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INVITED REVIEW

Recent advances in chronic obstructive pulmonary disease pathogenesis: from disease mechanisms to precision medicine

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Abstract

Chronic obstructive pulmonary disease (COPD) is a devastating lung disease with a high personal and societal burden. Exposure to toxic particles and gases, including cigarette smoke, is the main risk factor for COPD. Together with smoking cessation, current treatment strategies of COPD aim to improve symptoms and prevent exacerbations, but there is no disease-modifying treatment. The biggest drawback of today's COPD treatment regimen is the 'one size fits all' pharmacological intervention, mainly based on disease severity and symptoms and not the individual's disease pathology. To halt the worrying increase in the burden of COPD, disease management needs to be advanced with a focus on personalized treatment. The main pathological feature of COPD includes a chronic and abnormal inflammatory response within the lungs, which results in airway and alveolar changes in the lung as reflected by (small) airways disease and emphysema. Here we discuss recent developments related to the abnormal inflammatory response, ECM and age-related changes, structural changes in the small airways and the role of sex-related differences, which are all relevant to explain the individual differences in the disease pathology of COPD and improve disease endotyping. Furthermore, we will discuss the most recent developments of new treatment strategies using biologicals to target specific pathological features or disease endotypes of COPD.

Keywords: COPD; inflammation; aging; ECM; genome-wide association study; CELSR1; sex differences; small airways; precision imaging; personalized treatment; biologics

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Introduction

Chronic obstructive pulmonary disease (COPD) is an often severely disabling chronic lung disease with a high prevalence of over 250 million cases worldwide. It is currently the fourth leading cause of death globally and is predicted to be the third by 2020 [1]. In the most recent definition of the Global Initiative for Chronic Obstructive Lung Disease (GOLD), COPD is defined as 'a common, preventable and treatable disease that is characterized by persistent respiratory systems and airflow limitation that is due to airway and/or alveolar abnormalities usually caused by significant exposure to noxious particles or gases' [2]. This exposure can be quite variable, with smoking being the main risk factor in high-income countries and indoor cooking and occupational exposures representing important risk factors in low-income countries. The definition and staging by GOLD is rather uniform, as defined by lung function, symptoms and exacerbation history, despite COPD being a heterogeneous disease in its pathological manifestations in patients. It has long been recognized that

inflammation is a central hallmark of COPD, playing a role in the pathological changes in all different lung compartments [3]. Next to toxic exposures, genetic predisposition is an important risk factor for COPD and COPD represents a complex disease in which genetic abnormalities in combination with the type and duration of exposures determine the clinical phenotype [4]. The development of novel 2D and 3D *in vitro* and *ex vivo* models, as well as animal models that accurately recapitulate the main features of COPD pathology, has been very important for the study of disease mechanisms in COPD and recent developments in this area have been amply reviewed elsewhere [5–10].

In the present review we focus on recent developments related to the abnormal inflammatory response, ECM and age-related changes, structural changes in the small airways and the role of sex-related differences, which are relevant to explain the underlying individual differences in the disease pathology of COPD and are important to improve disease endotyping. Where possible, we will underpin the observed pathogenetic changes by their potential genetic drivers.

Finally, we will discuss the most recent developments of new treatment strategies using biologicals to target specific pathological features or disease endotypes (a specific group of patients who share a distinct pathobiological mechanism) of COPD.

Abnormal inflammatory responses in COPD

It has long been known that the innate immune system plays a main role in COPD, as reviewed previously [3]. Although it can be envisaged that noxious gases will evoke such an immune response, the peculiarity within COPD is that it is more extensive and damaging and sustained for a longer time than, for example, in smokers without COPD. Neutrophilic inflammation, as observed in the innate response, is strongly dependent on IL-1-alpha, which is reported to be increased in COPD patients [11,12] and also more readily induced in COPD airway epithelial cells [13].

In the adaptive immune response in COPD, the predominant cell is the CD8 cytotoxic T cell. The presence of this cell type in the airways as well as parenchyma remains sustained over a long period of time, even up to 3 years after smoking cessation [14,15]. The finding of lymphoid aggregates and follicles in COPD [16] and, in particular, the confirmation of oligoclonality in these follicles [17], fitted very well with the concept of autoimmunity. In severe COPD, IL-18, associated with lung lymphoid aggregates, has been shown to drive IFN-gamma production, contributing to a Th1 response [18]. Nevertheless, clonal B cell responses could be a consequence of antigenic exposure due to the disease (matrix components, infectious agents, immune components) and does not necessarily prove that this would also contribute to disease [19]. More recently, the role of innate lymphoid cells (ILC) in inflammatory disease has received more attention [20]. Although this role in COPD as yet is far from clear, group 3 ILC (ILC3) appear to be the main subtype in COPD [20], suggested to be involved in the initiation of the ectopic lymphoid aggregates [21]. In addition, ILC1 were found to be associated with lymphoid cell infiltration and have been postulated to play a role in emphysematous destruction in COPD [22]. In COPD exacerbations it was shown that ILC2 can switch to ILC1 and thus also contribute to IFN-gamma-driven inflammation [23].

