



## Oxidative stress in COPD, pathogenesis and therapeutic views

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| ARTICLE INFO  | ABSTRACT  |
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| <p><b>Article type</b><br/>Review article</p> <p><b>Article history</b><br/>Received: 2 Feb 2014<br/>Revised: 19 Mar 2014<br/>Accepted: 15 Apr 2014</p> <p><b>Keywords</b><br/>Chronic Obstructive Pulmonary Disease (COPD)<br/>Hypersecretion<br/>Oxidative stress</p> | <p>Chronic obstructive pulmonary disease (COPD), characterised by partially reversible contracture of small respiratory airways seems to be among leading causes of death in the world. COPD is characterized by inflammation, protease/antiprotease imbalance, genetic variability and oxidative stress. The latter refers to a condition in which oxidative agents overcome against antioxidants. In this review literature, the consequences of oxidative stress in COPD, such as systemic and pulmonary neutrophil influx, hypersecretion, dual and reciprocal effects with inflammatory contributors and systemic manifestations are discussed. In addition, a review of oxidative stress biomarkers as well as therapeutic strategies based on recent researches for antioxidant supplementation therapy is provided.</p> |

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### Introduction

The respiratory system contacts permanently to internal and external oxidant. Air pollutants and particles in smokes are external and phagocytic products are internal instances of oxidative substances.

Besides, intracellular organelles like mitochondria can produce radicals which act as oxidative agents. Enzymatic and non enzymatic antioxidants are defensive

system against invasive oxidative agents (1). Overcomes of oxidant burden to antioxidants and diminished antioxidants availability can lead to oxidative stress. In cigarette smokers that each puff contains 1017 oxidative particles, lung cells are susceptible to oxidative stress, due to excessive burden of oxidants over antioxidants. According to this occurred

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imbalance, cigarette smoking has the cardinal etiologic role in chronic obstructive pulmonary disease (COPD) (2).

In COPD patients, lung injuries occur because of both direct damage and inflammatory processes by oxidative agents not only in pulmonary system but also in other vital organs, explaining other non-pulmonary manifestations such as muscular atrophy and weight loss (3,4).

#### ***Measurable oxidative stress markers***

Hydrogen peroxide existing in bronchoalveolar lavage (BAL) fluid or in expired flow is a measurable indicator to assess oxidative stress activity directly and indirectly. Nitric Oxide (NO) measurement in exhaled flow can determine oxidative burden. Another way to meter oxidative stress is evaluating antioxidants changes in obtained sputum or BAL fluid that reflects oxidants amount inversely (5).

Oxidant agents catalyses proteolytic reactions, through breaking of peptide chain of large protein molecules, yielding residual degraded products like carbonyl residues, which accounts an important measurable marker to show oxidative stress quantity (6,7). Several other final products of chemical pathway, occurring during oxidative stress condition can be applied as valid markers, including nitrotyrosine (a protein nitration product), 8-hydroxydeoxyguanine (a product of DNA destruction), F<sub>2</sub>-isoprostanes, 4-hydroxynonenal (4-HNE), and hydrocarbons (final metabolite of lipid peroxidation); all of them are found in sputum, BAL fluid, exhaled breath and lung tissue (8,9) Cigarette smoking can cause elevation of some of these indicators (7,10). A parameter like gamma-glutamyltransferase, a plasma membrane enzymatic antioxidant, can successfully be applied for distinguishing a stable COPD from starting exacerbation (9).

#### ***Oxidative stress witnesses in respiratory system***

Numerous studies have been performed to discover any correlation between oxidative stress biomarkers and COPD. Merely all of these biomarkers showed significantly higher levels in smoker subjects as well as COPD patients in comparison to healthy population. Many of these markers showed a direct relationship between themselves and airway obstruction represented by FEV<sub>1</sub>. Some of these markers such as H<sub>2</sub>O<sub>2</sub> increases severely during exacerbation of COPD and some of them like 8-isoprostane share in inflammatory processes, expressing that oxidative stress can contribute to inflammation.

