Eur J Med Res (2010) 15(Suppl. II): 27-35 

Review

# Role of Elastases in the Pathogenesis of Chronic Obstructive Pulmonary Disease: Implications for Treatment

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**Abstract**
Neutrophil elastase, metalloproteinases, and their inhibitors play an important role in the development of chronic obstructive pulmonary disease (COPD), resulting in extensive tissue damage and malfunctioning of the airways. Nearly fifty years after the protease-antiprotease imbalance hypothesis has been suggested for the cause of emphysema, it is still appealing, but it does not explain the considerable variation in the clinical expressions of emphysema. However, there are many recent research findings to support the imbalance hypothesis as will be shown in this review. Although limited, there might be openings for the treatment of the disease.

*Key words:* antiproteinases, COPD, metalloproteinases, neutrophil elastase, pathogenesis, treatment

## Chronic Obstructive Pulmonary Disease

Chronic obstructive pulmonary disease (COPD) is one of the major health care problems in the present world. The global prevalence of COPD in adults 40 years or older is approximately 9-10% and is higher in smokers than in non-smokers, and higher in men than in women (Halbert et al 2006). COPD is an important cause of death in many countries and the incidence is still increasing because of the expanding epidemic of smoking and the increasingly aging population (Chapman et al 2006). Because of its increasing incidence, the World Health Organization (WHO) in collaboration with the US National Institutes of Health formed the Global Initiative for chronic obstructive lung disease (GOLD). The GOLD definition describes COPD as 'A disease state characterized by airflow limitation that is not fully reversible, and that is usually both progressive and associated with an abnormal inflammatory response of the lungs to noxious particles or gases' (Pauwels et al 2001). Classically, COPD involves the three morphological forms chronic bronchitis, emphysema, and small airway disease. However, these pathologic entities can be present in mixed forms in the same patient (Jeffery 2001).Chronic bronchitis is characterized by cough with expectoration due to mucus hypersecretion, which does not always lead to airway obstruction. Goblet cell hyperplasia is observed in the bronchial wall, but the excessive mucus production correlates poorly with the mucus gland mass (Yoshida and Tuder 2007). Emphysema is characterized by a permanent air space enlargement due to a process of alveolar destruction and is not associated with significant fibrosis (Snider et al 1985, 1986). Damage to the alveolar wall and attachment destruction leads to the loss of elastic recoil. There are two major types of emphysema, according to the distribution within the acinus: centrolobular and panlobular. The centrolobular form involves dilatation and destruction of the respiratory bronchioles, while the panlobular form of emphysema involves the destruction of the whole of the acinus. The former is the most common type of emphysema in COPD and is more prominent in the upper zones, while the latter predominates in patients with $\alpha_1$-antitrypsin deficiency and is more prominent in the lower zones.

## COPD and Inflammation

All these morphological forms of COPD, chronic bronchitis, small airway disease, and emphysema are accompanied by airway inflammation. The inflammatory cell profile in the alveolar walls and the air spaces is similar to that described in the airways and persists throughout the course of the disease (Finkelstein et al 1995). An increase in neutrophils, macrophages, and T-lymphocytes in various parts of the lung is characteristic and relates to the degree of airflow limitation (Saetta et al 1998). There may be an increase in eosinophils in some patients as well, particularly during exacerbations (Saetta et al 1994, 1996). These inflammatory cells are capable of releasing a variety of cytokines and inflammatory mediators. In addition, the airway epithelium is a rich source of cytokines and chemokines that recruit both neutrophils and macrophages into the airspaces. Many of these cytokines are overexpressed in COPD (Chung 2001; MacNee 2007). The pro-inflammatory cytokines IL-1$\beta$ and TNF-$\alpha$ are released by airway epithelial cells during inflammatory reactions induced by infection, injury, or smoking cigarettes. Both cytokines share biological functions through some common signal transduction pathways (Stewart and Marsden 1995). The expression of metalloproteinases and other enzymes involved in the degradation of connective tissue proteins is stimulated by IL-1$\beta$ in close connection with TNF-$\alpha$ (Cao et al 1996; Churg et al 2002, 2003a, 2004; Kusano et al 1998). The neutrophils release a large array of serine proteases including elastase, proteinase-3 and cathepsin G, all are able to induce emphysema in

animal models (Stockley 1983) by destroying the elastin and components of the alveolar wall (Saetta et al 2001). It has been shown that during cardiopulmonary bypass surgery increased levels of plasma elastase and metalloproteinases (MMPs) occur, which may cause pulmonary injury (De Backer et al 1996; Steinberg et al 2001). Neutrophils may also play a role in the mucus hypersecretion, which is characteristic for chronic bronchitis. All neutrophil proteases are potent secretagogues (Nadel 1991; Sommerhoff et al 1990; Witko-Sarsat et al 1999). Elastase is also a very potent inducer of mucus gland hyperplasia (Sommerhoff et al 1990).