Type 2 inflammation, normally associated with asthma, has also been described in COPD patients without a history of asthma and was suggested to represent an endotype of COPD [24]. Obviously, this has also been discussed as a representative of the 'asthma COPD overlap syndrome' [25]. Furthermore, a key role has been described for Th17 cells and their principal cytokine, IL-17, at least in a subset of COPD [26–28]. In particular, human lung dendritic cells from emphysema patients stimulated Th1 and Th17 responses [29] and emphysema patients had higher Th17 cell levels, which were responsive to elastin stimulation *in vitro* [29]. Furthermore, the response to elastin stimulation was associated with the percentage of emphysema on CT scans, supporting a possible (T cell-mediated) autoimmune phenomenon. A very recent study showed an airway epithelial IL-17A response signature identifying a steroid-unresponsive COPD patient subgroup [28]. The authors suggested that such a gene signature of IL-17 airway epithelial response distinguishes a biologically, radiographically and clinically distinct COPD subgroup that may benefit from personalized therapy [28].

ECM changes in COPD

The lung ECM forms the main building blocks of the lung. Disturbance of the ECM can have important consequences leading to lung tissue remodeling, which can affect all lung compartments (i.e. airway wall fibrosis and emphysema) in COPD. The main ECM compartments in the lung are the basement membrane, lamina propria of the airways and the alveolar interstitium, where the ECM connects the alveoli and blood vessels, forming the parenchyma. The main protein components of the basement membrane are collagen IV and laminin; in the lamina propria and interstitium they are fibrillar collagens, elastin, fibronectin, glycosaminoglycans and proteoglycans [30,31].

The main producers of ECM proteins in the lung are fibroblasts and activated myofibroblasts, followed by airway smooth muscle and airway epithelial cells [32,33]. Degradation of the ECM is regulated by endogenous proteases, of which MMPs are the main type in the lung; their activity is regulated by tissue inhibitors of metalloproteinases (TIMPs). The balance between these MMPs and TIMPs is essential for ECM homeostasis [30,34,35].

A potential contributor of emphysematous lung tissue destruction in COPD is the inflammation-related increase in MMPs (i.e. MMP-2, MMP-9 and MMP-12), leading to an imbalance between MMPs and TIMPs and a shift toward increased ECM degradation, especially elastin [36–38]. Several studies have shown a reduction in elastic fibers in COPD, as well as disrupted fibers and disturbed elastogenesis [39–41]. Additionally, increased elastin gene expression was demonstrated in severe COPD [37], as well as an upregulation of several elastogenesis-related genes, including elastin and fibulin-5, in COPD lung tissue [42]. Of interest, fibulin-5 appeared to be a key modulator of elastic fiber formation, with fibulin-5 knockout mice suffering from severe elastinopathy, resulting in loose skin, vascular abnormalities and severe emphysema [43,44]. Of interest, fibulin-5 protein was shown to be cleaved by serine proteases *in vitro*, resulting in disturbed elastogenesis [45]. Thus, the increase in elastin and fibulin-5 on the gene expression level and the decreased presence of elastic fibers and elastin fiber abnormalities in COPD suggest a defect in the formation and repair of elastic fibers in COPD, which is possibly related to increased levels of cleaved non-functional fibulin-5 protein. In addition to the concept that increased fibulin-5 gene expression is a response of hampered tissue repair in COPD, it has also been proposed that the increased fibulin-5 expression in COPD is involved in small airway fibrosis and is thus detrimental [46].

Other important ECM changes in COPD include increased collagen deposition in the (small) airway walls and structural changes in collagen fibrils, with more disorganized collagen fibrils in COPD [47,48]. An important contributing factor to this disorganization of collagen is the lack of decorin deposition in the adventitia of the small airways [49]. Decorin is a proteoglycan that binds to collagen fibrils, providing structural support. Decorin also binds many growth factors and their receptors, including TGF-beta, the main inducer of tissue repair, thereby inhibiting growth factor activity [50–52].

Together, the collagen changes in the airways and elastic fiber-related changes with loss of alveolar attachments in peripheral lungs leads to loose airway walls that lack their alveolar support, resulting in increased airway collapsibility and airflow limitation during expiration in COPD. An example of a diseased airway with lack of alveolar support is shown in Figure 1.

Genetic regulation of ECM changes in COPD

The list of genetic variants associated with COPD or reduced lung function is increasing rapidly [53–56]. Whether and which of these genetic variants have functional consequences for COPD pathology and are underlying ECM changes in COPD is a million dollar question. Of interest, several recent genome-wide association studies (GWAS) showed an enrichment of, partly the same, ECM-related genes among their lists of genetic variants for COPD and/or pulmonary function (Table 1). Gharib *et al* [57] performed GWAS analysis of pulmonary function measures (forced expiratory volume in the first second [FEV_1] and FEV_1/forced vital capacity [FVC]) in two large consortia (CHARGE and SpiroMeta) and used gene set enrichment analysis to identify pathways linked to pulmonary function. Interestingly, their pathway analysis for airflow obstruction (decreased FEV_1/FVC) resulted in a clear enrichment of gene sets involved in ECM remodeling, including the integrin pathway, proteinaceous ECM, collagen and ECM. Subsequent network analysis identified MMP-10 as a candidate gene and they demonstrated a pathophysiological role for MMP-10 in a mouse model of smoke-induced emphysema [57].

