Antioxidant responses to oxidant-mediated lung diseases

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Comhair, Suzy A. A., and Serpil C. Erzurum. Antioxidant responses to oxidant-mediated lung diseases. *Am J Physiol Lung Cell Mol Physiol* 283: L246–L255, 2002; 10.1152/ajplung.00491.2001.—Reactive oxygen species (ROS) and reactive nitrogen species (RNS) are generated throughout the human body. Enzymatic and nonenzymatic antioxidants detoxify ROS and RNS and minimize damage to biomolecules. An imbalance between the production of ROS and RNS and antioxidant capacity leads to a state of "oxidative stress" that contributes to the pathogenesis of a number of human diseases by damaging lipids, protein, and DNA. In general, lung diseases are related to inflammatory processes that generate increased ROS and RNS. The susceptibility of the lung to oxidative injury depends largely on its ability to upregulate protective ROS and RNS scavenging systems. Unfortunately, the primary intracellular antioxidants are expressed at low levels in the human lung and are not acutely induced when exposed to oxidative stresses such as cigarette smoke and hyperoxia. However, the response of extracellular antioxidant enzymes, the critical primary defense against exogenous oxidative stress, increases rapidly and in proportion to oxidative stress. In this paper, we review how antioxidants in the lung respond to oxidative stress in several lung diseases and focus on the mechanisms that upregulate extracellular glutathione peroxidase.

redox; reactive oxygen species; reactive nitrogen species

OXYGEN IS ONE OF THE MOST abundant elements in our world, constituting 21% of the air we breathe (20, 122). It is essential for the oxidation of organic compounds, which is the process by which mammalian cells generate the energy needed to sustain life. However, oxygen may also damage the lung. Inhaled ozone and nitric oxide may induce toxic processes that impair lung function (20, 38, 82, 119, 122). Under normal conditions, potentially toxic oxygen metabolites are generated at a low level in lung cells by the transfer of a single electron during aerobic metabolism (25, 33, 46). The resulting reactive oxygen species (ROS), which include hydroxyl radicals, superoxide (O$_2^-$), and hydrogen peroxide (H$_2$O$_2$), play an integral role in the modulation of several physiological functions but can also be destructive if produced in excessive amounts (31, 38, 82, 99, 104, 107). Similarly, reactive nitrogen species (RNS) such as nitric oxide, nitrite, and peroxynitrite (ONOO$^-$) are both physiologically necessary and potentially destructive.

Another oxygen-mediated mechanism of damage is inflammation, during which leukocytes, macrophages, and mast cells release mediators that may cause bronchoconstriction and edema as observed during an asthmatic reaction (15, 38, 64). Lung tissue can also be destroyed during reperfusion after an ischemic period such as that produced by surgery (42, 94, 99, 104, 107, 134). All these mechanisms have one thing in common: damage is at least partly mediated by oxidants and nitrogen species.

To minimize oxidant damage to biological molecules, the human lung is endowed with an integrated antioxidant system of enzymatic and expendable soluble antioxidants. This system includes several antioxidant defense mechanisms that detoxify reactive products or convert them to products that are quenched by other antioxidants (47, 58). If the oxidant burden is sufficiently great, the reactive species may overwhelm or inactivate the antioxidant system. The resulting excess
Oxygen species can damage major cellular components, including membrane lipids, protein, carbohydrates, and DNA. The pathophysiological consequences of this injury are inflammation and widespread tissue damage (46).

**OXYGEN AND REACTIVE OXYGEN SPECIES**

More than 90% of all the oxygen we breathe undergoes a concerted tetravalent reduction to produce water in a reaction catalyzed by cytochrome oxidase in the mitochondrial electron transport chain. Oxygen (O2) can also be reduced via a nonenzymatic pathway through four successive one-electron (e\(^{-}\)) reductions (6, 34) (Eq. 1).

\[
O_2 + 4H^+ + 4e^- \rightarrow 2H_2O
\]  
(1)

Cytochrome oxidase is the terminal electron acceptor in the respiratory chain and must donate its reducing equivalents to oxygen to allow continued electron transport. Otherwise, ATP production cannot continue. Thus the major role for oxygen in all aerobic organisms is simply to act as a sink or dumping ground for electrons (34). The tetravalent reduction of oxygen by the mitochondrial electron-transport chain is considered a relatively safe process. Nonetheless, the electron carriers catalyze alternating one-electron oxidant-reduction reactions, and they can react with oxygen to generate ROS such as O2\(^{\cdot}\) (47, 96, 97). Mitochondria are the major intracellular sites of O2\(^{\cdot}\) generation under physiological conditions (53). One other potentially major source for the generation of O2\(^{\cdot}\) is the NADPH oxidase enzymatic system, which is found in neutrophils, monocytes, macrophages, cytochrome P-450, monoamine oxidase, and lipooxygenase (4, 22, 31, 34). O2\(^{\cdot}\) is also generated by other mechanisms such as molybdenum hydroxylase reactions (including the xanthine, sulfite, and aldehyde oxidases) and arachidonic acid metabolism.