## Proteases and their Inhibitors

Elastase is a serine proteinase with a primary translational product of 267 amino acids and variable glycosylation, migrating with a molecular mass of 28 to 31 kiloDalton (kDa) (Sinha et al 1987). The N-terminal amino acid sequence is strongly homologous with that of pancreatic elastase. Interestingly, the pancreatic elastase is generated from pro-elastase by tryptic cleavage of an N-terminal activation peptide, whereas no pro-enzyme has been detected for the elastase (Barrett 1981). Elastase is synthesized in the promyelocytic stage of myeloid development and stored in large quantities in its active form in neutrophil azurophilic granules (Takahashi et al 1988). Elastase has potent catalytic activity against a broad array of extracellular matrix substances, such as elastin (Senior et al 1976), cartilage proteoglycan (Roughley and Barrett 1977), collagen types I and II (Starkey et al 1977), type III (Gadek et al 1980), type IV (Davies et al 1978), and fibronectin (McDonald and Kelley 1980). Both neutrophils and macrophages are also able to produce metalloproteinases, which are likely to contribute to injury of the lung. Cigarette smoke exposure induces either directly or indirectly the production of MMP-1 (collagenase), MMP-9 (gelatinase B), MMP-12 (macrophage elastase), and other metalloproteinases by macrophages (Shapiro 1999). A deregulated expression of these metalloproteinases could lead to the lung destruction characteristic of emphysema. MMP-12 is a 54 kDa proenzyme that is processed by loss of both N- and C-terminal residues into a 40 kDa and then to a 22 kDa active form. It has been suggested that MMP-12 gene polymorphisms may account for the activity of MMP-12 in various diseases and that it is one of the causative factors in smoking related lung injury (Belvisi and Bottomley 2003). In healthy humans, the lung is protected by a variety of antiproteases that quickly inactivate free proteases. The major antiproteases which are also involved in the pathogenesis of COPD include $\alpha_1$-antitrypsin, $\alpha_1$-antichymotrypsin, secretory leukocyte proteinase inhibitor (SLPI), elafin, cystatins and tissue inhibitors of metalloproteinases (TIMP), including TIMP-1, TIMP-2, and TIMP-3. In the lung, alpha-1-antitrypsin (A1AT), also known as serum trypsin inhibitor, is the most predominant antiprotease. A1AT is a single chain glycoprotein consisting of 394 amino acids. A1AT belongs to the family of serine protease inhibitors, known as serpins. A1AT has the ability to inhibit a wide variety of proteases, but it has the highest affinity for elastase. Normal physiological concentrations of A1AT in adults' serum ranges from 1.5-3.5 g/l, but the concentration will rise many folds upon acute inflammation. This up-regulation is required to balance the secretion of elastase by neutrophilic granulocytes. Also during infection, cancer and pregnancy an increase in serum concentration of A1AT occurs. SLPI has a lower association rate constant for elastase than A1AT and is partially reversible, meaning that SLPI is not equipotent to A1AT, although it does inhibit on a one-to-one molar basis. Elafin has a similar affinity to elastase as A1AT, but both SLPI and elafin are present at lower concentrations than A1AT (Gettins 2002; Williams et al 2006).