Sakornsakolpat *et al* [58] used a new approach integrating GWAS and existing gene expression data to identify new genes for severe COPD. Using pathway enrichment analysis, they demonstrated enrichment of cell matrix adhesion of collagen binding, which included three ECM-binding integrin genes (*ITGA10*, *ITGA2*, *ITGA1*).

In 2017, Wain *et al* [54] published a large GWAS study for lung function and COPD using the UK Biobank. Integration of the genetic variants with existing expression Quantitative Trait Loci (eQTL) datasets resulted in the identification of 234 genes with potentially causal effects on lung function that were enriched for several ECM-related genes, including molecules related to elastic fibers, elastic fiber formation, ECM organization and extracellular structure organization. These gene sets included fibulin-5 as a potential causal gene for impaired lung function. More recently, two even larger GWAS studies for lung function and COPD were published by Shrine *et al* [55] and Sakornsakolpat *et al* [56], identifying 139 new genetic loci for lung function and 35 new loci for COPD, respectively. Again, pathway analysis of the lung function-associated genes showed that most significantly enriched pathways were related to elastic fibers and ECM organization and this also included a new signal for an ECM-binding integrin, integrin alpha-V. A significant enrichment of ECM-related pathways was also found among the COPD-associated genes, including laminin binding, integrin binding, mesenchyme development, cell matrix adhesion and actin filament bundles. Together with the enrichment of lung development pathways among the COPD-associated genes, these findings strongly support a role for disturbed ECM development and homeostasis in the development of COPD. Follow-up functional studies are now warranted to disentangle the exact disease-driving mechanism and to identify new potential targets for intervention.

Aging and lung ECM changes in COPD

Aging is the progressive decline of homeostasis, resulting in increased risk of disease or death [59]. The aging

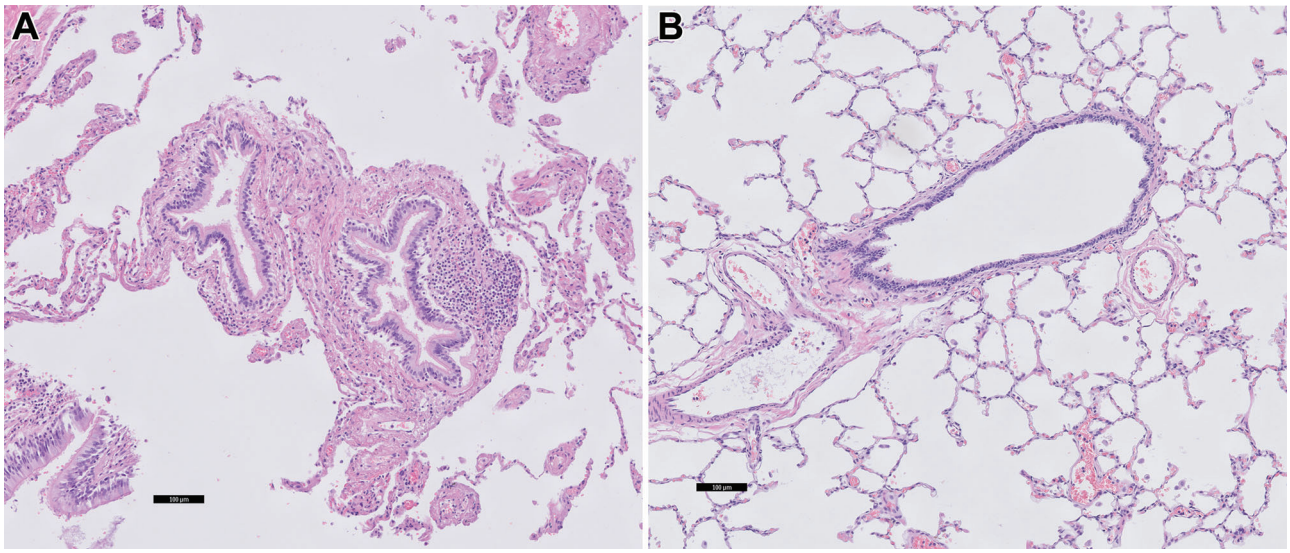

Figure 1. (A) COPD lung tissue with severe emphysema showing a small airway with extensive loss of alveolar attachments. (B) Comparable image of normal lung tissue with a small airway with normal parenchymal surroundings and attachments.

lung is characterized by several physiological and structural changes that are also present in COPD, including a decline in lung function, decreased mucociliary clearance, decreased antioxidant levels, senile emphysema and altered ECM proteins [60]. Furthermore, mutations in genes responsible for telomere shortening, a hallmark of aging, have been associated with severe emphysema [61,62]. Therefore, it has been suggested that accelerated lung aging plays an important role in the pathogenesis of COPD [59,63].

Although ECM dysregulation was proposed as an additional hallmark of the aging lung [64], it is not yet completely clear how lung ECM homeostasis is affected by aging and how this may contribute to COPD. The occurrence of airspace enlargement or senile emphysema [65] and the decrease in elastic fibers and increased collagen deposition in the aging lung [66,67] are, however, indicative of lung remodeling.