O2\(^{\cdot}\) is relatively unstable, with a half-life of only milliseconds. Because it is charged, it does not easily cross cell membranes (6). O2\(^{\cdot}\) will react, however, with proteins that contain transition metal prosthetic groups, such as heme moieties or iron-sulfur clusters. These reactions may damage amino acids or cause protein/enzyme function to be lost (50, 138). Most of the O2\(^{\cdot}\) generated in vivo undergoes a nonenzymatic or superoxide dismutase (SOD)-catalyzed reaction, resulting in the nonradical H2O2 (Eq. 2) (79). H2O2 can also be directly produced by several oxidase enzymes, including xanthine oxidase, monoamine, and amino acid oxidase (24).

\[
O_2^{\cdot} + O_2^{\cdot} + 2H^+ \rightarrow H_2O_2 + O_2
\]  
(2)

H2O2 can be oxidized by eosinophil-specific peroxidase (EPO) and neutrophil-specific peroxidase (MPO) using halides (X\(^{-}\)) as a cosubstrate to form the potent oxidant hypohalous acids (HOX) and other reactive halogenating species (Eq. 3) (44, 56, 70, 130).

\[
H_2O_2 + X^- + H^+ \rightarrow HOX + H_2O \quad X = Br^-, Cl^-
\]  
(3)

Much of the damage done by O2\(^{\cdot}\) and H2O2 in vivo is due to their production of hydroxyl radicals (\(\cdot\)OH) in a series of reactions catalyzed by traces of transition ions. One such example is the iron-catalyzed Haber-Weiss reaction in which Fe3\(^{\cdot}\) is reduced to Fe2\(^{\cdot}\), followed by the Fenton reaction in which the Fe2\(^{\cdot}\) catalyzes the transformation of H2O2 into (\(\cdot\)OH) (Eq. 4) (54).

\[
O_2^{\cdot} + Fe^{3+} \rightarrow Fe^{2+} + O_2 \quad \text{Haber-Weiss reaction}
\]

\[
H_2O_2 + Fe^{2+} \rightarrow Fe^{3+} + OH^- + H_2O \quad \text{Fenton reaction}
\]  
(4)

An alternative pathway for \(\cdot\)OH formation in vivo may involve MPO and EPO. Under physiological concentrations of halides, MPO produces hypochlorous acid (HOCl), and EPO produces hypobromous acid (HOBr). Studies of \(\cdot\)OH with spin-trapping agents (41, 124) and chemical traps (57, 95) have demonstrated that hypohalous acids can generate \(\cdot\)OH after reacting with O2\(^{\cdot}\) (Eq. 5). \(\cdot\)OH can react with different molecules such as protein (16), DNA, and lipids (49).

\[
O_2^{\cdot} + HOX \rightarrow \cdot\text{OH} + X^- + O_2
\]  
(5)

**RNS**

The discovery that nitric oxide (NO) is endogenously formed throughout the human body has led to intense interest in the variety of roles this unique molecule plays in vivo. NO is involved in a wide variety of regulatory mechanisms. In addition, NO is also a cytotoxic agent present in environmental pollutants and cigarette smoke (109). NO is formed from the semiesential amino acid L-arginine by the action of nitric oxide synthase (NOS; Fig. 1) (5, 90). Several forms of NOS have now been characterized, and several distinct NOS genes have been identified (73). The NOS are classified as either constitutive or inducible (76, 89, 124). NO synthase (NOS; Fig. 1) (5, 90).

