



# LUND UNIVERSITY

## Extracellular Matrix Remodelling In COPD

Westergren-Thorsson, Gunilla; Bjermer, Leif; Hallgren, Oskar

*Published in:*  
European Medical Journal

2014

*Document Version:*  
Publisher's PDF, also known as Version of record

[Link to publication](#)

*Citation for published version (APA):*  
Westergren-Thorsson, G., Bjermer, L., & Hallgren, O. (2014). Extracellular Matrix Remodelling In COPD. *European Medical Journal*, 1-6.

*Total number of authors:*  
3

### General rights

Unless other specific re-use rights are stated the following general rights apply:  
Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal

Read more about Creative commons licenses: <https://creativecommons.org/licenses/>

### Take down policy

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

LUND UNIVERSITY

PO Box 117  
221 00 Lund  
+46 46-222 00 00

# EXTRACELLULAR MATRIX REMODELLING IN COPD

Gunilla Westergren-Thorsson,<sup>1</sup> Leif Bjermer,<sup>2</sup> \*Oskar Hallgren<sup>1,2</sup>

1. Department of Experimental Medical Science, Lund University, Lund, Sweden

2. Department of Respiratory Medicine and Allergology, Skåne University Hospital, Lund, Sweden

\*Correspondence to oskar.hallgren@med.lu.se

**Disclosure:** No potential conflict of interest.

## ABSTRACT

Chronic obstructive pulmonary disease (COPD) is a chronic inflammatory disease that involves major remodelling of cells and tissue in the lung. There has been much focus on the inflammatory response, while less attention has been paid to structural changes and alterations of the extracellular matrix (ECM). In this review we revisit some of the latest findings on what is currently known about ECM alterations in COPD. We also discuss mechanisms involved in tissue repair and pay extra attention to the potential role of glycosaminoglycans in the disease.

**Keywords:** Chronic obstructive pulmonary disease (COPD), airway remodelling, extracellular matrix, collagen, proteoglycan, glycosaminoglycan, matrix metalloproteinase.

## INTRODUCTION

Chronic obstructive pulmonary disease (COPD) is characterised by a slowly progressive development of airflow limitations that have been considered to be poorly reversible.<sup>1</sup> This dogma has been questioned in several studies both in patients with mild disease and moderate-to-severe disease, and so has the assumption that the use of inhaled bronchodilators to measure improvement in forced expiratory volume in 1 second can differentiate COPD patients from asthma patients.<sup>2-4</sup> However, the airflow limitations in COPD are caused by loss of elastic recoil, degradation of alveolar walls (i.e. emphysema), and increased resistance in conducting airways. It is also associated with a wide range of phenotypical changes of structural cells and extracellular matrix (ECM) alterations. The major risk factor for developing COPD in developed countries is cigarette smoking although there are also other risk factors, including air pollution, exposure to indoor burning fuels, and occupational exposure.<sup>5</sup> In the lung, repeated exposure to noxious particles, such as cigarette smoke, has been suggested to trigger an immunological response elicited by the epithelial lining.<sup>6</sup> Epithelial cells signal to leukocytes and underlying mesenchymal cells that are recruited and activated; this starts a repair process. During normal tissue repair, the

process is terminated when the initial trigger is removed; however, when the trigger remains, as is the case in smokers that are continuously exposed to cigarette smoke, the process may become pathological.<sup>7-9</sup> However, different repair processes may be active in different parts of the lung as the lung architecture, including the ECM scaffold, differs very much throughout the airway tree. This hypothesis was confirmed by Churg et al.<sup>10</sup> who suggested that genes involved in tissue repair are differentially expressed in small airways and in the lung parenchyma following exposure to cigarette smoke. Genes that promoted ECM production and deposition were upregulated in small airways but were at baseline levels or downregulated in the parenchyma. There may also be a difference between central airways and the parenchyma, as several studies have identified phenotypically unique sub-populations of fibroblasts, a cell type that is a key player in tissue repair, in central airways, and in the parenchyma.<sup>11-14</sup>