It has been proposed that the aged lung is characterized by subtle changes in the ECM and lung architecture, which does not directly cause disease or changes in lung function, so-called transitional remodeling [68]. When the lung is then exposed to an additional insult, like in COPD, these alterations may trigger an aberrant repair response and cause permanent tissue damage. This theory is in line with the findings of a recent lung tissue gene expression study where it was shown that genes involved in ECM–receptor interactions, including three collagen genes, decreased more with age in COPD patients compared with controls, suggesting a defective repair response in COPD with aging [69]. In addition, Calhoun *et al* [70] demonstrated an increase in ECM proteins, including collagen, with increasing age in mice and human lungs, together with an increase in senescent cells. In a more recent study [71], the link between growth differentiation factor 11 (GDF11), a member of the TGF family, and senescence in COPD was investigated. GDF11 was decreased in plasma of COPD patients and the protein was localized in mesenchymal cells in the airway wall and the airway epithelium. Treatment with GDF11 attenuated emphysema and cellular senescence in an elastase-induced emphysema model, indicating a role for GDF11 in cellular senescence in COPD.

The true role of age-related ECM changes in COPD pathology is still to be determined, but it certainly represents an interesting area for research. More so because targets for pharmaceutical intervention can potentially be applicable to other age-related diseases as well. In this respect, it is of interest that novel anti-aging treatment strategies are emerging that target cellular senescence with beneficial effects on lifespan, lung aging and lung fibrosis in animal models [72–74] and very recently also with beneficial effects in patients with idiopathic pulmonary fibrosis [75]. This first-in-human, open-label pilot study, where 14 idiopathic pulmonary fibrosis patients were treated with the combination of dasatinib (a tyrosine kinase inhibitor that targets anti-apoptosis pathways; used for the treatment of leukemia) and quercetin (a natural product, flavonoid, targeting anti-apoptosis pathways) showed significant and clinically meaningful physical improvement in these patients. Further evaluation of these senolytic drugs in larger randomized controlled trials is warranted.

Precision imaging for new insights into COPD pathology

The major site of airflow obstruction in COPD has been shown, by direct measurements of resistance, to occur in the small conducting airways that are <2 mm in internal diameter [76–78]. Since then, there has been much debate over the most important contributor to airflow obstruction: structural changes within small airways or loss of elastic recoil due to emphysematous destruction.

Table 1. Enrichment of genes involved in ECM remodeling processes among genetic variants associated with COPD and lung function

Study	Phenotype	Gene set*	*P* value enrichment†	Key enriched genes‡
Gharib *et al* [57]	Airflow obstruction (FEV_1/FVC)	Integrin pathway	0	§*ACAN, COL1A2, COL3A1, COL4A2, COL4A4, COL5A1, COL5A3, COL18A1, COL15A1, FBN1, FBN2, FBLN1, FBLN2, HAPLN1, LAMA4, LAMC1, LUM, MFAP5, MMP10, NID2, THBS4*
		Proteinaceous ECM	1.45E−04	
		ECM part	1.75E−04	
		ECM	2.50E−04	
		Collagen	2.52E−04	
		ECM structural constituent	9.48E−04	
Wain *et al* [54]	Lung function (FEV_1, FVC, FEV_1/FVC)	**Molecules associated with elastic fibers**	0.006	*EFEMP1, TGFB2, LTBP4, MFAP2, FBLN5*
		Elastic fiber formation	0.008	*EFEMP1, TGFB2, LTBP4, MFAP2, FBLN5*
		ECM organization	0.029	*HSD17B12, MMP15, TGFB2, CTSK, ADAMTSL4, EFEMP1, ITGA1, THSD4, NTN4, NPNT, LTBP4,MFAP2, CTSS, LEMD3, FBLN5*
		Extracellular structure organization	0.019	*HSD17B12, MMP15, TGFB2, CTSK, ADAMTSL4, EFEMP1, ITGA1, THSD4, NTN4, NPNT, LTBP4,MFAP2, CTSS, LEMD3, FBLN5*
		Fibronectin binding	0.014	*HSD17B12, CTSS, CTSK, MFAP2*
Sakornsakolpat *et al* [58]	Severe COPD	Collagen binding involved in cell matrix adhesion	2.70E−03	*ITGA10, ITGA2, ITGA1*
Sakornsakolpat *et al* [56]	COPD	Basement membrane	<0.01	*COL15A1, COL18A1, PRSS23, ITGA1, GPX8, DSP, TIMP3, SERPING1, GJA1, DST*
		ECM part	<0.01	*COL15A1, SERPING1,GPX8, COL18A1, B3GNT9, PRSS23, GJA1, TIMP3, SCARF2, EMID1*
		Laminin complex	<0.01	*COL15A1, ELMO3, COL18A1, ITGA2, DST, GPX8, DSP, FGD6, ENSG00000228536, ESRP2*
		ECM	<0.05	*COL15A1, GPX8, SERPING1, COL18A1, LRP1, B3GNT9, SCARF2, GJA1, PRSS23, TIMP3*
		Collagen binding	<0.05	*COL15A1, LRP1, TIMP3, GPX8, PRSS23, GJA1, ADAMTSL3, SERPING1, DLC1, SCARF2*
		Proteinaceous ECM	<0.05	*COL15A1, GPX8, COL18A1, SERPING1, B3GNT9, LRP1, GJA1, SCARF2, ADAMTSL3, PRSS23*
		ECM binding	<0.05	*TIMP3, COL15A1, COL18A1, EMP1, PRSS23, ITGAV, ARHGEF17, GPX8, SCARF2, GJA1*
		Cell matrix adhesion	<0.05	*ASAP2, SH3PXD2A, ENSG00000173517, KIAA0754, ADAMTSL1, ITGA1, PARVA, ENSG00000251175, ENSG00000223561, ARHGEF17*
Shrine *et al* [55]	Lung function (FEV_1, FVC, FEV_1/FVC)	**Molecules associated with elastic fibers**	9.33E−05	*ITGAV, TGFB2, LTBP4, MFAP2, GDF5*
		Elastic fiber formation	0.000104	*ITGAV, TGFB2, LTBP4, MFAP2, GDF5*
		ECM organization	0.00241	*MMP15, TGFB2, LTBP4, DST, ITGAV, P4HA2, MFAP2, GDF5, ADAM19*
		Alpha6Beta4Integrin	0.018468	*MET, DST, DSP, SMAD3*
		TGF-Core	0.036822	*TGFB2, GDF5, SMAD3*
		TGF-beta2 production	0.034539	*TGFB2, SMAD3*
		Extracellular structure organization	0.045883	*MMP15, TGFB2, THSD4, ITGAV, SMAD3, NPNT, MFAP2*
		TGF-beta receptor binding	0.012484	*TGFB2, GDF5, SMAD3*
		TGF-beta binding	0.026674	*LTBP4, ITGAV*

