ first reacts with LOO˙ ($k = 2 \times 10^9\ M^{-1}\ s^{-1}$) to form LOONO. Following decomposition of LOONO to LO˙ and $NO_2$, a second NO˙ is consumed via reaction

with $LO^{\cdot}$, with the composite rate constant for this reaction being $k = 7 \times 10^4\ M^{-1}\ s^{-1}$. At equal concentrations, greater inhibition of oxidation was observed with NO than with α-tocopherol. At last, they proposed a serious of reactions as the mechanism of their observation [1]:

$$2\ NO^{\cdot} + 2ROO^{\bullet} \rightarrow 2ROONO \quad (7)$$

$$2ROONO \rightarrow 2RO^{\bullet} + NO^{\cdot} \quad (8)$$

$$RO^{\cdot} + NO_2 \rightarrow RONO_2 \quad (9)$$

$$RO^{\cdot} + NO^{\cdot} \rightarrow RONO \quad (10)$$

$$NO^{\cdot} + NO_2 \rightarrow N_2O_3 \quad (11)$$

$$N_2O_3 + H_2O \rightarrow 2HNO_2 \quad (12)$$

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$$4\ NO^{\cdot} + 2ROO^{\cdot} + H2O \rightarrow 2HNO2 + RONO_2 + RONO \quad \text{Reaction 13}$$

The direct chemical interactions of NO with superoxide ($O_2^{\cdot -}$) or lipid peroxyl radicals ($LOO^{\cdot}$) are of particular interest because they illustrate a molecular basis for some of the apparently antagonistic outcomes of similar chemical reactions when placed in a biological setting (Fig.2). The reaction of $NO^{\cdot}$ and $O_2^-$ is rapid and facile ($k=6.7\times10^9$ M/s), resulting in the formation of peroxynitrite ($ONOO^-$). Similarly, $NO^{\cdot}$ can scavenge $LOO^{\cdot}$ with $k=2\times10^9$ M/s [3, 4].

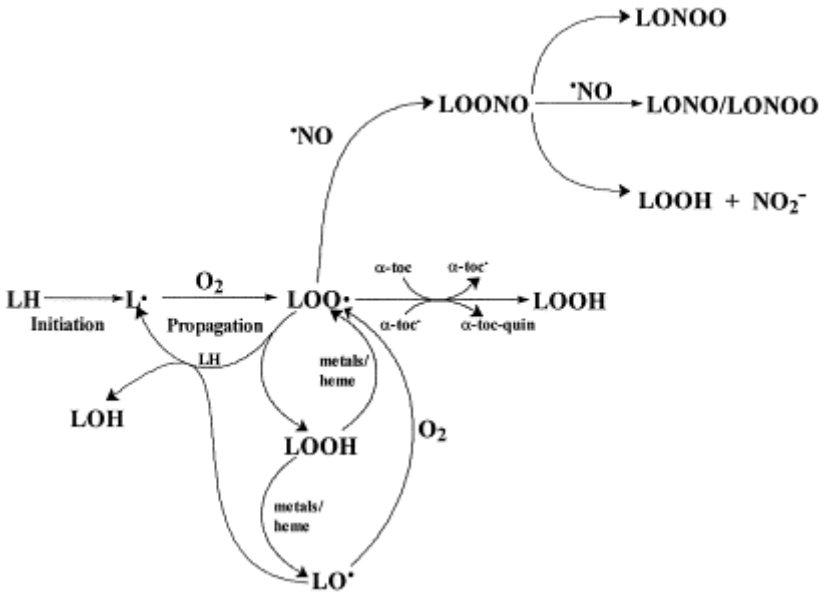


Fig. 2. Inhibition of lipid peroxidation by NO˙. Initiation of lipid peroxidation occurs by abstraction of an allylic hydrogen atom from an unsaturated fatty acid. The rapid reaction between the corresponding carbon centered radical and oxygen forms a lipid peroxyl radical. Propagation occurs via the reaction with LOO˙ and another fatty acid which forms a lipid hydroperoxide (LOOH) and re-generates a LOO˙. Lipid peroxyl radicals can also be formed by decomposition of LOOH in reactions catalyzed by transition metal ions (e.g. copper or iron) either free or in the form of heme proteins (e.g myoglobin and hemoglobin). The central role of LOO˙ is thus apparent and scavenging of these radicals by antioxidants prevents lipid peroxidation [4].

### *4. Reaction between nitric oxide and RNOS*

The reaction between NO˙ and $O_2$ is termed NO˙ autoxidation. In the gas phase and in hydrophobic layers of cellular membranes, NO˙ autoxidation reaction initially produces $NO_2$. With an oxidation potential of 1.2 V versus a standard hydrogen electrode, $NO_2$ is a strong oxidant that can mediate lipid peroxidation reactions. But NO˙ can react with $NO_2$ to form $N_2O_3$, with an oxidation potential of 0.7 V, which is the quintessential species in most biological nitrosative conditions [10].

$$2\,NO^{\cdot} + O_2 \rightarrow 2NO_2 \quad (13)$$

$$2NO_2 + NO^{\cdot} \rightarrow N_2O_3 \quad (14)$$

The reaction between NO· and led to formation of $ONOO^-$, which can cause lipid peroxidation, base modification, strand breaks, cysteine oxidization and dityrosyl-bridges formation. Decomposition of $ONOO^-$ is suggested to proceed through peroxynitrous acid (ONOOH, pKa 6.8). NO· can also can decompose ONOOH [12].

$$NO\cdot + O_2^{\bullet -} \rightarrow ONOO^- \quad (15)$$

$$ONOOH + NO\cdot \rightarrow NO_2 + NO_2^- + H^+ \quad (16)$$

By these mechanisms, NO· serves to abate the oxidation chemistry of RNOS.

***5. Conclusion***

NO·can be a very effective antioxidant to both ROS and RNOS. The antioxidant mechanisms is through the versatile chemistry of NO· with ligand-metal, radical-radical and RNOS. Because of the extremely complexity in nitric oxide function and mechanism, much more research is needed. The current view is that where, when, and how much NO· is present or is being produced under a given circumstance determines the biological response and whether the result is beneficial or deleterious [10,11,12].

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