## Alpha-1-antitrypsin Deficiency

Deficiency of A1AT, a rare genetic condition with autosomal co-dominant inheritance (Köhnlein and Welte 2008), was first recognized in 1963 (Laurell and Eriksson 1963) and classically presents with emphysema in young- to middle-aged adults. They described an association between familial emphysema and deficiency of A1AT. Shortly thereafter Fagerholm described the allelic variation of A1AT (Fagerholm and Laurell 1967). Two mutations in the A1AT gene have been associated with decreased A1AT serum levels (Siafakas and Tzortzaki 2002). Other researchers proposed that A1AT deficiency is one of the most common hereditary disorders in the world (de Serres 2002). In total, there are at least 120 million carriers of defective alleles, which are populations at risk for emphysema development and 3.4 million carriers of deficiency allele combinations who may eventually develop COPD. These individuals with A1AT deficiency only account for a small proportion of the total COPD burden. Much more than 70 different variants are recognized and they are classified according to their proteinase inhibitor (Pi) properties (Brantly et al 1998). The most common phenotype presenting with clinical evidence of A1AT deficiency is PiZZ or Znull. The serum concentration of A1AT in patients with genotype PiZ is about 10-15% of normal concentrations. Acquired deficiency is much more common and occurs because of the inactivation of a methionine residue in the active site of A1AT by free oxygen radicals and substances present in cigarette smoke. A1AT is an acute-phase reactant with anti-protease properties mainly synthesized by liver cells and the Z-variant of A1AT is retained as inclusion bodies in the endoplasmic reticulum of hepatocytes (Sandford and Silverman 2002). Thus A1AT deficiency is merely a failure in the secretion rather than in the biosynthesis of A1AT that leads to the deficiency (Crystal 1989; Luisetti and Seersholm 2004). Vitamin D-binding protein (VDBP, also known as a group-specific component or Gc-globulin) is the main systemic transporter of $1,25(OH)_2$ vitamin D3 and is essential for its cellular endocytosis (Ongagna et al 2005). It is a highly expressed polymorphic protein that is a precursor to inflammation-primed macrophage activating factor (GcMAF) (Nagasawa et al 2004) and can significantly enhance leukocyte chemotactic activity to C5a and C5a des-Arg (Zhang and Kew 2004). Thus, it can regulate the inflammatory

response or diminish antioxidative capacity of the host. A decreased frequency of 2-2 genotype of VDBP was reported in COPD patients (Zhang and Kew 2004), but this was not replicated (Kueppers et al 1977a, 1977b). In the last 10 years, many publications reviewed the genetic profile of COPD (Hoidal 2001; Sampsonas et al 2006; Sandford et al 2002; Sandford and Silverman 2002; Snider 2003). These data indicate that in addition to A1AT deficiency, other forms of genetic susceptibility also play a crucial role in COPD development. Alpha-2-macroglobulin is a protease inhibitor, and its serum deficiency is very rare. There are only case reports of COPD patients and $\alpha_2$-macroglobulin polymorphism (Poller et al 1989).

## The Imbalance Hypothesis

The 'protease-antiprotease imbalance' hypothesis for the development of emphysema has been postulated more than forty years ago (Eriksson 2008), who demonstrated for the first time the existence of A1AT deficiency (Laurell and Eriksson 1963), and this hypothesis is still supported. The hypothesis is appealing but does not explain the considerable variation in the clinical expression of emphysema in individuals who smoke cigarettes, as is shown in this review. Only 10-15% of A1AT-deficient individuals (Burdon et al 1996) and 10-20% of cigarette smokers develop emphysema (Postma and Boezen 2004; Tetley 1993). Several lines of evidence are suggestive for the validity of the hypothesis: 1. instillation of elastase can induce emphysema in the lungs of animals (Gross et al 1965; Janoff et al 1977; Snider et al 1984); 2. destruction of elastins by elastase and the loss of elastic recoil (Campbell et al 1987; Shapiro et al 2003); and 3. genetic deficiency of $\alpha$1-proteinase inhibitor increases the susceptibility to the development of emphysema (Laurell and Eriksson 1963; Stoller and Aboussouan 2005). Together with the fact that proteolytic enzymes produce many of the clinical and pathological features of COPD, these observations firmly establish the concept that enzyme activity and tissue damage in emphysema are the result of excess release of proteinases, or a reduction in the levels of inhibitors required to control them.