Only gene sets involved in ECM remodeling processes were selected from the referred publications.
*Gene sets enriched in more than one study are highlighted in bold.
† *P* values are corrected for multiple testing using different methods.
‡ Genes enriched in more than one study are underlined.
§ Twenty-one focus genes derived from network analysis based on enriched gene sets.

To date, clinical thoracic CT scans have been validated as a non-invasive imaging technique to correlate with regional lung function [79] and to quantify emphysema (<−950 Hounsfield Units) [80,81]. However, the spatial resolution of clinical CT scans, 800–1000 μm, does not permit the analysis of the smallest conducting airways, or parenchymal structures [82–84]. The recent application of micro-CT, with a spatial resolution of up to 1 μm, has provided the ability to resolve alveolar structures and reliably identify terminal bronchioles, with an average lumen diameter of 424 μm [85]. McDonough and colleagues [85] used micro-CT to image tissue samples from explanted lungs from patients with very severe (GOLD4) COPD, and provided the first evidence that terminal bronchioles are destroyed in end-stage COPD, even in regions of lung with no emphysema measured using mean linear intercept (a measure for airspace enlargement). These data therefore suggested that loss of terminal bronchioles may occur prior to the development of emphysema. To test this hypothesis, Koo and

colleagues [86] used a cross-sectional analysis of smokers with normal lung function and patients with mild, moderate and very severe COPD, and demonstrated that terminal bronchioles are significantly destroyed by 41% in mild (GOLD1), 43% in moderate (GOLD2) and 69% in end-stage (GOLD4) COPD patients. Using the robust measure of alveolar surface area, which translates to the functional tissue involved in gas exchange, the study also reported that terminal and transitional (first generation of respiratory airways) bronchioles are lost in lung tissue in which no emphysematous destruction is present, indicating that small airways disease is an early pathological feature of mild and moderate COPD.

Although micro-CT has the resolution to assess small airways, only CT and MRI modalities hold promise for the early detection of obstructive lung diseases and the characterization of disease pathology over time. The recent application of parametric response mapping (PRM) [87], which uses image registration to match inspiratory and expiratory CT scans to examine local changes in lung density, is now able to detect emphysema (PRM^{emph}) from non-emphysematous gas trapping, termed functional small airways disease (PRM^{fSAD}). In a subsequent study using excised lungs and micro-CT [88], it was validated that PRM^{fSAD} is associated with a lower number of terminal bronchioles and a reduced terminal bronchiole lumen area, whereas changes in alveolar surface area only correlated with PRM^{emph}. This is important, as Bhatt and colleagues [89], using the COPDGene cohort of 1508 current and former smokers, have shown that PRM^{fSAD} is associated with early disease, before the development of airflow limitation, and contributed to a faster FEV_1 decline than PRM^{emph}. Furthermore, Labaki and colleagues [90], in a cohort of 725 smokers with and without COPD, reported that PRM^{fSAD} is associated with regions of normal tissue over a range of COPD severities and that subjects with the highest PRM^{fSAD} at baseline had the greatest increase in emphysema. PRM has also been shown to correlate with GOLD stage and several clinical parameters used to assess COPD morbidity, such as BMI, 6 min walking distance and quality of life as determined by the St-George Respiratory Questionnaire (SGRQ) [91]. In addition, PRM^{fSAD} has been shown to be sensitive enough for short-term (1 year) follow-up of COPD patients, with PRM^{fSAD} decreasing as PRM^{emph} and tissue destruction increase [92].

These studies therefore support the notion that small airways disease is a precursor to the development of emphysema. As a CT biomarker, PRM^{fSAD} has the potential to improve patient care through improved screening, disease subtyping and monitoring the response to treatment as an outcome measure. Future GWAS to associate genes with PRM disease subtypes will probably be informative compared with the traditional lung function measures to identify potential therapeutic targets and early disease markers.

