Arch. Immunol. Ther. Exp. (2016) 64:177–193
DOI 10.1007/s00005-015-0375-5

REVIEW

Matrix Metalloproteinases and Their Inhibitors in Chronic Obstructive Pulmonary Disease

Zdenka Navratilova[1] · Vitezslav Kolek[2] · Martin Petrek[1]

Received: 19 December 2014 / Accepted: 25 September 2015 / Published online: 26 November 2015

Abstract Chronic obstructive pulmonary disease (COPD) is characterised by irreversible airflow limitation associated with chronic inflammation. Matrix metalloproteinases (MMPs) are proteolytic enzymes that contribute to the inflammatory response in COPD and degrade extracellular matrix components. Their enzymatic activity is inhibited by a four-member family of tissue inhibitors of metalloproteinases (TIMPs). In COPD, the MMP/TIMP network, mainly MMP-9, has been repeatedly observed to be dysregulated at both the local (lung) and systemic levels. Here, we review the findings reported in numerous cross-sectional studies with our primary focus on longitudinal observations in human COPD studies. The data from longitudinal prospective studies on the MMP/TIMP network may lead to the introduction of novel prognostic biomarkers into clinical management of COPD. We address the relationship between the systemic and local lung MMP/TIMP network in COPD patients and briefly describe the involvement of microRNAs. Finally, the role of the MMP/TIMP network in COPD treatment is discussed.

✉ Martin Petrek
martin.petrek@fnol.cz

Zdenka Navratilova
zdenka1navratilova@gmail.com

Vitezslav Kolek
vitezslav.kolek@fnol.cz

[1] Laboratory of Immunogenomics, Department of Pathological Physiology, Faculty of Medicine and Dentistry, Palacky University, Hnevotinska 3, 775 15 Olomouc, Czech Republic

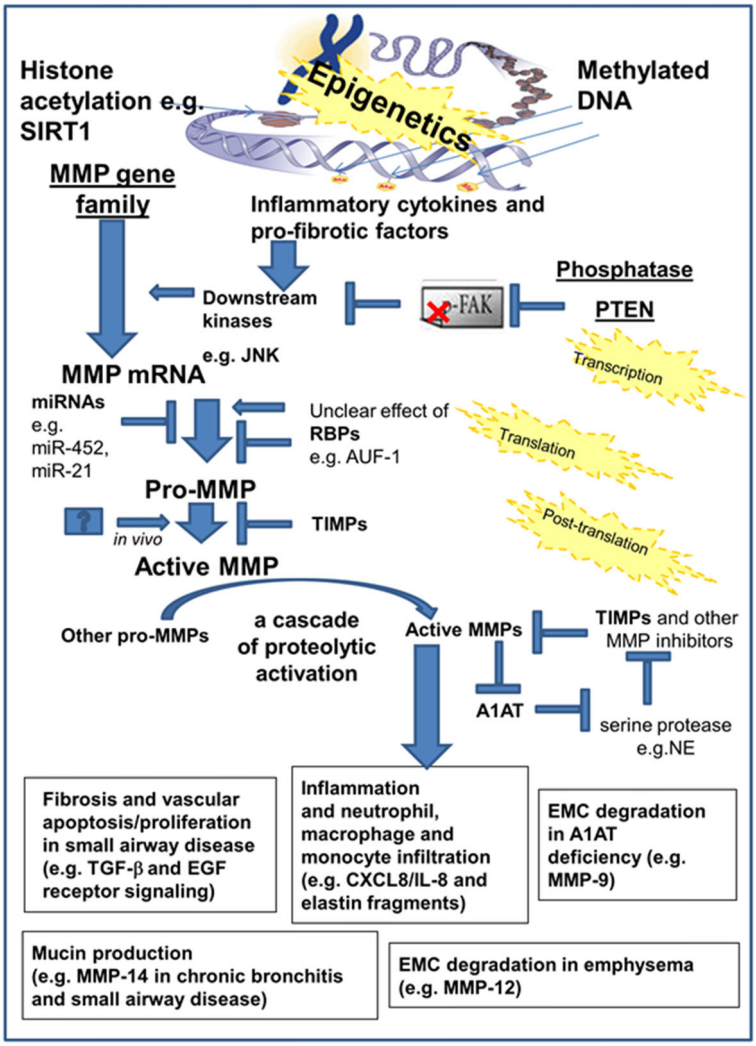
[2] Department of Respiratory Medicine, Faculty of Medicine and Dentistry, Palacky University, Olomouc, Czech Republic