Moreover, in parallel with the structural remodelling of the airway tree and the subsequent airflow limitations, many patients also have remodelling of the lung vasculature, which often results in pulmonary arterial hypertension (PAH).<sup>15</sup> These two processes are very tightly connected and it has been shown that vascular remodelling in COPD patients is similar to what is observed in high-

altitude natives, which indicates that alveolar hypoxia can be one cause of these changes.<sup>16</sup> In addition, the degree of hypoxaemia has been shown to be related to the increased pulmonary arterial pressure and vascular resistance in COPD patients.<sup>17</sup> Although it is very probable that vascular and airway remodelling are two sides of the same coin, less attention has been paid to ECM changes in lung vasculature in COPD patients. Studies on the interplay between these two processes will therefore be important to better understand the complexity of the disease. The subject of vascular remodelling in COPD is beyond the scope of this text and has been reviewed elsewhere.<sup>15</sup>

The turnover of ECM is influenced by two processes: production and degradation. Mesenchymal cells, such as fibroblasts and smooth muscle cells, are the bulk producers of ECM components, but other cell types also contribute to the production of more specialised ECM molecules, including components of basement membranes. Moreover, the degradation of the ECM is mainly dictated by the delicate balance between proteases and anti-proteases, and especially matrix metalloproteinases (MMPs) and their natural inhibitors, tissue inhibitor of metalloproteinases (TIMP).<sup>18</sup> Since the discovery that lack of the protease inhibitor  $\alpha$ 1-antitrypsin may cause emphysema, many efforts have been focused on the influence of these molecules on the disease.<sup>19</sup> Clinical trials and animal models that target proteases and MMPs have so far been less successful in limiting the clinical manifestations of the disease.<sup>20</sup>

There have been many studies with the aim to characterise structural remodelling and ECM alterations in COPD, but many of the data generated in these papers are not consistent. One reason for this is that COPD is a very heterogeneous disease with varying degrees of emphysema, fibrosis, and small airway involvement. To further understand the dynamics of structural remodelling in COPD it is therefore crucial to further phenotype patients in the studies. Another source of variability is that immunohistochemistry has been used for identification and quantification of the ECM proteins in many studies. Since different antibodies have different binding capacity to antigens, and the preparation of the tissue may vary between different laboratories, this may contribute to the variability between studies. Herein we review some of the latest findings on what is currently known about ECM alterations in COPD.

ECM consists of a large number of fibrous proteins, including collagens and elastin, but also glycoproteins (GPs) and proteoglycans (PGs). Fibrous collagens, such as collagen Type 1 and 3 form rope-like fibril bundles that give the tissue stability and strength. Elastin, the most abundant protein in elastic fibres, contributes to the elastic properties of the lung. Other important players of the ECM are the PGs that are found in the ECM, in basement membranes and on cell membranes. They have long unbranched polysaccharide side-chains, glycosaminoglycan (GAG), attached to the core protein.<sup>21,22</sup> GAG chains are negatively charged at neutral pH due to uronic acid groups and varying amounts of sulphate bound to the polymer which makes them very bioactive. However, it is not only the degree of sulphation, but also the location of sulphate groups that dictates the bioactivity of GAGs.

Depending on the content of the polysaccharide chain there are several types of GAGs: heparan sulphate (HS), heparin, chondroitin sulphate (CS), dermatan sulphate (DS), keratan sulphate (KS), and hyaluronic acid (HA).<sup>23</sup> They serve as a tissue reservoir that binds and immobilises growth factors, cytokines, and proteases that can be released and form local gradients within the tissue, and may contribute to diverse functions such as chemotaxis, proliferation, and differentiation. Due to their negative charge, they bind water and thereby influence the viscosity of the tissue.<sup>24,25</sup> Moreover, GAGs have been shown to interact with and influence the bioactivity of several mediators involved in the COPD pathogenesis. Growth factors involved in tissue repair that may induce fibroblast differentiation and altered production of ECM proteins, including tumour growth factor-beta, connective tissue growth factor, fibroblast growth factor, hepatocyte growth factor, and platelet-derived growth factor, have been shown to bind to GAGs.<sup>26-28</sup>