## Secretory Leukocyte Proteinase Inhibitor

Since most of the other anti-proteases, such as SLPI and elafin, are considerably smaller molecules than A1AT, these molecules are thought to be active in areas such as intercellular spaces that are not accessible to the larger A1AT molecule (Ayad et al 2003; Vogelmeier et al 1991). However, even very small molecular weight inhibitors are no better than A1AT in studies looking at the process of quantum proteolysis (Campbell et al 1999). SLPI has a major benefit that relates to the fact that it can inhibit elastase bound to connective tissue substrate, whereas A1AT is not very good at this. This gives an impression that SLPI actually gets under the cell, whereas it is a very different mechanism. Although SLPI, like A1AT, is inactivated by cigarette smoke-induced oxidation, oxidized SLPI maintains its anti-proteinase activity particularly at high concentrations (Boudier and Bieth 1994). It has been proposed that SLPI is important for protecting the respiratory epithelium because of its location in the bronchi (Ohlsson and Tegner 1976; Tegner 1978). In contrast to A1AT, SLPI blocks elastase in the alveolar walls (Bruch and Bieth 1986). Ayad et al (2003) demonstrated higher concentrations of SLPI in serum and BAL fluid in A1AT deficient individuals as compared to normal PiM subjects. They speculated that there may be some genetic, or other, compensatory processes leading to an increase in SLPI in A1AT deficient individuals. SLPI can protect the lung against the development of emphysema in A1AT deficient subjects and cigarette smokers have lower SLPI concentrations than never-smokers, presumably due to irreversible bronchial epithelial damage. The relationship may be false, because A1AT deficient subjects may have more mucus hypersecretion, which could raise the SLPI level above that of individuals who have no mucus hypersecretion. However, in studies reported by Hill et al (1999, 2000), having appropriate controls between deficient and non-deficient subjects, it was clear that SLPI was actually lower and this is consistent with an effect of proteolytic enzymes, which cause a reduction in SLPI secretion from airway cells. It is also consistent with the reduction in SLPI that occurs during acute exacerbations when elastase activity rises. In contrast, Hollander et al (2007) could not demonstrate increased levels of SLPI and $\alpha_1$-1-antichymotrypsin in plasma of A1AT deficient individuals. Their argument is that increased protease inhibitor levels in plasma occur at young age, and decrease to normal levels with increasing age. This is supported by some reports (Ganrot 1968; Tunstall et al 1975). They also question if plasma is the relevant biological fluid to measure SLPI, since SLPI is produced locally in the airways and is regulated by various pro-inflammatory stimuli. Other serine protease inhibitors, such as $\alpha$-1-antichymotrypsin, $\alpha$-2-macroglobulin, anti-thrombin, and anti-plasmin play also important roles in controlling serine protease activity. While these inhibitors are produced mainly by the liver and reach the other tissues by passive diffusion (Kalsheker et al 2002; Morrison et al 1987; Vignola et al 1998), SLPI and elafin are produced locally by airway epithelial cells (Sallenave et al 1994, 1997). Thus, it cannot be excluded that under inflammatory conditions the compensatory increase in SLPI and/or other protease inhibitors may reduce the severity of A1AT deficiency. SLPI is considered equipotent to A1AT as an airway anti-proteinase. DNA derived from individuals with a broad range of A1AT mutations, many of which occur in high frequency, and analyzed for SLPI mutations has been found to have no polymorphisms in the major SLPI coding exons (Abe et al 1991). In addition, no polymorphisms were detected in individuals who had obstructive lung disease at an early age, but did not have A1AT deficiency or cystic fibrosis, i.e., hypothetically those who had an increased chance of having an abnormal SLPI gene that might be responsible for their disease (Abe et al 1991; Chang et al 2006).These data suggest that structural mutations in SLPI may not be involved in the pathogenesis of COPD (Ning et al 2004).

## Metalloproteases and Elastase

One of the most prominent families of proteases cleaved by elastase is the group of matrix metalloproteases. Serine proteases such as elastase, cathepsin G, and proteinase-3 have been shown to activate MMP-2, and this activation could be blocked by $\alpha_1$-antitrypsin, but not by an MMP-inhibitor (Shamamian et al 2001). Geraghty et al (2007) have demonstrated that elastase could induce cathepsin G and MMP-2 expression and activity in macrophages. Their study provides data that elastase presides over a novel hierarchy in protease regulation, causing tissue destruction in diseases associated with a high burden of elastase. The up-regulation and release of cathepsin B by elastase is an interesting observation. Zheng et al (2000) were the first to demonstrate that cathepsins are released in response to cigarette smoke. Mediators that generate Th2-type tissue inflammatory responses are also able to activate proteolytic pathways. By using an overexpressing transgenic modelling system to target IL-13 to the murine lung, they were able to induce a phenotype that is equal to human COPD with prominent emphysema, enlarged lungs, enhanced pulmonary compliance, mucus metaplasia, and a mixed tissue inflammation. The cathepsins, but also metalloproteinases were induced by IL-13 and it has been suggested that IL-13 is an important regulator of the protease/antiprotease balance contributing to the pathogenesis of COPD. Earlier it was demonstrated that MMP-12 is essential for cigarette smoke-induced inflammation and emphysema in the murine lung (Hautamaki et al 1997) and that MMP-12 expression is enhanced in alveolar macrophages from smokers and patients with emphysema (Shapiro 2000).