**Fig. 1.** Reactive nitrogen species (RNS) synthesis. NO, nitric oxide; NO2, nitrite; NO3, nitrate; NO2, nitrate; NO2, oxyhemoglobin; Hb3\(^{\cdot}\), methemoglobin; ONOOH, peroxynitrous acid; MPO, myeloperoxidase; HOCl, hypochlorous acid; SNO: S-nitrosothiol; NOS II, nitric oxide synthase II.
The constitutive forms (NOS I and NOS III) are cytosolic and originally described and cloned from neuronal and endothelial cells, respectively. They are dependent on Ca\textsuperscript{2+} and calmodulin and release low amounts of NO for short periods in response to receptor and physical stimulation (89). The inducible form (NOS II) is independent of Ca\textsuperscript{2+}. Once expressed, NOS II generates NO in large amounts for long periods (131). The biochemical effect of NO is largely defined by the concentration of NO. The paramagnetic NO molecule contains an odd number of electrons, which explains its highly reactive and radical nature (Fig. 1) (65, 113). Autooxidation of NO with O\textsubscript{2} results in the formation of nitrite (NO\textsubscript{2}\textsuperscript{-}). However, at physiological concentrations of NO and O\textsubscript{2}, this reaction may be too slow to be important in vivo (8, 66). NO\textsubscript{2} is also a substrate for hemoperoxidases such as MPO and EPO, which catalyze peroxidase-mediated oxidation and chlorination of biological targets (40, 41, 66, 124, 130, 135, 136). Moreover, peroxidase-catalyzed oxidation of NO\textsubscript{2} results in the formation of a nitrogen dioxide radical (NO\textsubscript{2}•) or related molecules. These substances can contribute to the nitrification of phenolic compounds such as tyrosine to form dimerized (dityrosine) and nitrated (3-nitrotyrosine) products, which are stable (40, 41, 66, 124, 130, 135, 136).

Although NO\textsubscript{2} is a major end product of NO, it does not accumulate in vivo but is rapidly oxidized to nitrate (NO\textsubscript{3}•). NO is also rapidly oxidized by oxyhemoglobin (HbO\textsubscript{2}), which results in the formation of methemoglobin (Hb\textsuperscript{3+}) and NO\textsubscript{3} (1, 14, 51). The oxidative metabolism of NO may also lead to the formation of carcinogenic nitrosoamines (72, 75) and the rapid formation of free or protein-associated 3-nitrotyrosine (91, 131). NO\textsubscript{2}•-nitrosothiols (SNO) have been proposed as a mechanism whereby NO groups are transported and/or repair damaged molecules. However, an antioxidant cannot distinguish between radicals that play a physiological role and those that cause damage (6). Moreover, some antioxidant compounds also have prooxidant actions (6). This section will review the enzymatic and nonenzymatic primary antioxidant defenses.

Enzymatic antioxidants. SOD (EC 1.15.1.11) is a ubiquitous enzyme with an essential function in protecting aerobic cells against oxidative stress (79). It catalyzes \textgreek{O}_2• radicals to H\textsubscript{2}O\textsubscript{2}. There are three forms of SOD. The copper-zinc SOD is located in the cytosol, the manganese SOD is primarily a mitochondrial enzyme, and extracellular SOD is usually found on the outside of the plasma membrane (43).

Catalase (EC 1.11.1.6) is a tetrameric hemoprotein that undergoes alternate devalent oxidation and reduction at its active site in the presence of H\textsubscript{2}O\textsubscript{2} and catalyzes the dismutation reaction (22, 36, 102). As a result, catalase has appreciable reductive activity for small molecules such as H\textsubscript{2}O\textsubscript{2} and methyl or ethyl hydroperoxide. It does not metabolize large molecular peroxides such as lipid hydroperoxide products of lipid peroxidation (129). Catalase is most effective in the presence of high H\textsubscript{2}O\textsubscript{2} concentrations. However, in the presence of low concentrations of either H\textsubscript{2}O\textsubscript{2} or other peroxides, the glutathione system plays a critical role (20).

The glutathione system is a central mechanism for reducing H\textsubscript{2}O\textsubscript{2}. It complements catalase as a reducing system for H\textsubscript{2}O\textsubscript{2} but exceeds catalase in its capacity to eliminate additional varieties of toxic peroxides (105). Other metabolized substrate species include large molecule lipid peroxides, formed by free radical attack on polyunsaturated lipid membranes and products of lipoxygenase-catalyzed reactions (58). The key enzyme in the reduct cycle responsible for the reduction of H\textsubscript{2}O\textsubscript{2} is GPx. This reaction specifically requires reduced glu-
tathione (GSH) to serve as the electron donor. The glutathione disulfide (GSSG) formed in the course of the reaction is subsequently reduced back to GSH by glutathione reductase, which uses NADPH generated from the hexose monophosphate shunt system as an electron donor (37, 80). Healthy, nonstressed cells maintain a high intracellular GSH:GSSG ratio to ensure the availability of GSH and thereby promote active reduction of H₂O₂ through the glutathione system (37, 80). In a role unrelated to its role in the GSH system, free GSH can also function as a water-soluble antioxidant by interacting directly with radical intermediates in nonenzymatic catalyzed reactions. Scavenging of O₂⁻ by GSH leads to the formation of thyl radicals (GS·) and H₂O₂ via several steps, which is a radical propagation reaction. This reaction leads to the formation of GS· and H₂O₂ and can occur in physiologically relevant concentrations. Hence, a substance that is generally accepted to be an antioxidant may possess prooxidant activity under certain conditions (6, 38).