In COPD, activated epithelial cells and macrophages release chemotactic mediators such as tumour necrosis factor-alpha, chemokine (C-C motif) ligand 2, chemokine (C-X-C motif) ligand 1 (CXCL1), CXCL8, CXCL9, CXCL10, and CXCL11 that form gradients to attract leukocytes and lymphocytes to the site of inflammation. To retain gradients and protect mediators from proteolysis they readily bind to HS and/or CS/DS-GAGs in the

ECM.<sup>29-32</sup> Importantly, this interaction depends on the disaccharide composition within the GAG sequence in addition to merely the charge. GAGs also bind and regulate the activity of many extracellular proteases. One example is HS which has been demonstrated to be a natural inhibitor of neutrophil elastase.<sup>33</sup> Furthermore, the activity of MMP-2 and MMP-9 has been shown to be influenced by GAG. While some GAGs - HS, DS, and CS - increase the activity of MMP-2 and MMP-9, KS decreases the activity.<sup>34</sup>

The view of proteins belonging to the ECM is normally limited to collagens, elastin, PGs, and GPs. However, Naba and co-workers<sup>35-37</sup> suggested that a wider range of proteins should be sorted into this compartment. By mass spectrometry and a subsequent *in silico* bioinformatic approach to analyse the non-cellular part of tissue, i.e. the ECM, they identified the proteins mentioned above but also additional groups of proteins, including ECM-associated proteins and secreted factors, which they collectively called the matrisome. In total, 1,050 proteins were predicted to belong to the matrisome including cytokine growth factors and proteases. Importantly, the additional proteins that are resident in the matrisome may also contribute to ECM property and function, but have so far been overlooked in this context in most studies.

## ECM ALTERATIONS IN COPD

### Bronchus and Bronchioles

Thickening of the reticular basement membrane in central airways is an established part of the histopathology in asthma, but lack of unequivocal data makes it less clear in COPD.<sup>38-40</sup> In studies by Sohal and co-workers<sup>41</sup> it was shown that the bronchial and bronchiolar reticular basement membrane is fragmented in current smokers, and especially in COPD patients. In the fragmented regions cells positive for both epithelial and mesenchymal markers, the hallmark for epithelial mesenchymal transition (EMT), were observed.<sup>42,43</sup> These data were confirmed in a study showing that freshly isolated primary bronchial epithelial cells show an increase in the EMT expressing profile. In central airways in patients with mild-to-moderate disease, collagen Type 1 and 3 is increased in basement membranes, lamina propria, and in the adventitia.<sup>39,44,45</sup> These changes were negatively correlated to lung function parameters. An increase in total collagen has been recorded also in

bronchiolar tissue in moderate and severe disease.<sup>46,47</sup> The increase was due to an increased staining area of collagen Type 1 and 3 at the expense of other collagens. In parallel, the total amount of bronchiolar tissue was increased in moderate COPD patients while, in more severe disease, the amount is comparable to control subjects. In severe disease the level of total collagen decreases to levels lower than control subjects, suggesting that there may be different processes active at different parts of the disease.