The occurrence of oxidative stress in lungs of COPD patients and in cigarette smokers is demonstrated by higher levels of oxidative markers in their pulmonary samples compare to healthy or non smoker subjects (2,11,12). H<sub>2</sub>O<sub>2</sub> as a marker is measurable and was shown with even higher levels in COPD exacerbation. The sources of this marker include alveolar macrophages making super oxide radicals (O<sup>-2</sup>) (13,14), increased iron content of macrophages in COPD runs the Fenton reaction by which reactive oxygen species (ROS) formation is exited (10,15) and xanthin/xanthin oxidase reaction is the alternative source (16,17).

NO, another marker presents with variable proportion in COPD patients compare to normal subjects which is probably due to its unstable molecular structure but nitrotyrosine generated from reaction of NO with superoxide is increased in sputum of COPD patients and inversely correlates with FEV<sub>1</sub> (8,18).

Additional marker found in higher levels in patients with COPD is carbon monoxide (CO), with limited use for evaluation of oxidative stress, because of its production directly by cigarette as well as NO (19).

Furthermore, lipid peroxidation metabolites such as 8-isoprostane are in higher levels in COPD patients, additionally these markers reflect inflammation in respiratory airways, and correlate directly with airway obstruction (20,21). Hydrocarbon gases like ethan produced from lipid destruction is another marker that is higher in COPD patients (9).

### ***Antioxidants and COPD***

Based on oxidants/antioxidants imbalance which lead to higher oxidants burden in COPD patients, many documented data have demonstrated the effect of antioxidants augmentation in protecting lung airway and tissues from more damaging of oxidative agents (22,23).

According to this hypothesis many studies have shown increased dietary intake of vitamin C, vitamin A and other antioxidant fortified nutritional regimen in COPD patients can markedly improve their pulmonary function reflecting by increased FEV1 in patients (23-30). Some of studies failed to show a protective role for vitamin E (23-25).

### ***How oxidative stress play role in COPD pathogenesis?***

#### ***Oxidative stress and protease/antiprotease imbalance***

Antiprotease enzymes are among the targets of oxidizing agents, which are found in cigarette smoking or released by leukocytes. These agents inactivate antiproteases, leading to excessive protease activity which is the main cause of emphysema developing (31,32). Oxidative agents oxidise methionine in its active site thereby inactivate  $\alpha$  1-antitrypsin contributing to decreasing of antiprotease activity and increasing protease activity which result in more destruction of histological architecture of lungs, events

are demonstrated in vivo only immediately after cigarette smoking and not in later time (33,34).

#### ***Oxidative stress and mucus hypersecretion***

hypersecretion induced by oxidant agents such as xanthin/xanthin oxidase (35) can cause airway limitation (36). More mucus production owing to epithelial growing stimulated by oxidant agents occurs along with ciliary impairment contributing to more accumulation and finally stasis of mucus in pulmonary airways (37,38).

#### ***Oxidative stress and damage to airspace***

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#### ***Oxidative stress and damage to airspace epithelial***

Airway epithelium is an enlarged surface area exposed directly to environmental oxidants including cigarette smoking and air pollutants which make this surface completely vulnerable and cause increased permeability of epithelium which means inflammation of respiratory airways (39-41). Intra and extracellular antioxidant glutathione is the major defensive system against oxidative agents in this area, confirmed by in vitro and in vivo changes of this antioxidant following cigarette smoking (41-44).

The protective effects of related antioxidant system located in epithelium vary in kinds and amounts in persons probably regulated by genes. This can be one description why only 10% and not all the cigarette smokers

progress to COPD (45).