Keywords Chronic obstructive pulmonary disease · Matrix metalloproteinases · Tissue inhibitors of metalloproteinases · MicroRNAs · Local and systemic response · Biomarker

Introduction

Chronic obstructive pulmonary disease (COPD) is characterised by largely irreversible airflow limitation (airflow obstruction) that is associated with chronic inflammation (Brusselle et al. 2011). A leading cause of COPD is cigarette smoking that is assumed to induce the chronic inflammatory response affecting lung tissue. In particular, inflammatory cells infiltrate the surface epithelium with their released products causing permanent irritation to functional tissue. The subsequent tissue repair leads to collagen deposition with accompanying bronchial wall thickening and narrowing of the small airways (Harju et al. 2010; Vestbo et al. 2012, 2013). With disease progression, a diminished capacity to produce extracellular matrix components (ECM) and/or a parallel shift in the protease-anti-protease balance towards degradation results in breakdown of the lung parenchyma and large airway disease—emphysema (Harju et al. 2010; Vestbo et al. 2012, 2013).

Matrix metalloproteinases (MMPs) are zinc-dependent endopeptidases that degrade ECM. Their important role in lung disease has been emphasised in the European Respiratory Journal series "matrix metalloproteinases in lung health and disease" (Chelladurai et al. 2012; Churg et al. 2012; Davey et al. 2011; Loffek et al. 2011). In addition to proteolytic degradation of the lung parenchyma, MMP-associated activity also participates in a number of other processes, including the inflammatory response, mucus



hypersecretion, vascular apoptosis/proliferation, and pro-fibrotic pathways (Churg et al. 2012; Davey et al. 2011; Deshmukh et al. 2009; Loffek et al. 2011; Wang et al. 2008). During inflammation, pro-protein MMPs are activated through a cascade of proteolytic cleavage to yield active MMPs that in turn activate cytokines (e.g. tumour necrosis factor-α) and/or modify the binding interaction between some chemokines and their receptors (Churg et al. 2012; Davey et al. 2011; Loffek et al. 2011) (Fig. 1). Simultaneously, MMPs with collagenase (e.g. MMP-8), gelatinase (e.g. MMP-2 and MMP-9) and elastase activity (e.g. MMP-12) collectively cleave all ECM and are thus actively involved in the degradation of lung parenchyma that leads to the development of emphysema (Davey et al. 2011; Loffek et al. 2011). Initially latent ECM-bound growth factors such as transforming growth factor (TGF)-β are released by ECM degradation, increasing TGF bioavailability to trigger the signalling pathways resulting in fibrotic changes (Davey et al. 2011; Loffek et al. 2011). Importantly, the effect of MMPs may be strengthened by their mutual cascade of enzymatic activation (Davey et al. 2011; Loffek et al. 2011) (Fig. 1).

The broad spectrum of MMP biological activity is regulated at several levels: epigenetically (e.g. by DNA methylation of MMP promoters and histone deacetylation by SIRT1) (Gao and Ye 2008; Loffek et al. 2011; Nakamaru et al. 2009; Philibert et al. 2012; Vlahos et al. 2012; Vucic et al. 2014; Yao et al. 2013), transcriptionally (e.g. PTEN) (Boosani and Agrawal 2013; Shaykhiev et al. 2011), post-transcriptionally by microRNAs (miRNAs) and, importantly, at the level of post-translational modification by natural inhibitors of MMPs (e.g. TIMPs and RECK) (Loffek et al. 2011; Takahashi et al. 1998) (Fig. 1). In this review, we aim to provide an informative overview on MMPs and their inhibitors in COPD with a focus on reviewing longitudinal prospective studies that may lead to