The amount of the small PGs decorin and biglycan has been shown to decrease in the adventitia of bronchioles and in alveolar walls in patients with COPD, and especially in more severe disease.<sup>48,49</sup> Biglycan and decorin play an important role in cross-linking collagen fibrils, and this finding may, therefore, indicate loss of tissue integrity. These data do not agree with a study by Annoni et al.<sup>50</sup> who did not see any difference in the staining of decorin and biglycan in central or small airways, but the patients had milder disease in this study. The volume fraction of the GPs tenascin and fibronectin were, on the other hand, elevated in bronchi and bronchioles, which has also been shown by others.<sup>39,51</sup> These proteins have demonstrated to be elevated following tissue injury and inflammation, regulating cell adhesion and differentiation.<sup>52</sup> The non-sulphated GAG HA has been shown to increase in both alveolar tissue and in bronchioles in COPD.<sup>46</sup> HA has, in an animal model, been shown to have a protective role in bronchial tissue by blocking bronchial obstruction caused by inhaled elastase.<sup>53,54</sup> In a study by Kunz and co-workers<sup>55</sup> the effect of inhaled corticosteroids on expression of ECM proteins in COPD patients was examined. Tissue staining for collagen Type 3 and versican increased in bronchial biopsies following corticosteroid treatment for 18 months. The data suggest that long-term treatment with steroids can influence the remodelling process in the airways.

### Alveolar Tissue

Elastic fibres in the alveoli of COPD patients have been demonstrated to be abnormal and partly fragmented.<sup>56</sup> In addition it has also been shown that the volume fraction of elastic fibres is decreased in alveolar tissue and bronchioles in COPD patients.<sup>57-59</sup> This has been confirmed in other studies that show a decrease in elastin, the major component in elastic fibres, in alveoli and small airways in COPD patients of varying severity.<sup>46,60</sup> In agreement with these findings, fibroblasts isolated

from distal lung from COPD patients have impaired capability to produce elastin in response to TGF- $\beta$ 1.<sup>61</sup> Furthermore, a decreased amount of elastin has been suggested to be linked to an increase in the PG versican in alveolar walls. The increased versican is associated with a decrease in elastin binding protein that is required to assemble *de novo* produced elastin and other components of elastic fibres.<sup>60</sup> These data are in agreement that fibroblasts isolated from alveolar tissue from COPD patients have increased production of versican.<sup>11</sup> This is an example of the impaired repair function of fibroblasts in COPD that have been described by many groups.

Togo and colleagues<sup>62</sup> have shown that fibroblasts from mild-to-moderate COPD patients have impaired potential to migrate and contract. However, fibroblasts from severe COPD patients have been shown to be more contractile, suggesting that there may be different processes in different severities of the disease.<sup>12</sup> The levels of the eicosanoid prostacyclin have been shown to be elevated in COPD, and fibroblasts from severe COPD patients respond improperly to iloprost, a prostacyclin analogue, which could lead to impaired collagen network fibrillogenesis.<sup>63</sup> In line with these results, it has been shown by several groups that fibroblasts isolated from COPD patients show markers of senescence and are less prone to proliferate.<sup>61,64-66</sup> Van Straaten and coworkers<sup>49</sup> have also found diminished staining of HS PGs (perlecan, agrin, and collagen Type 18) in alveolar basement membranes, which may be an indication of alveolar damage.<sup>67</sup> So far, less is known about the GAG moiety of PGs in COPD patients, although both the amount of GAGs and the degree of sulphation are increased in other chronic airway diseases, including asthma and idiopathic pulmonary fibrosis. In addition, patients with idiopathic PAH have been shown to have increased levels of HA.<sup>68,69</sup>

### MMPs in COPD

Since the breakdown of alveolar tissue and ECM is one of the major causes of airflow limitations in COPD, much effort has been made to examine the role of proteases and MMPs in particular. MMPs do not only contribute to the breakdown of the ECM

in alveolar tissue but also to the remodelling process in bronchi and bronchioles, including to goblet cell metaplasia.<sup>70</sup> MMP-2 has been shown to increase in bronchioles and in surrounding parenchyma in COPD patients.<sup>71,72</sup> Increased levels of MMP-9 have been observed in bronchoalveolar lavage fluid from patients with emphysema.<sup>73,74</sup> Mercer and co-workers<sup>75</sup> have shown that the ratio between MMP-9 and its natural inhibitor, TIMP-1, becomes skewed in the direction of MMP-9 during an exacerbation, but during stable disease the relative level of MMP-9 decreases and the level of TIMP-1 increases. MMP-1 and MMP-8 have also been found to be elevated in COPD.<sup>76-78</sup> Further supporting the role of MMPs in COPD is that single nucleotide polymorphisms in the genes of MMP-1, MMP-2, MMP-9, and MMP-12 have been shown to be linked to disease.<sup>79</sup>