Zheng et al (2000) have also studied the effects of metalloproteinase and cathepsin proteinase inhibitors on the outcome of IL-13 induced emphysema. These experiments show that these antagonists partially abrogated the IL-13-induced emphysema and that they were more effective at reducing lung volume and alveolar destruction when used in combination. Levels of a variety of important antiproteases in the lungs of IL-13 overexpressing animals, such as TIMP-2, TIMP-3, TIMP-4, SLPI, and cystatin C, were not changed. An increased expression of TIMP-1 was noted as being not of sufficient magnitude to inhibit the induced emphysema. However, the expression of A1AT was decreased. The biological significance of the latter observations is questionable because the level of A1AT produced by the liver is exceeding that of lung-derived A1AT. When cathepsin B is converted into an active form by elastase (Burnett et al 1995), it is able to inactivate important respiratory tract innate immune proteins such as SLPI, human β-defensins 2 and 3, and lactoferrin (Geraghty et al 2007; Rogan et al 2004; Taggart et al 2003). In that way, elastase-induced cathepsin B release may not only lead to degradation of the extracellular matrix, it will also have an impact on important antimicrobial peptides and proteins.

Although tobacco smoking is an important inducer of COPD, it is not the sole course, because other causes are abnormal recruitment and activation of inflammatory cells and excess levels of free radicals. Nevertheless, smoking-induced emphysema both in humans and in animal models results from enzymes derived from alveolar macrophages, more precisely the metalloproteinases (MacNee 2007). Mice deficient in macrophage elastase (MMP-12) and rats depleted of macrophages did not develop emphysema after long-term smoke exposure (Hautamaki et al 1997; Ofulue and Ko 1999). However, neutrophils also secrete cathepsin and MMP-8 and MMP-9 beside elastase. Several studies still confirm the importance of the neutrophil in emphysema (Churg et al 2003a; Shapiro et al 2003). There is a considerable interaction between the metalloproteinases, with MMP-1 activating MMP-2, MMP-2 activating MMP-1 and MMP-12, and MMP-13 activating MMP-9 (McCawley and Matrisian 2001; Wright and Churg 2007). Which metalloproteinases are actually important in emphysema is a controversial issue. Increased levels of MMP-1, MMP-9, and MMP-12 have been found in the lungs of emphysema patients (Demedts et al 2006; Finlay et al 1997; Imai et al 2001; Molet et al 2005; Ohnishi et al 1998; Russell et al 2002). Thus, collagenases, gelatinases, and elastases appear to be important in emphysema, but the different metalloproteinases may play different roles in animals compared to humans. TNF-α serves as a central mediator. It activates a proteinase cascade and the mediator itself is activated by proteinases. Macrophage-derived MMP-12 is able to liberate TNF-α from the surface of the macrophage, resulting in infiltration of neutrophils by activation of endothelial cells. The migrated neutrophils, in turn, release elastase upon TNF-α stimulation. Moreover, MMP-12 and the neutrophil-derived elastase potentiate the action of each other by inactivating A1AT and TIMP-1, respectively (Shapiro et al 2003). In a recent study, it has been shown that the number of MMP-12 positive macrophages in lavage fluid of smokers with COPD was higher compared with ex-smokers with COPD, healthy smokers, and healthy never-smokers (Babusyte et al 2007). A similar result was observed for MMP-12 positive macrophages in induced sputum from these patients. Although the number of macrophages in induced sputum was not significantly higher in smokers with COPD compared with the ex-smokers with COPD, several studies show that MMP-12 may indeed play a role in COPD in humans. The expression of MMP-12 in healthy smokers is increased compared with non-smokers, supporting the suggestion that smoking may increase the expression of this enzyme (Babusyte et al 2007; Demedts et al 2006; Molet et al 2005). The contribution of MMP-12 to smoke-induced emphysema is probably enhanced by indirect effects, such as inactivation of A1AT (Gronski, Jr. et al 1997), and MMP-12 mediated recruitment of neutrophils to the lung (Churg et al 2003a). Otherwise, MMP-12 may accumulate and would not rapidly decreases or become inactivated after smoking cessation, exaggerating a persistent inflammation. In a very recent paper, Lowrey et al (2008) have described that MMP-9 protein, but not metalloproteinase activity, was higher in sputum of COPD patients compared with smoking controls. They suggest that MMP-9 levels may not reflect the overall metalloproteinase activity in the airways of COPD patients, reflecting a

complex relationship between MMP-9 levels and activity.