◀**Fig. 1** Role of matrix metalloproteinases (MMPs) and neutrophil elastase (NE) in the pathogenesis of COPD. Cigarette smoke components and other noxious particles induce epigenetic modifications including DNA methylation, histone acetylation and miRNome (miRNAs) that are able to regulate MMP expression directly (Nakamaru et al. 2009; Yao et al. 2013) and indirectly via a broad spectrum of inflammatory cytokines [e.g. tumour necrosis factor (TNF)-α] and growth factors (e.g. TGF-β (Loffek et al. 2011; Philibert et al. 2012). Aberrant DNA methylation, miRNA expression and down-regulated expression of class I–III histone deacetylases (e.g. SIRT1) were also reported in COPD patients (Nakamaru et al. 2009; Vucic et al. 2014; Yao et al. 2013). In addition, SIRT1 reduction was associated with imbalance of TIMP-1 and MMP-9 in COPD patients (Nakamaru et al. 2009; Yao et al. 2013). At the transcriptional level, down-regulated tyrosine phosphatase activity of PTEN (or phosphatase and tensin homologue) can contribute to the increased MMP expression in COPD (Shaykhiev et al. 2011). This pathway may act via insufficient PTEN-associated inhibition of cytoplasmic focal adhesion kinase (FAK) and other downstream kinases that induce MMP-9 expression (Boosani and Agrawal 2013). In addition, the decreased expression of PTEN is related to its gene hypermethylation in COPD (Vucic et al. 2014). Post-transcriptional regulation of MMPs is mediated by miRNAs and RNA-binding proteins that together recognise and subsequently affect the stability and decay of MMP mRNAs. MMP proteins are then synthesised as inactive pro-proteins (zymogens) that are activated by serine proteinases or other members of the MMP family in a cascade of proteolytic cleavage resulting in amplified activation. However, the in vivo inducers of pro-MMPs have not been described to date. Pro-MMPs and active MMPs are inhibited by a four-member family of TIMPs (TIMP1-4) and other natural inhibitors [e.g. α2-macroglobulin and reversion-inducing-cysteine-rich protein with kazal motifs (RECK)]. In COPD pathogenesis, increased proteolytic activity of MMPs contributes to pulmonary inflammation and neutrophil/macrophage/monocyte chemotaxis by pro-cytokine activation (e.g. "shedding" of cellular membrane-bound pro-TNF-α) and aminoterminal truncation of neutrophil chemokines leading to several-fold potentiation of their biological activity [e.g. CXCL8/interleukin (IL)-8]. However, MMPs may also have an anti-inflammatory effect in COPD as they inactivate the chemotactic activity of CXCL1 and CXCL4 and generate CC7 chemokine receptor antagonists (Loffek et al. 2011). Three common forms of COPD are emphysema, small airway disease and chronic bronchitis. The protease-mediated degradation of extracellular matrix components (ECM) is generally accepted to contribute to emphysema development. MMPs with collagenase (e.g. MMP-8), gelatinase (e.g. MMP-2 and MMP-9) and elastase activity (e.g. MMP-12) collectively cleave all ECM. In addition, MMPs (such as MMP-8, MMP-9, MMP-12 and MMP-13) degrade serine protease inhibitors "serpins", especially alpha-1 antitrypsin (A1AT) that is the major inhibitor of neutrophil elastase (NE)]. To amplify the proteolytic effect, NE inhibits TIMPs. Initially latent ECM-bound growth factors such as pro-fibrotic factor (TGF)-β are released during the ECM degradation and thus MMPs increase their bioavailability to induce signalling pathways leading to pro-fibrotic changes in small airway diseases. Furthermore, MMPs may contribute to mucus hypersecretion and vascular remodelling through mobilisation of mitogenic and anti-apoptotic epidermal growth factor (EGF) (Deshmukh et al. 2009)

the introduction of novel prognostic biomarkers into clinical management of COPD. We also briefly review selected aspects of MMP/TIMP network regulation and its implications for treatment.