Role of sex differences in COPD pathology

Although COPD has long been considered a disease of males, its prevalence and mortality among females have risen rapidly during the last decades and are now equal to those of males [93]. Several studies have shown that sex differences exist with respect to susceptibility to smoking and clinical presentation of COPD. Females report more symptoms of dyspnea and cough and have a faster annual decline in FEV_1, even with similar pack-years of smoking [94–96]. In a population-based cohort study (SAPALDIA) of 1792 current smokers, Downs *et al* [96] investigated the effects of the number of cigarettes smoked per day on the course of lung function in males and females measured over a period of 11 years (Figure 2). In both sexes, the number of packs of cigarettes smoked per day was related to lung function decline, the mean annual FEV_1 decline being −10.4 ml per pack per day in males and −13.8 ml in females. Interestingly, smoking-related FEV_1 decline was accelerated in females with airflow obstruction (FEV_1/FVC <0.7) compared with those without (FEV_1/FVC >0.7), the decline being −39.4 versus −12.2 ml/year per pack per day, respectively ($p < 0.002$), whereas this was not the case in males (FEV_1 decline of −12.9 versus −8.8 ml/year per pack, $p = 0.07$). The difference between men and women with a reduced FEV_1/FVC was significant ($p = 0.05$), indicating that women with airway obstruction experience a greater smoking-related lung function decline than men (Figure 2). Another argument in support of the notion that females are more susceptible to the adverse effects of smoking is the finding that females are over-represented among patients with severe early onset COPD, defined as the occurrence of COPD before the age of 53 years with an FEV_1 below 40% predicted. In a study by Silverman *et al* [97], a group of 84 probands with severe early onset COPD (in

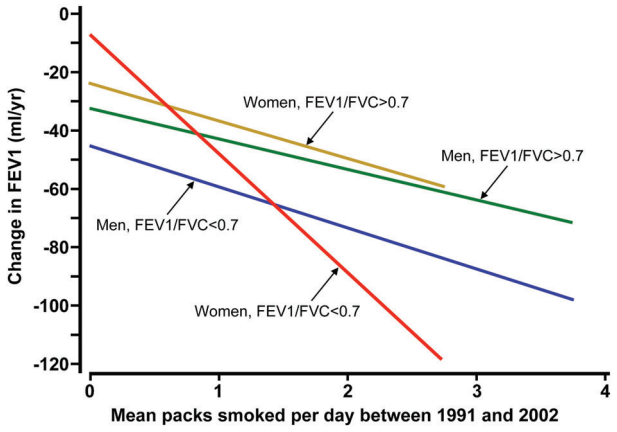

Figure 2. Relationship between mean packs per day and annual change in FEV_1 in men and women: greater smoking-related lung function decline in women compared with men with airflow obstruction. Reproduced from Downs *et al* [96] © 2005 Downs et al; Licensed under Creative Commons Attribution 4.0 International Public License (https://creativecommons.org/licenses/by/4.0/legalcode).

the absence of alpha1-antitrypsin deficiency) and 348 of their first-degree relatives was assembled. Probands with severe early onset COPD were more often female (71%). Among the entire group of first-degree relatives, males and females demonstrated similar lung function. However, among current and ex-smokers, female first-degree relatives had significantly lower FEV_1/FVC values compared with males (82 ± 17% in females versus 87 ± 13% in males, $p = 0.009$) [98,99]. In addition, they more often had severe lung function impairment ($FEV_1 < 40\%$ predicted) compared with male first-degree relatives, suggesting that females are more susceptible to developing severe COPD.

It has been reported that females develop a different type of COPD than males. They demonstrate less severe emphysematous destruction for the same degree of lung function impairment [95,98,99]. However, they have more airway wall remodeling, with thicker small airway walls relative to the lumen [95]. A recent analysis of COPDGene subjects confirmed earlier findings that males display more emphysema than females overall for the same degree of airflow obstruction [100]. However, this was not the case in females with severe early onset COPD, severe emphysema (percentage of lung with low attenuation areas > 25%) and GOLD4 COPD. These three phenotypic subgroups with advanced disease had comparable radiographic emphysema despite fewer pack-years of smoking. This finding that females with severe COPD demonstrate a similar degree of emphysema with less smoke exposure is in line with the notion that a subgroup is particularly susceptible to developing COPD with smoking. The difference in susceptibility to smoking already starts at an early age, as the growth in lung volumes is less in girls compared with boys when they start smoking as adolescents [101].