### ***Neutrophil influx in oxidative stress in the lungs***

Among the effects are thought for oxidative stress is decreased ability of neutrophils to deform for passing across the intravascular to interalveolar spaces or moving through capillaries which are smaller in diameter than neutrophils, leading to increased neutrophil sequestration in lungs; a fact investigated in cigarette smoker by in vitro methods and also confirmed in vivo (46,47).

Cigarette smoking reduces neutrophil deformability and then increases their sequestration. Protective antioxidants act opposite performance and diminish oxidative effects resulting from cigarette smoking reflecting by depletion in plasma concentration of antioxidants during cigarette smoking shown in Drost et al. study (48).

Another reason for developing COPD in only a proportion of smokers can be addressed to variability of latter effect of antioxidants which regulate neutrophil deformability and sequestration (49,50). Changes in plasma concentration of antioxidants followed by cigarette smoking reflect systematic stimulation of protective antioxidants which in turn can provoke circulating neutrophils without need for neutrophil emigration to destruct alveolar walls and develop emphysema.

### ***Relationship between oxidative stress and lung inflammation***

Oxidative stress and inflammation have reciprocal influence on each other in domain of respiratory diseases like COPD and healthy cigarette smokers also. Releasing of inflammatory mediators, nuclear instigation to produce new genes and susceptibility to fall in COPD trap, are among the most important implication, briefly are discussed

here. Investigation have shown increased secretion of many of mediators like IL-8 and TNF-  $\alpha$  in COPD patients and in cigarette smokers (51,52).

Furthermore many correlation have confirmed by diverse driven studies including relationship between oxidative stress marker in exhaled respiration and neutrophil count in sputum (21), and relation between peripheral blood neutrophil count and restricting of airways reflected by FEV1 (53,54). Oxidant/antioxidant imbalance as occurs in COPD and cigarette smoker causes activating many signalling mechanisms which has a central role in developing inflammation. All of these pathways have common goal; increased gene transcription to upregulate inflammatory mediators. Among them the most important are nuclear factor kappa-b (NF-KB), activating protein 1(AP-1), extracellular signal-regulated kinase (ERK), and c-jun N-terminal kinase (JNK) and p38 mitogen-activated protein, all of them exited by cigarette smoking (55-58). All of these processes are redox-sensitive (59,60).

As demonstrated in many studies oxidizing agents and inflammatory mediators share in a vicious circle in which oxidants stimulate secretion of inflammatory mediators by macrophages and airway epithelial cells and these mediators in turn provoke gene expression to transcript and produce more inflammatory mediators resulting in enhanced inflammation (61,62). Oxidative stress is responsible for initiating of this defected circle because antioxidants are able to abolish it completely.

In another view DNA-Histone structural rearrangement by oxidizing agents like cigarette smoking, as shown by Anderson et al. study, can explain how gene expression is prominently undergone producing inflammatory mediators in oxidative stress condition (63). In normal condition DNA

is tightly twisted around histones axes, not accessible for transcription by RNA polymerase and NF- $\kappa$ B. Histone acetylation can change electrical charge and therefore modifies histone structure leading to untwist of DNA, providing more space and freedom for transcriptional factors for ending gene silencing and beginning to produce inflammatory mediators. Whole of this process is thought to be redox-sensitive, can be initiated by cigarette smoking exposure and can be reversed by antioxidants (63).

Histone deacetylase enzymes (HDAC) director of histone deacetylation are found with lower activity in cigarette smoking exposed animals and in retrieved macrophages from COPD patients, conversely acetylated histones are in higher levels in these animal models and COPD patients reflecting a redox-sensitive enzymatic regulation (64-66). Additionally a vice versa correlation between HDAC activity and FEV1 was proved (67).

Resistance to corticosteroids in COPD patients may be due to lower activity of HDAC, a fact if confirmed, HDAC therapeutic application could be reasonable (68). An alternative to reveal associated intervention of inflammation and oxidative stress in COPD formation is substantiated by E1A, a derivative protein from concealed adenoviral infection. This protein activate nuclear cofactors such as NF- $\kappa$ B, have essential role in gene expressing and enhancing inflammatory mediator releasing in response to oxidative agents like cigarette smoking (69).