## CONCLUSION

Many of the airflow limitations in COPD are caused by structural remodelling resulting from an imbalance between synthesis and breakdown of ECM proteins. However, the involvement of these two counteracting processes is different throughout the airways as there is often excessive deposition of ECM in bronchi and bronchioles and loss of alveolar tissue. In addition, the involvement of these processes varies between different patients because of the great heterogeneity of the disease. To deepen the understanding of the dynamics and impact of these changes, there is therefore a great need to develop better tools for clinical and molecular phenotyping of patients. Moreover, since structural remodelling is an endogenous physiological response, it cannot be excluded that it may not always be detrimental but also protective, at least in some windows of disease progression. These possibilities should be considered in the development of new therapeutic approaches. One group of molecules that has so far been neglected in the exploration of structural remodelling in COPD are GAGs, the carbohydrate moiety of PGs. These molecules have been shown to interact with many cells and mediators that drive the disease, and should therefore be given more attention in the future.

## Acknowledgements

The study was supported by the Swedish Medical Research Council (11550), the Swedish Heart-Lung Foundation, Greta and John Kock, the Alfred Österlund Foundation, the Anna-Greta Crafoord Foundation, the Konsul Bergh Foundation, the Royal Physiographic Society in Lund, and the Medical Faculty of Lund University.