Four GPx have been described, all selenium enzymes: 1) the classic cytosolic form, found in all cells (9); 2) a membrane-associated GPx phospholipid H₂O₂ (39); 3) another cytoplasmic enzyme, gastrointestinal GPx, which was first found in cells of the gastrointestinal tract (23); and 4) an extracellular GPx (eGPx), first identified as a distinct enzyme in human plasma (137). All members of this family of enzymes can be oxidized by organic hydroperoxides, hydroperoxide, or both, and can subsequently be reduced by glutathione (137). The existence of multiple forms of GPx is due to the expression of different genes (103). All GPx contain a selenium atom in the active site in the form of selenocystine.

Nonenzymatic antioxidants. Cells use nonenzymatic antioxidant compounds to react directly with oxidizing agents and disarm them. Such antioxidants are said to be “scavengers”; their roles are unavoidably suicidal. For example, vitamin E (α-tocopherol) is a membrane-bound antioxidant that terminates the chain reaction of lipid peroxidase by scavenging lipid peroxyl radicals (LOO·) (6, 34, 123). In this reaction, vitamin E becomes a radical, but it is much less reactive than LOO· (123). However, at high concentrations, the radical form of vitamin E may function as a prooxidant (6). Vitamin C can also directly scavenge O₂⁻ and -OH by forming the semidehydroascorbate free radical that is subsequently reduced by GSH (78). Vitamin C, however, is usually not considered a major antioxidant because it also has prooxidant properties. It is probably the only cellular reducing agent other than O₂⁻ capable of converting Fe³⁺ to Fe²⁺, which then reacts with H₂O₂ to form -OH (106). Whether the prooxidant or antioxidant properties of vitamin C prevail in any particular tissue is determined by the extent of available iron stores; iron overload favors excess oxidant generation (6, 106). Other nonenzymatic antioxidants include β-carotene (scavenger of O₂⁻, anions and peroxyl radicals), uric acid (hydroxyl radical, O₂⁻, peroxyl radical scavenger), glucose (hydroxyl radical scavenger), bilirubin (LOO⁻ scavenger), taurine (hypochlorous acid quencher), albu-

**ANTIOXIDANTS IN THE LUNG**

Lungs are unique because they have a large epithelial surface area that is at risk for oxidant-mediated attack. The tracheobronchial tree and the alveolar space are exposed to reactive oxidizing species in the form of inhaled airborne pollutants, tobacco smoke, and products of inflammation. The lung, therefore, requires additional antioxidant resources to prevent airway-borne oxidant injury (58). The major airways contain high-molecular-weight mucopolypeptide glycoproteins, which are synthesized by the epithelial cells and glands that increase mucus production in the presence of inflammation (58). The lung contains intracellular antioxidant enzymes to maintain a normal redox state. The alveolar space can recruit additional antioxidant activity from the epithelial lining fluid (ELF). This fluid contains large amounts of GSH (100-fold higher than in plasma), 90% of which is in the reduced form (19, 27, 35, 114, 115). The ELF also contains catalase, SOD, and GPx (19, 27, 114, 115). Additional antioxidants contained in ELF include ceruloplasmin, transferrin, ascorbate, vitamin E, ferritin, other serum proteins, and small molecules such as bilirubin (58). The multiplicity of the antioxidant systems available to the lung and their overlapping specific activities suggest that to maintain normal pulmonary cellular function, it is critically important for the lung to adequately control redox balance. Disequilibrium, either through increased oxidant stress or decreased antioxidant resources, can result in a series of pathophysiological events in the lung that culminate in cellular death and pulmonary dysfunction (58). A partial list of major lung diseases associated with oxidants is presented in Table 1.