MMP/TIMP Network in Cross-Sectional Studies of COPD

The involvement of the MMP/TIMP network in remodelling processes in the small airways has been reported by numerous independent studies, utilising bronchoalveolar lavage (BAL) samples (Segura-Valdez et al. 2000; Vlahos et al. 2012), airway epithelial cells (Deshmukh et al. 2009), induced sputum (Chaudhuri et al. 2012, 2013; Ilumets et al. 2007; Paone et al. 2011; Simpson et al. 2013), lung tissue (Gosselink et al. 2010; Noguera et al. 2012) and exhaled breath condensate (Kwiatkowska et al. 2012). The up-regulation of MMPs was observed at both mRNA and protein levels with increased enzymatic activities in both the upper airways from induced sputum (Culpitt et al. 2005) and the lower airways from BAL (Vlahos et al. 2012). In addition, increased systemic concentrations of circulating MMP-1 to MMP-3 and MMP-7 to MMP-10 were observed in COPD (Tables 1, 2).

A problem frequently encountered in these studies is an underestimation of the impact of cigarette smoking on MMP levels. As COPD usually affects people in their middle or old ages, patients enrolled in current case–control studies frequently have had prolonged exposure to cigarette smoke with high numbers of individual pack-years. By contrast, few older people exposed to an equivalent smoking load do not suffer from some form of COPD-related disease and are eligible, therefore, to be considered as healthy controls. In this regard, it has been common in non-matched case–control studies for current, never and ex-smokers to be analysed together (Finlay et al. 1997; Navratilova et al. 2012; Pinto-Plata et al. 2012). If the matching of the percentages of representation of the three smoking statuses between case and controls was close, the healthy controls were usually significantly younger and with a lower smoking load compared to the COPD patients (Culpitt et al. 2005; Pinto-Plata et al. 2007; Segura-Valdez et al. 2000).

Although the lingering difficulties associated with COPD and healthy subject recruitment are understandable, inappropriately matched cohorts should raise a suspicion that the consistently observed elevation in MMP production is not necessarily associated with COPD pathogenesis but more with age or cigarette smoking. Indeed, subsequent studies by Simpson et al. and others have shown that the MMP/TIMP network may be influenced by both age and cigarette smoking (Table 3) and the response to cigarette smoking is more pronounced with increasing age (Ilumets et al. 2011; Simpson et al. 2013; Snitker et al. 2013). The major observations on the relationship between smoking and MMP/TIMP network are summarised in Table 3. Here, some conflicting data are also mentioned to show that the effect of smoking has not yet been characterised in detail.

Table 1 Matrix metalloproteinases (MMPs) in COPD (stable COPD, ECOPD or emphysema)

MMP	COPD	Expression	Biological material	*n*	Method	References
MMP-1 (interstitial collagenase/collagenase 1)	COPD	↑	Sputum	15	ELISA	Culpitt et al. (2005)
		–	Sputum	24	Fluorescence microspheres	Ropcke et al. (2012)
		↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		–	Serum	24	Fluorescence microspheres	Ropcke et al. (2012)
		↑°	Lung tissue surrounding small bronchioles	8	RT-PCR	Gosselink et al. (2010)
	Emphysema	–	BAL	101	ELISA	D'Armiento et al. (2013)
		↑	Alveolar macrophages, conditioned media	10	RT-PCR	Finlay et al. (1997)
		↑	Lung parenchyma	23	RT-PCR + ELISA + SDS-PAGE	Imai et al. (2001)
		↑°	Alveolar macrophages	54	RT-PCR	Wallace et al. (2008)
MMP-2 (gelatinase A)	COPD	↑	BAL	8	Gelatin zymography	Segura-Valdez et al. (2000)
		↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		–	Serum	24	Fluorescence microspheres	Ropcke et al. (2012)
		↓°	Lung tissue surrounding small bronchioles	8	RT-PCR	Gosselink et al. (2010)
		↓°	Small bronchioles	8	RT-PCR	Gosselink et al. (2010)
	Emphysema	–	Alveolar macrophages	10	RT-PCR + gelatin zymography	Finlay et al. (1997)
MMP-3 (stromelysin 1)	COPD	↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		–	Serum	160	Fluorescence microspheres	Loza et al. (2012)
MMP-7 (matrilysin)	COPD	–	Sputum	24	Fluorescence microspheres	Ropcke et al. (2012)
		↑	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
		↑	Serum	74	Fluorescence Microspheres	Navratilova et al. (2012)
MMP-8 (neutrophil collagenase/collagenase 2)	COPD	↑	Sputum	23	ELISA	Ilumets et al. (2007)
		↑	Sputum	17	Immunocapture assay assessing active/total levels	Vernooy et al. (2004)
		↑	Sputum	15	ELISA	Culpitt et al. (2005)
		↑	Plasma	201	Protein array	Dickens et al. (2011)
		↑	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
		↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		↑°	Plasma	201	Protein array	Dickens et al. (2011)