## REFERENCES

1. Barnes PJ et al. Chronic obstructive pulmonary disease: molecular and cellular mechanisms. *Eur Respir J*. 2003;22:672-88.
2. Calverley PM et al. Bronchodilator reversibility testing in chronic obstructive pulmonary disease. *Thorax*. 2003;58(8):659-64.
3. Anthonisen NR, Wright EC. Bronchodilator response in chronic obstructive pulmonary disease. *Am Rev Respir Dis*. 1986;133(5):814-9.
4. Brand PL et al. Interpretation of bronchodilator response in patients with obstructive airways disease. The Dutch Chronic Non-Specific Lung Disease (CNSLD) Study Group. *Thorax*. 1992;47(6):429-36.
5. Postma DS et al. Risk factors and early origins of chronic obstructive pulmonary disease. *Lancet*. 2014;doi:10.1016/S0140-6736(14)60446-3. [Epub ahead of print].
6. Petecchia L et al. Bronchial airway epithelial cell damage following exposure to cigarette smoke includes disassembly of tight junction components mediated by the extracellular signal-regulated kinase 1/2 pathway. *Chest*. 2009;135(6):1502-12.
7. Araya J, Nishimura SL. Fibrogenic reactions in lung disease. *Annu Rev Pathol*. 2010;5:77-98.
8. Morty RE et al. Transforming growth factor-beta signaling across ages: from distorted lung development to chronic obstructive pulmonary disease. *Proc Am Thorac Soc*. 2009;6(7):607-13.
9. Schiller M et al. TGF-beta-induced SMAD signaling and gene regulation: consequences for extracellular matrix remodeling and wound healing. *J Dermatol Sci*. 2004;35(2):83-92.
10. Churg A et al. Expression of profibrotic mediators in small airways versus parenchyma after cigarette smoke exposure. *Am J Respir Cell Mol Biol*. 2009;40(3):268-76.
11. Hallgren O et al. Altered fibroblast proteoglycan production in COPD. *Respir Res*. 2010;11:55.
12. Hallgren O et al. Enhanced ROCK1 dependent contractility in fibroblast from chronic obstructive pulmonary disease patients. *J Transl Med*. 2012;10:171.
13. Kotaru C et al. Regional fibroblast heterogeneity in the lung: implications for remodeling. *Am J Respir Crit Care Med*. 2006;173(11):1208-15.
14. Pechkovsky DV et al. Human lung parenchyma but not proximal bronchi produces fibroblasts with enhanced TGF-beta signaling and alpha-SMA expression. *Am J Respir Cell Mol Biol*. 2010;43(6):641-51.
15. Sakao S et al. The vascular bed in COPD: pulmonary hypertension and pulmonary vascular alterations. *Eur Respir Rev*. 2014;23(133):350-5.
16. Wright JL et al. The structure and function of the pulmonary vasculature in mild chronic obstructive pulmonary disease. The effect of oxygen and exercise. *Am Rev Respir Dis*. 1983;128(4):702-7.
17. Chaouat A et al. Pulmonary hypertension in COPD. *Eur Respir J*. 2008;32:1371-85.
18. Churg A et al. Series "matrix metalloproteinases in lung health and disease": matrix metalloproteinases in COPD. *Eur Respir J*. 2012;39(1):197-209.
19. Laurell CB et al. Alpha1-antitrypsin deficiency. Pi genotype ZO, SO and MO. *Acta Paediatr Scand*. 1974;63(6):855-7.
20. Barnes PJ. Development of new drugs for COPD. *Curr Med Chem*. 2013;20(12):1531-40.
21. Capila I, Linhardt RJ. Heparin-protein interactions. *Angew Chem Int Ed Engl*. 2002;41(3):391-412.
22. Turnbull J et al. Heparan sulfate: decoding a dynamic multifunctional cell regulator. *Trends Cell Biol*. 2001;11(2):75-82.
23. Sugahara K et al. Recent advances in the structural biology of chondroitin sulfate and dermatan sulfate. *Curr Opin Struct Biol*. 2003;13(5):612-20.
24. Toole BP. Hyaluronan: from extracellular glue to pericellular cue. *Nat Rev Cancer*. 2004;4(7):528-39.
25. Suki B et al. Biomechanics of the lung parenchyma: critical roles of collagen and mechanical forces. *J Appl Physiol* (1985). 2005;98(5):1892-9.
26. Deakin JA et al. The binding properties of minimal oligosaccharides reveal a common heparan sulfate/dermatan sulfate-binding site in hepatocyte growth factor/scatter factor that can accommodate a wide variety of sulfation patterns. *J Biol Chem*. 2009;284(10):6311-21.
27. Kemp LE et al. Signalling by HGF/SF and Met: the role of heparan sulphate co-receptors. *Biochem Soc Trans*. 2006;34(Pt 3):414-7.
28. Chen Q et al. Potential role for heparan sulfate proteoglycans in regulation of transforming growth factor-beta (TGF-beta) by modulating assembly of latent TGF-beta-binding protein-1. *J Biol Chem*. 2007;282(36):26418-30.
29. Proudfoot AE et al. Glycosaminoglycan binding and oligomerization are essential for the in vivo activity of certain chemokines. *Proc Natl Acad Sci USA*. 2003;100(4):1885-90.
30. Handel TM et al. Regulation of protein function by glycosaminoglycans--as exemplified by chemokines. *Annu Rev Biochem*. 2005;74:385-410.
31. Donnelly G et al. The effect of extracellular matrix on the growth of mouse olfactory tissue in vitro. *Neuroreport*. 1998;9(17):3837-40.
32. Spillmann D et al. Defining the interleukin-8-binding domain of heparan sulfate. *J Biol Chem*. 1998;273(25):15487-93.
33. Spencer JL et al. New insights into the inhibition of human neutrophil elastase by heparin. *Biochemistry*. 2006;45(30):9104-20.
34. Isnard N et al. Effect of sulfated GAGs on the expression and activation of MMP-2 and MMP-9 in corneal and dermal explant cultures. *Cell Biol Int*. 2003;27(9):779-84.
35. Hynes RO, Naba A. Overview of the matrisome: an inventory of extracellular matrix constituents and functions. *Cold Spring Harb Perspect Biol*. 2012;4(1):a004903.
36. Naba A et al. The matrisome: in silico definition and in vivo characterization by proteomics of normal and tumor extracellular matrices. *Mol Cell Proteomics*. 2012;11(4):M111.014647.
37. Naba A et al. Towards definition of