**EXTRACELLULAR ANTIOXIDANT RESPONSE IN LUNGS EXPOSED TO OXIDATIVE STRESS**

Normally, the homeostasis of cellular functions during oxidative stress depends on the rapid induction of protective antioxidant enzymes. Naturally occurring antioxidants exist to protect cells and tissue against the continuous production of ROS/RNS during normal metabolism (58). Tissues and cells respond to mild oxidative stress by increasing antioxidant defenses (119). However, high levels of ROS/RNS may overwhelm antioxidant defenses, resulting in oxidant-mediated injury or cell death (4, 20).

Numerous studies have revealed that oxidant stress plays a crucial role in the initiation and progression of a wide range of diseases and in the regulation of a number of important biological processes. Pulmonary diseases associated with oxidative stress include asthma, hyperoxia, sarcoidosis, and chronic beryllium disease (CBD). ROS play a key role in the initial lung response to asbestos and silica that leads to interstitial pulmonary fibrosis (87, 88). Interestingly, during the development of pulmonary diseases, antioxidant re-

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sponse are different. For example, asbestosis and sarcoidosis lead to an increase of SOD, whereas there are no changes found in silicosis or hyperoxic lung injury (27, 62, 71). In contrast, SOD activity is significantly lower in patients with asthma and decreases further during an asthmatic exacerbation.

The glutathione system is altered in lung inflammatory conditions. For instance, GSH levels are elevated in the ELF of chronic smokers and in chronic beryllium disease, an immune-specific granulomatous inflammation (29). Levels of GSH in ELF decrease rapidly in patients with mild asthma during an asthma exacerbation (27). Similarly, GSH levels are decreased in ELF in idiopathic pulmonary fibrosis (17, 74), asbestosis (11), acute respiratory distress syndrome (13), and in human immunodeficiency virus-positive patients (12). Levels of glutathione modulate the T helper type 1 (Th1) vs. the Th2 immune response pattern (93). For example, high levels of glutathione in patients with CBD may contribute to the development and/or maintenance of a chronic Th1 cell-mediated immune response to beryllium, whereas the low GSH levels may contribute to the development or maintenance of Th2 cell immune response in asthma.

Other enzymes of the glutathione system are also influenced by oxidant stress. Some studies have shown increased GPx activity in ELF of smokers compared with nonsmokers (29, 104), whereas others have shown decreased GPx in smokers (81). The difference in GPx activity may be due to the difference in smoking history (19, 86). GPx activity is not altered in asthma but is increased in lungs of CBD patients. Overall, the antioxidant response is inconsistent across different oxidant-mediated lung diseases.

eGPx AND ITS ROLE IN OXIDATIVE STRESS

Expression of eGPx. eGPx transcripts have been found in epithelial cells with well-developed brush borders that contain lipids and alkaline phosphatase activity, e.g., the human airways, intestine, and renal tubules (2, 3, 68, 120). Alveolar macrophages are also able to synthesize and secrete eGPx (2, 28, 30). The GPx family is an important enzymatic component of the mechanisms for detoxifying ROS in the lung and may play a significant role in preventing pulmonary oxidant stress. eGPx gene expression is upregulated in bronchial epithelial cells and ELF as a result of oxidative stress occurring in individuals with asthma or CBD and in those who have been exposed to exogenous oxidants such as cigarette smoke (Fig. 2) (2, 28–30). The upregulation of eGPx occurs rather late after exposure (after 24 h) (28), which may explain why eGPx was not induced after 12 h of hyperoxia. In support of this, levels of eGPx mRNA and protein increase only after 72 h of hyperoxia in a mouse model (67). Induction of eGPx mRNA in bronchial epithelial cells is associated with elevated protein levels in ELF, suggesting that the increase of eGPx occurs, in part, by bronchial epithelial cell synthesis and secretion (28). However, alveolar macrophages can also express eGPx (2, 30). It is not known whether other lung cells or inflammatory cells upregulate eGPx gene in response to oxidative stress.

Bronchial epithelial cells significantly increase eGPx mRNA expression in response to increased intracellular or extracellular ROS in vitro. Supplementation of GSH in cell culture to physiological levels potentiates

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<th>Table 1. Lung diseases associated with oxygen radicals</th>
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<td><strong>Disease</strong></td>
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<tr>
<td>Emphysema</td>
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<td>Adult respiratory distress syndrome</td>
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<td>Hyperoxia</td>
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<td>Idiopathic pulmonary fibrosis</td>
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α1PI, α-1-proteinase inhibitor.
the induction of eGPx mRNA in response to ROS (28). This effect is not reproduced by N-acetylcysteine, suggesting that other effects of thiol groups are necessary to achieve the synergistic effect on induction of the eGPx gene (28). Although GSH is usually considered an antioxidant, it can also act as an oxidant when present at physiological levels (6). GSH may participate in oxidizing processes and/or accelerate the generation of ROS, specifically O$_2^-$ (6, 84, 121, 133). Previous reports have shown that GPx can function as an ONOO$^-$ reductase and thereby prevent nitration reactions caused by RNS (45). On the other hand, NO donors (S-nitroso-N-acetyl-D,L, penicillamine/GSNO) induce eGPx gene expression in a time- and dose-dependent manner.