Another striking finding is that A1AT administration to a model of cigarette smoke exposed mice is able to suppress smoking-induced increases in serum TNF-α, and a decreased inflammatory cell influx into the lung (Churg et al 2003b). Recent work has shown that A1AT inhibits lipopolysaccharide-mediated increases in TNF-α and IL-1β release from human blood monocytes (Janciauskiene et al 2004, 2007; Nita et al 2005). Similarly, administration of A1AT to patients with deficiency of the anti-proteinase decreased sputum elastase activity, and sputum levels of leukotrienes B4 (Stockley et al 2002). In addition, A1AT can inhibit both thrombin and plasmin, preventing release of MMP-12 and TNF-α (Churg et al 2007). Important in this case is also the enhancing effect of A1AT on LPS-stimulated IL-10 generation, because it influences a specific mechanism for the effects of A1AT rather than a general depressive effect of A1AT on cell function (Janciauskiene et al 2004). Thus, we might say that A1AT not only is a protease inhibitor, it also exhibits anti-inflammatory properties.

## Implications for Treatment

Medical treatment of elastase-induced emphysema is similar to the treatment of general COPD and should be tailored to each individual patient. The targets of treatment are the prevention of accelerated decline of pulmonary function, reduction of lung infections, and improvements in exercise capacity. However, none of the existing pharmaceutical treatments for COPD has been shown to modify the long-term decline in lung function. Therefore, pharmacotherapy is used to decrease symptoms and/or complications (Chrystyn et al 1988; Gross et al 1989; Higgins et al 1991; Vathenen et al 1988). Pharmaceutical treatment consists of application of long-acting $\beta_2$-agonists, formoterol or salmeterol. The combination of $\beta_2$-agonists with long-acting anticholinergic tiotropium or theophylline may provide additional improvement in lung function and health status (Guyatt et al 1987; The COMBIVENT Inhalation Solution Study Group 1997). Short-acting $\beta_2$-agonists may only provide relief in acute situations. The mucolytic and antioxidant N-acetylcysteine has proven itself as a mucolytic, but its anti-inflammatory properties are rather limited (Decramer et al 2005; Petty 1990; van Overveld et al 2005). The benefit of inhaled corticosteroids is widely debated. Oxygen therapy enables many patients with severe COPD to lead a more normal life and increase survival (Nocturnal Oxygen Therapy Trial Group 1980; Report of the Medical Research Council Working Party 1981). Physical exercise will improve breathing ability by relieving dyspnea and fatigue (Berry et al 1999). Surgical approaches to improving dyspnea by removing areas of major lung damage from emphysema are only effective in a very small and carefully selected part of the patient population (Mehran and Deslauriers 1995; Naunheim et al 2006). Finally, substitution therapy with human A1AT in non-smoking patients with low A1AT serum levels (<0.8 g/l) is performed for about ten years in a small number of countries. In addition to the high cost of this therapy, the evidence for its efficacy is limited (Köhnlein and Welte 2008). The development of elastase inhibitors has significantly increased our knowledge. In the last 15 years, elastase inhibitors have been used in many animal studies. Among the inhibitors are chloromethylketone, ICI-200,355, L-658,758, ONO-5046, and SC-39026. However, SLPI is easy to produce synthetically. In addition, when delivered by aerosol SLPI is retained in the epithelial lining fluid of the lungs with a sufficient long half-life of 12 hours. A third advantage is the insensitivity to MMP-8, a factor generally abundant at sites with high elastase concentrations (Fitch et al 2006). Delivery of exogenous proteinase inhibitors might thus have many advantages in the treatment of COPD, and SLPI seems to be a promising candidate.

## Conclusions

In general, we might say that the major proteinases involved in the pathogenesis of COPD include those produced by neutrophils (elastase, cathepsin G and proteinase-3) and macrophages (cathepsins), and various metalloproteinases. The major anti-proteinases involved in the pathogenesis of COPD include alpha-1-antitrypsin, secretory leukocyte proteinase inhibitor, and the tissue inhibitors of MMPs. As soon as the fragile equilibrium is disturbed by external or internal factors, the individual is prone to the development of chronic and destructive lung disease, leading to disability, and ultimately death.

*Conflicts of interest:* The authors declare no conflict of interest in relation to this article.

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