- an ECM parts list: an advance on GO categories. *Matrix Biol.* 2012;31(7-8): 371-2.
38. Jeffery PK. Remodeling in asthma and chronic obstructive lung disease. *Am J Respir Crit Care Med.* 2001;164(10 Pt 2):S28-38.
39. Liesker JJ et al. Reticular basement membrane in asthma and COPD: similar thickness, yet different composition. *Int J Chron Obstruct Pulmon Dis.* 2009;4: 127-35.
40. Polosukhin VV et al. Association of progressive structural changes in the bronchial epithelium with subepithelial fibrous remodeling: a potential role for hypoxia. *Virchows Arch.* 2007;451(4): 793-803.
41. Sohal SS et al. Reticular basement membrane fragmentation and potential epithelial mesenchymal transition is exaggerated in the airways of smokers with chronic obstructive pulmonary disease. *Respirology.* 2010;15:930-8.
42. Sohal SS et al. Evaluation of epithelial mesenchymal transition in patients with chronic obstructive pulmonary disease. *Respir Res.* 2011;12:130.
43. Sohal SS, Walters EH. Epithelial mesenchymal transition (EMT) in small airways of COPD patients. *Thorax.* 2013;68(8):783-4.
44. Harju T et al. Variability in the precursor proteins of collagen I and III in different stages of COPD. *Respir Res.* 2010;11:165.
45. Kranenburg AR et al. Enhanced bronchial expression of extracellular matrix proteins in chronic obstructive pulmonary disease. *Am J Clin Pathol.* 2006;126(5):725-35.
46. Eurlings IM et al. Similar matrix alterations in alveolar and small airway walls of COPD patients. *BMC Pulm Med.* 2014;14:90.
47. Hogg JC et al. What drives the peripheral lung-remodeling process in chronic obstructive pulmonary disease? *Proc Am Thorac Soc.* 2009;6(8):668-72.
48. Noordhoek JA et al. Different modulation of decorin production by lung fibroblasts from patients with mild and severe emphysema. *COPD.* 2005;2(1): 17-25.
49. van Straaten JF et al. Proteoglycan changes in the extracellular matrix of lung tissue from patients with pulmonary emphysema. *Mod Pathol.* 1999;12(7): 697-705.
50. Annoni R et al. Extracellular matrix composition in chronic obstructive pulmonary disease. *Eur Respir J.* 2012;40(6):1362-73.
51. Amin K et al. Relationship between inflammatory cells and structural changes in the lungs of asymptomatic and never smokers: a biopsy study. *Thorax.* 2003;58(2):135-42.
52. Jones FS, Jones PL. The tenascin family of ECM glycoproteins: structure, function, and regulation during embryonic development and tissue remodeling. *Dev Dyn.* 2000;218(2):235-59.
53. Cantor JO, Turino GM. Can exogenously administered hyaluronan improve respiratory function in patients with pulmonary emphysema? *Chest.* 2004;125(1):288-92.
54. Scuri M et al. Hyaluronic acid blocks porcine pancreatic elastase (PPE)-induced bronchoconstriction in sheep. *Am J Respir Crit Care Med.* 2001;164(10 Pt 1):1855-9.
55. Kunz LI et al. Inhaled steroids modulate extracellular matrix composition in bronchial biopsies of COPD patients: a randomized, controlled trial. *PLoS One.* 2013;8(5):e63430.
56. Finlay GA et al. Elastin and collagen remodeling in emphysema. A scanning electron microscopy study. *Am J Pathol.* 1996;149(4):1405-15.
57. Black PN et al. Changes in elastic fibres in the small airways and alveoli in COPD. *Eur Respir J.* 2008;31(5):998-1004.
58. Hogg JC, Timens W. The pathology of chronic obstructive pulmonary disease. *Annu Rev Pathol.* 2009;4:435-59.