Table 1 continued

MMP	COPD	Expression	Biological material	*n*	Method	References
	Emphysema	↑°	Plasma	201	Protein array	Dickens et al. (2011)
	ECOPD	↑	Sputum	10	Immunofluorometry	Ilumets et al. (2008)
	COPD	↑	EBC	45	ELISA	Kwiatkowska et al. (2012)
		–	BAL	24	Fluorescence microspheres	Ropcke et al. (2012)
		↑	Alveolar macrophages	8	ELISA + zymography	Russell et al. (2002)
		↑	Sputum	42	ELISA	Paone et al. (2011)
		↑	Sputum	23	ELISA	Ilumets et al. (2007)
		↑	Sputum	15	ELISA + zymography	Culpitt et al. (2005)
		↑	Sputum	17	Immunocapture assay assessing active/ total MMP	Vernooy et al. (2004)
		–	Sputum	22	ELISA	Baines et al. (2011)
		↑	Sputum	20	Gelatin zymography	Cataldo et al. (2000)
		–	Sputum	24	Fluorescence microspheres	Ropcke et al. (2012)
		↑	BAL	8	Gelatin zymography	Segura-Valdez et al. (2000)
		–	Sputum and cells	53	ELISA, FRET assay	Chaudhuri et al. (2013)
		–	Blood granulocytes	11	Zymography	Cataldo et al. (2001)
		↑	Blood neutrophiles	13	ELISA	Baines et al. (2011)
		–	Plasma	201	Protein array	Dickens et al. (2011)
		–	Plasma	20	ELISA	Shaker et al. (2008)
		↑	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
		↑	Serum	23	ELISA	Brajer et al. (2008)
		↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		↑	Serum	70	ELISA	Bolton et al. (2009)
		↑	Serum	45	ELISA	Kwiatkowska et al. (2012)
		–	Serum	24	Fluorescence microspheres	Ropcke et al. (2012)
		–	Serum	72	ELISA	Higashimoto et al. (2005)
		↑°	BAL	58	ELISA + gelatin zymography	Vlahos et al. (2012)
		↑°	Lung tissue surrounding small bronchioles	8	RT-PCR	Gosselink et al. (2010)

Table 1 continued

MMP	COPD	Expression	Biological material	*n*	Method	References
		↑°	Sputum cells	27	DNA array	Chaudhuri et al. (2013)
		↑°	Plasma	201	Protein array	Dickens et al. (2011)
		↑°	Serum	73	ELISA	Higashimoto et al. (2009)
		↓°	Serum	253	ELISA	Pinto-Plata et al. (2012)
	Emphysema	–	BAL	101	ELISA	D'Armiento et al. (2013)
		–	Lung parenchyma	23	RT-PCR	Imai et al. (2001)
		↑	Alveolar macrophages	10	RT-PCR	Finlay et al. (1997)
		↑°	Sputum	53	ELISA, FRET assay	Chaudhuri et al. (2013)
		↑°	Plasma	201	Protein array	Dickens et al. (2011)
		↓	Plasma	101	Fluorescence microspheres	D'Armiento et al. (2013)
	ECOPD	↑	EBC	17	ELISA	Kwiatkowska et al. (2012)
		↑	Sputum	12	Gelatin zymography	Mercer et al. (2005)
		–	Plasma	90	Protein array	Hurst et al. (2006)
		–	Plasma	33	Protein array	Dickens et al. (2011)
		↑	Serum	17	ELISA	Kwiatkowska et al. (2012)
MMP-10 (stromelysin 2)	COPD	↑°	Small bronchioles	8	RT-PCR	Gosselink et al. (2010)
		↑°	Lung tissue surrounding small bronchioles	8	RT-PCR	Gosselink et al. (2010)
		↑	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
MMP-12 (macrophage metalloelastase)	COPD	↑	Sputum	28	ELISA + casein zymography	Demedts et al. (2006)
		–	Sputum	24	Fluorescence microspheres	Ropcke et al. (2012)
		–/↑	Sputum/cells	53	ELISA, FRET assay/ RT-PCR	Chaudhuri et al. (2012)
	Emphysema	–	BAL	66	Fluorescence microspheres	D'Armiento et al. (2013)
		–	Alveolar macrophages	10	RT-PCR	Finlay et al. (1997)
		0	Macrophage-conditioned media	10	Casein zymography	Finlay et al. (1997)
		0	Lung parenchyma	23	RT-PCR	Imai et al. (2001)
		↑°	Sputum	53	ELISA, FRET assay	Chaudhuri et al. (2012)
MMP-13 (collagenase 3)	COPD	↑	Lung tissue	7	Western blot and immunohistochemistry	Lee et al. (2009)
		0	Lung tissue	10	Immunohistochemical analysis	Segura-Valdez et al. (2000)
MMP-14	COPD	↑	Airway epithelium	2	Immunostaining	Deshmukh et al. (2009)