Several factors might contribute to the increased susceptibility to smoking. A possible explanation is that females have smaller lungs and therefore one pack of cigarettes may represent a higher dose in females than males. Another possible explanation would be different brands of cigarettes smoked or differences in inhalation technique. Finally, hormonal and genetic factors may drive the sex differences observed in COPD. Tam *et al* [102] showed increased airway wall thickness in female compared with male smokers at risk for or with GOLD1 COPD. They recapitulated these findings in a smoke-induced mouse model of COPD, where female mice developed greater airway remodeling, greater small airway resistance, increased TGF-beta activity and a reduced antioxidant response upon smoke exposure. These effects were prevented by ovariectomy and in part also by tamoxifen treatment, indicating a role for sex hormones. The CYP450 pathway, which is important in drug metabolism and detoxification, has been suggested to play a role, as it is known to be affected by estrogen and several enzymes of this family are increased in women (reviewed in [103]). CYP1A1 and CYP1B1 expression increases in cultured bronchial epithelial cells after estrogen stimulation [104] and higher CYP450A1 and CYP450B1 expression has been observed in primary epithelial cells of patients with COPD compared with those without COPD [105]. CYP450 enzymes convert the cigarette smoke constituent naphthalene to the far more toxic intermediate metabolite naphthalene oxide, leading to a rapid death of club cells. Another study reporting different underlying mechanisms of COPD in males and females was carried out by Kohler *et al* [106]. They investigated the bronchoalveolar lavage fluid proteome in smokers with and without COPD and found a total of 19 proteins to be significantly differentially expressed [106]. Interestingly, COPD-associated differential protein expressions were completely driven by the female patients and included cathepsin B (downregulated), ATP synthase (upregulated) and chaperonin (upregulated). Pathway analysis revealed that the lysosome activity pathway was significantly downregulated in females with COPD, which was primarily driven by the decrease in cathepsin B. Downregulation of lysosomal function has been linked to autophagy, a conserved homeostatic pathway for protein degradation and regeneration. There is growing evidence in the literature that autophagy plays an important role in the pathogenesis of COPD [107].

Finally, genetic factors may play a role. In a recent study by Hardin *et al* [108], sex-related dimorphic genetic risk factors were examined in a GWAS meta-analysis using three large COPD cohorts (COPDGene, ECLIPSE, GenKOLS). A genome-wide SNP*sex interaction analysis for COPD was performed. Although no genome-wide significant hits were identified, several variants were identified that approached the prespecified significance threshold of $5*10^{-8}$, including four SNPs located in the *CELSR1* gene, a member of the cadherin superfamily. The top-hit was SNP rs9615358, located in *CELSR1*. *CELSR1* is a known early lung development gene and was found to show higher expression in female compared with male fetal lung tissue, suggesting that in females *CELSR1* may play a role in the susceptibility to COPD with a developmental origin.

Emerging treatment strategies in COPD; potential use of biologics

The chronic airway inflammation in COPD has been traditionally characterized by neutrophilic inflammation. However, a subgroup of patients with COPD has an eosinophilic phenotype [109], based on increased peripheral blood eosinophil counts or increased percentages of eosinophils in induced sputum. Increased blood eosinophil counts in COPD patients have been associated with an increased risk of future exacerbations [110,111]; chronic treatment with inhaled corticosteroids decreases this risk [112–114]. This and other observations have led to the concept of treatable traits in chronic airway diseases [115], as in both asthma and COPD the presence of eosinophilic airway inflammation (as evidence by increased blood eosinophil levels) has

been shown to be a theragnostic biomarker, predicting the therapeutic response to inhaled corticosteroids.

The next step is to investigate whether a precision medicine approach using specific biologics targeting eosinophilic inflammation would be as successful in patients with COPD as in asthmatics. Indeed, in patients with severe eosinophilic asthma, three monoclonal antibodies targeting the IL-5/IL-5 receptor axis have been shown to significantly reduce exacerbation rates [116–118]. Mepolizumab and reslizumab inhibit the ligand IL-5, whereas benralizumab binds to the IL-5 receptor expressed on eosinophils and basophils, inducing apoptosis and thus depleting these cells. In two phase III double-blind, randomized, controlled trials (METREX and METREO), monthly subcutaneous injections of mepolizumab were associated with a lower rate of exacerbations than placebo in patients with COPD and an eosinophilic phenotype [119]. However, whereas mepolizumab reduced exacerbation rates in severe eosinophilic asthma by 50%, the effect of mepolizumab in eosinophilic COPD was very modest (approximately 15% reduction), although the same cut-off values for blood eosinophil counts had been applied as inclusion criteria in these trials for both diseases. Moreover, whereas mepolizumab treatment was associated with significant improvements in quality of life in patients with severe eosinophilic asthma, the effects on patient-reported outcomes, such as SGRQ and the COPD Assessment Test, did not reach the minimal clinically important difference.

Several mechanisms might underlie the very modest effects of mepolizumab in patients with eosinophilic COPD: first, the role of eosinophils in eliciting acute exacerbations in COPD might be less prominent as compared with severe eosinophilic asthma; second, the eosinophilic inflammation might be driven by other mediators than IL-5, such as IL-3, GM-CSF, eotaxins and leukotrienes. As the cytolytic anti-IL5 receptor monoclonal antibody benralizumab depletes eosinophils (completely in blood and significantly in sputum and mucosal tissues), clinical studies with benralizumab can help to disentangle the mechanisms of eosinophilic airway inflammation in COPD, as well as their contribution to acute exacerbations. Interestingly, in two large trials, the GALATHEA and TERRANOVA studies, benralizumab – at different doses (10, 30 or 100 mg s.c.) – did not affect the annualized exacerbation rate as compared with placebo in patients with moderate to very severe COPD, a blood eosinophil count of 220/μl or greater and a history of frequent exacerbations [120]. Therefore, in contrast to the clear clinical benefits of benralizumab treatment in patients with severe eosinophilic asthma, eosinophil depletion in COPD patients did not translate to a reduction in exacerbations. This important