59. Turino GM et al. Lung elastin content in normal and emphysematous lungs. *Bull Eur Physiopathol Respir.* 1980;16 Suppl:43-57.
60. Merrilees MJ et al. Changes in elastin, elastin binding protein and versican in alveoli in chronic obstructive pulmonary disease. *Respir Res.* 2008;9:41.
61. Zhang J et al. Pro-inflammatory phenotype of COPD fibroblasts not compatible with repair in COPD lung. *J Cell Mol Med.* 2012;16(7):1522-32.
62. Togo S et al. Lung fibroblast repair functions in patients with chronic obstructive pulmonary disease are altered by multiple mechanisms. *Am J Respir Crit Care Med.* 2008;178(3):248-60.
63. Larsson-Callert AK et al. Defective alterations in the collagen network to prostacyclin in COPD lung fibroblasts. *Respir Res.* 2013;14:21.
64. Holz O et al. Lung fibroblasts from patients with emphysema show a reduced proliferation rate in culture. *Eur Respir J.* 2004;24(4):575-9.
65. Nobukuni S et al. Cigarette smoke inhibits the growth of lung fibroblasts from patients with pulmonary emphysema. *Respirology.* 2002;7(3):217-23.
66. Noordhoek JA et al. Different proliferative capacity of lung fibroblasts obtained from control subjects and patients with emphysema. *Exp Lung Res.* 2003;29(5):291-302.
67. Smits NC et al. Heparan sulfates in the lung: structure, diversity, and role in pulmonary emphysema. *Anat Rec (Hoboken).* 2010;293(6):955-67.
68. Venkatesan N et al. Allergen-induced airway remodeling in brown norway rats: structural and metabolic changes in glycosaminoglycans. *Am J Respir Cell Mol Biol.* 2012;46(1):96-105.
69. Venkatesan N et al. Allergen-induced airway remodeling in brown Norway rats: structural and metabolic changes in glycosaminoglycans. *Am J Respir Cell Mol Biol.* 2012;46(1):96-105.
70. Elkington PT, Friedland JS. Matrix metalloproteinases in obstructive pulmonary pathology. *Thorax.* 2006;61(3):259-66.
71. Baraldo S et al. Matrix metalloproteinase-2 protein in lung periphery is related to COPD progression. *Chest.* 2007;132(6):1733-40.
72. Chen Y et al. Enhanced levels of prostaglandin E2 and matrix metalloproteinase-2 correlate with the severity of airflow limitation in stable COPD. *Respirology.* 2008;13(7):1014-21.
73. Betsuyaku T et al. Neutrophil granule proteins in bronchoalveolar lavage fluid from subjects with subclinical emphysema. *Am J Respir Crit Care Med.* 1999;159(6):1985-91.
74. Finlay GA et al. Elevated levels of matrix metalloproteinases in bronchoalveolar lavage fluid of emphysematous patients. *Thorax.* 1997;52(6):502-6.
75. Mercer PF et al. MMP-9, TIMP-1 and inflammatory cells in sputum from COPD patients during exacerbation. *Respir Res.* 2005;6:151.
76. Segura-Valdez L et al. Upregulation of gelatinases A and B, collagenases 1 and 2, and increased parenchymal cell death in COPD. *Chest.* 2000;117(3):684-94.
77. Montano M et al. FEV1 inversely correlates with metalloproteinases 1, 7, 9 and CRP in COPD by biomass smoke exposure. *Respir Res.* 2014;15:74.
78. Craig VJ et al. Mononuclear phagocytes and airway epithelial cells: novel sources of matrix metalloproteinase-8 (MMP-8) in patients with idiopathic pulmonary fibrosis. *PLoS One.* 2014;9(5):e97485.
79. Moccigiani E et al. Metalloproteases/anti-metalloproteases imbalance in chronic obstructive pulmonary disease: genetic factors and treatment implications. *Curr Opin Pulm Med.* 2011;17 Suppl 1:S11-19.