**Transcriptional regulation.** The regulation of genes in response to oxidative stress occurs via transcription and/or stabilization of mRNA. Studies support that the ROS regulation of the eGPx gene expression occurs at a transcriptional level (28). In general, ROS and RNS regulate the expression of numerous genes via signaling mechanisms. Redox-sensitive transcription factors such as signal transducers and activators of transcription (STAT), nuclear factor-κB, and transcription factor activator protein-1 (AP-1) are activated in epithelial cells and inflammatory cells during oxidative stress (98, 100). Although the STAT family of transcription factors is activated by many cytokines and growth factors, it can also be activated by oxidative stress such as H$_2$O$_2$ (108, 112, 116). The activation of STATs by oxidative stress is inhibited by antioxidants. Several ROS-induced target genes have known STAT binding sites in their promoters. These include genes involved in antioxidant defense (111) such as SOD1 (10) and genes involved in cell growth regulation such as c-fos (128).

Studies from several laboratories have demonstrated that oxidant stress such as cigarette smoke, treatment with H$_2$O$_2$, depletion of intracellular GSH, or an increase in the GSH:GSSG ratio stimulates AP-1 activation and binding (83, 99). AP-1 regulates many of the inflammatory and immune genes in oxidant-mediated diseases (69, 110, 118). AP-1 is a protein dimer composed of the Jun and Fos gene products (100). These

![Fig. 3. Proposed scheme for eGPx gene induction by oxidative stress.](image)

![Fig. 4. Detoxification of ROS and RNS by eGPx in asthmatic individuals.](image)

![Fig. 5. Nitrotyrosine staining of asthmatic bronchial mucosa.](image)
gene products can form homodimeric (Jun-Jun) or heterodimeric (Jun-Fos) complexes (100). The 5′-flanking region of the eGPx gene contains the DNA-binding element for AP-1 (137) (Fig. 3). Cigarette smoke increases AP-1 DNA binding in human epithelial cells in vivo (99). Based on our results and those of others, we propose that increased formation of ROS and RNS in inflammatory cells and epithelium leads to alterations in the redox system, sensed by the epithelial cells, and leads to the activation of transcription factors such as STATs and AP-1 and downstream target gene expression.

Function. On the basis of our studies and the studies of others, we have generated a model for the function of eGPx in lung diseases associated with oxidative stress (Fig. 3). NO is produced in mammalian airways, and increased levels are found in many inflammatory lung diseases such as asthma and hyperoxia (28, 30, 61). Inflammation leads to increased levels of ROS (Fig. 4, pathway a). NO reacts slowly with $O_2^-$ to form the cytotoxic compound $NO_2$ or very rapidly with $O_2^\cdot$ to form ONOO$^-$ (Fig. 4, pathway b) (51). This results in increased tyrosine nitration in lung tissue (Fig. 5 and Fig. 4, pathway c). Thus when $O_2^-$ is produced at high rates during lung inflammation, NO may accelerate the formation of ONOO$^-$, leading to tyrosine nitration and aggravate lung damage (61). The NO/O$_2^\cdot$ pathway in vitro appears to be significantly shifted toward the formation of GSNO in the presence of physiological levels of GSH (Fig. 4, pathway d) (30, 45, 77). Recent studies have shown that GPx (Fig. 4, pathway c) can protect against NO-mediated protein oxidation (48) and can reduce GSNO. Thus the increased eGPx in lung inflammation may have two functions: reduction of ROS (e.g., $H_2O_2$ and $O_2^\cdot$) and possibly protection against NO-mediated protein oxidation by regulation of RSNO levels.

In conclusion, lung diseases occur in response to oxidative and nitrosative stress. The lung’s ability to respond to oxidative stress depends largely on its capacity to upregulate protective antioxidants. The antioxidant responses to lung disease vary widely, but the upregulation of eGPx in oxidant-mediated lung diseases is likely important to defend airway surfaces from ROS and RNS injury.

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