It seems that E1A facilitates inflammatory processes in response to oxidative stress by multiplying production of inflammatory mediators like IL-8, thereby making more predisposition to form COPD in smoking subjects (70-72). This can also explain how latent viral infection plays role in pathogenesis of COPD.

Furthermore oxidative stress may lead to lower activity of vascular endothelial growth factor (VEGF) R2 receptors, consequently emphysema develops in human lungs and antioxidants as protective agents which can prevent from VEGF downregulation (73,74). In addition to animal studies neutralization of the antioxidant gene activators like Nrf2, predisposes the experimental mice to develop emphysema more and more probable than in wild mice in contact with cigarette smoking (75).

Gluthation antioxidant system plays an important role as a protective agent against oxidative stress. Its changes are regulated by genes expressing whenever faced with oxidative agents like cigarette smoking (76-81).

In addition, mice with lower capability to induce gene expressing for antioxidant production are more susceptible to develop emphysema compare to normal races in chronic exposure to cigarette smoking (82).

### ***Systemic oxidative stress***

Clinical manifestations in COPD patients such as cachexia, weight loss, and increased cardiac accidents as well as subclinical changes including increasing peripheral neutrophil releasing ROS, prominent decrease of plasma level of antioxidant capacity, increased lipid peroxidation products in plasma and eventually increased level of nitrotyrosine, are evident that oxidative stress has its own systemic influences on other organs in additive to pulmonary system, (83-85) due to passing through respiratory system to general circulation and spread out their effects to many other vital organs (86).

Scanning of related published data in recent decade revealed that researchers are interested in a few categories in oxidative stress in COPD era, including more basic studies to prove oxidative stress role in

COPD pathogenesis, new therapeutic strategies (both described previously), the role of dietary supplementation in COPD alleviation and evaluation of new marker which could be more accurate and less invasive. All the engaged basic studies have presented evidences by which the role of oxidant/antioxidant imbalance is established convincingly, nevertheless increasing sample size and meta analytic studies can produce more accuracy in future studies (87-90).

Based on approved role of oxidative stress in COPD, many studies conducted to evaluate therapeutic effect of antioxidants in COPD treatment. According to some of these studies antioxidant agents not only prevent direct damages of oxidative agents but also may subside the signaling pathways leading to inflammation (10). A common instance is N-acetylcysteine, a drug used widely as oral, inhalation and intravenous forms (91).

Another trial studied the efficacy of a therapeutic antioxidant group with beneficial effects belong to super oxide dismutase (SOD) compounds. Synthetic SOD drugs investigated in plenty of animal and human studies, improve the clinical and paraclinical condition of patients significantly. EUKs, M40403, MnTBAP are instances of latter group (92-95).

Many other surveys focused on the role of antioxidant vitamins A, C, D and E in COPD patients, although results are somewhat inconsistent about the real effect of these vitamins (specially vitamin E) on symptoms and FEV1 improvement, proportional benefits from added supplementary vitamins are mentioned for COPD patients and cigarette smokers (96,97).

## Conclusion

Current survey has provided plenty of evidences to confirm existence of

oxidant/antioxidant imbalance in COPD developing. Evidences are accumulated to show a central role of this imbalance in pathogenesis of COPD.

Through diverse mechanisms such as upregulation of redox-sensitive transcription factors and then pro-inflammatory gene expression, stress oxidative can cause an inflammatory response to cigarette smoking in respiratory system as well as other vital organs. In other hand antioxidant gene induction can be stimulated by oxidative stress also. According to these facts future studies should be focused on finding new therapeutic strategies including drugs which regulate oxidants and antioxidant gene expression or development of potential antioxidant drugs.

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## Conflict of Interest

The authors declare no conflict of interest.

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