Table 1 continued

MMP	COPD	Expression	Biological material	*n*	Method	References
Neutrophil elastase[a]	COPD	↑	Sputum	42	ELISA	Paone et al. (2011)
		↑	Sputum	22	ELISA	Baines et al. (2011)
		↑	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
		↑°	BAL	58	NE specific substrate	Vlahos et al. (2012)
	Emphysema	0	Alveolar macrophages	10	RT-PCR	Finlay et al. (1997)
		↑	Macrophage-conditioned media	10	NE-sensitive chromogenic peptide	Finlay et al. (1997)
	ECOPD	↑	Sputum	10	Synthetic peptide substrate and spectrophotometry	Ilumets et al. (2008)

° Primarily investigated the correlation with disease progression assessed as the extent of airflow obstruction or emphysema

↑ up-regulation, ↓ down-regulation, – no significant difference, 0 not detected, *EBC* exhaled breath condensate, *BAL* bronchoalveolar lavage, *RT-PCR* real-time polymerase chain reaction, *ELISA* enzyme-linked immunosorbent assay

[a] Belong among serine proteases

Whatever the effect of cigarette smoke, increased concentrations of MMP-9 are related to COPD pathogenesis based on two case–control studies of closely pack-year-matched cohorts (Paone et al. 2011; Russell et al. 2002).

By contrast, other clinical studies provide data that are in conflict with the observations that MMP-9 production increases in smokers compared to non-smokers culminating in the development of COPD. These studies failed to confirm increased MMP-9 levels in COPD but suggested that smoking history alone could explain MMP-9 increases at least in part due to a higher smoke load in COPD (pack-years or mean smoking history) than in healthy control smokers (Baines et al. 2011; D'Armiento et al. 2013; Ropcke et al. 2012). Furthermore, anti-inflammatory therapy with inhaled bronchodilators/corticosteroids should be considered as a potential confounder explaining the inconsistent observations (Baines et al. 2011). Alternatively, it may be only the level of MMP-9 in response to specific pro-inflammatory stimuli (e.g. bacterial infection) that plays a critical role in the development of COPD (Ishii et al. 2014); both of these concepts are discussed later. The presence of specific phenotype and other comorbidities among COPD patients could be another source of inconsistent observations as MMP-9 and other MMPs have been reported to be affected by conditions other than airflow obstruction (Bolton et al. 2009; Chaudhuri et al. 2013; Chelladurai et al. 2012; Pinto-Plata et al. 2007). In particular, Bolton et al. (2009) observed MMP-9 to be decreased in COPD patients without osteoporosis compared to those with osteoporosis. Pulmonary hypertension among COPD patients could also represent a relevant bias as increased MMP levels have been reported in pulmonary hypertension (Chelladurai et al. 2012). Furthermore, emphysema grades have been reported to correlate with increasing concentrations of MMP-1 and MMP-12 (Ishii et al. 2014; Wallace et al. 2008) and frequent exacerbations may be related to increased MMP-9 levels (Pinto-Plata et al. 2007) (Table 1). In addition to emphysema and frequent exacerbations, chronic bronchitis with(out) mucus hypersecretion is another clinically relevant and common form of COPD (Global Strategy for the Diagnosis, Management and Prevention of COPD, Global Initiative for Chronic Obstructive Lung Disease (GOLD) 2015; available at: http://www.goldcopd.org/). MMP elevation has been reported to induce mucin expression in pulmonary mucoepidermoid carcinoma (MMP-14) and to be related to mucus hypersecretion in asthmatic patients (MMP-9) (Deshmukh et al. 2009; Ko et al. 2005). Vascular endothelial growth factor, a growth factor whose bioavailability may be regulated by MMP proteolytic activity, was decreased in emphysema compared to chronic bronchitis (Kanazawa 2007). However, patients with COPD and chronic bronchitis were not stratified in the study by Vignola et al. (1998) who focused on MMPs. In addition, MMP-8 concentrations were reported to be lower in non-symptomatic smokers compared to symptomatic smokers with cough and sputum but no airway obstruction (Ilumets et al. 2007). There are, however, no MMP data on possible differences among emphysema, other small airways' diseases and chronic bronchitis with mucus hypersecretion in COPD patients. In this context, comparisons of different forms of COPD may be challenging in