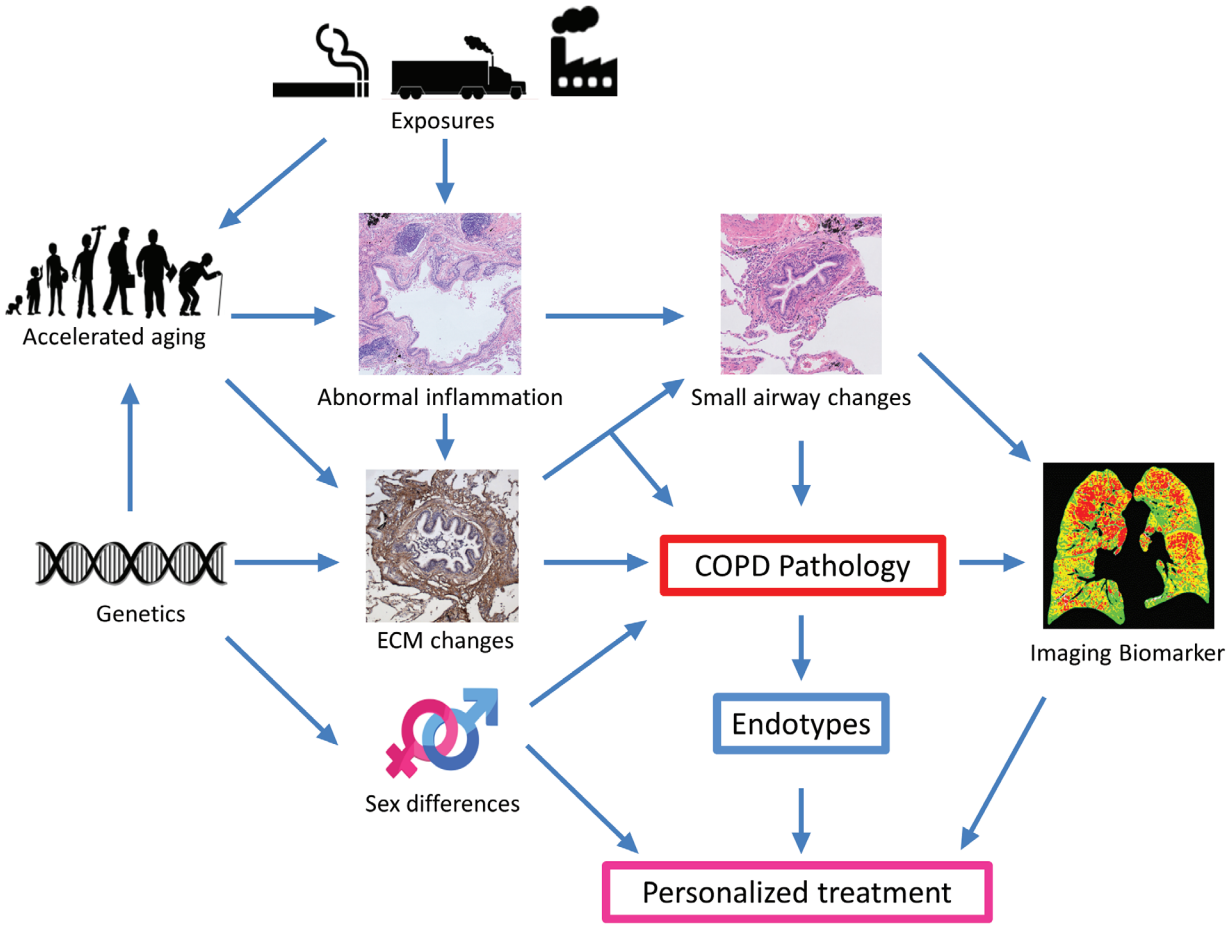

Figure 3. From key contributors to the individual's disease pathology to disease endotyping and personalized treatment in COPD.

discrepancy highlights that the 'treatable traits' concept should not replace but should complement the clinical diagnoses of asthma and COPD, which appear to be dominant in predicting therapeutic responses to targeted biologics in chronic airway diseases.

Take home message

To halt the worrying increase in the burden of COPD, improved and personalized treatment strategies are needed. Genetic risk factors, accelerated aging and sex differences are important contributors to key features of the individual's disease pathology in COPD, i.e. Th2, Th17 and/or autoimmune-type inflammatory responses, ECM changes and disturbed cell–matrix interactions, and small airways disease in COPD (Figure 3). Improved insight into these pathological features and more accurate diagnosis using precision imaging will enable the discovery of new endotypes in COPD that can be targeted by precision medicine in the future.

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Author contributions statement

CAB and WT drafted the initial design. All authors (CAB, MvdB, T-LH, GB, WT) contributed equally to the contents, contributed to adaptations of the concept and design, and approved the final version of the manuscript.

Abbreviations

COPD, chronic obstructive pulmonary disease; FEV_1, forced expiratory volume in the first second; FVC, forced vital capacity; GDF11, growth differentiation factor 11; GOLD, Global Initiative for Chronic Obstructive Lung Disease; GWAS, genome-wide association study; ILC3, group 3 innate lymphoid cells; PRM, parametric response mapping; SGRQ, St-George Respiratory Questionnaire; TIMP, tissue inhibitor of metalloproteinase.

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