Table 2 Tissue inhibitors of matrix metalloproteinases (TIMPs) in COPD (stable COPD, ECOPD and emphysema)

TIMP	COPD	Expression	Biological material	*n*	Method	References
TIMP1	COPD	↑	EBC	45	ELISA	Kwiatkowska et al. (2012)
		↑	BAL	24	ELISA	Ropcke et al. (2012)
		–	Alveolar macrophages	8	ELISA	Russell et al. (2002)
		↑	Induced sputum	20	ELISA	Cataldo et al. (2000)
		–	Induced sputum	15	ELISA	Culpitt et al. (2005)
		–	Induced sputum	24	ELISA	Ropcke et al. (2012)
		↓	Plasma	20	ELISA	Shaker et al. (2008)
		↑	Serum	160	Fluorescence microspheres	Loza et al. (2012)
		↑	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
		↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		↑	Serum	45	ELISA	Kwiatkowska et al. (2012)
		↑	Serum	72	ELISA	Higashimoto et al. 2005)
		↓°	Lung tissue surrounding small bronchioles	8	RT-PCR	Gosselink et al. (2010)
	ECOPD	↑	EBC	17	ELISA	Kwiatkowska et al. (2012)
		↓	Sputum	12	ELISA	Mercer et al. (2005)
		–	Plasma	33	Protein array	Dickens et al. (2011)
		–	Serum	17	ELISA	Kwiatkowska et al. (2012)
	emphysema	–	BAL	101	ELISA	D'Armiento et al. (2013)
		↓	Plasma	101	Fluorescence microspheres	D'Armiento et al. (2013)
TIMP2	COPD	–	Induced sputum	24	ELISA	Ropcke et al. (2012)
		–	Plasma	20	ELISA	Shaker et al. (2008)
		–	Serum	47	Protein microarray	Pinto-Plata et al. (2007)
		–	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)
		↓°	Small bronchioles	8	RT-PCR	Gosselink et al. (2010)
TIMP3[a]	emphysema	↑	Lung fibroblasts	3	cDNA array	Muller et al. (2006)
TIMP4	COPD	↑	Serum	74	Fluorescence microspheres	Navratilova et al. (2012)

° Primarily investigated the correlation with disease progression assessed as the extent of airflow obstruction or emphysema

↑ up-regulation, ↓ down-regulation, – no significant difference, *EBC* exhaled breath condensate, *BAL* bronchoalveolar lavage, *RT-PCR* real-time polymerase chain reaction, *ELISA* enzyme-linked immunosorbent assay

[a] TIMP3 protein is localised entirely to the extracellular matrix

Table 3 The effect of age and cigarette smoking on the MMP/TIMP network

Local lung MMP/TIMP network
MMP-9 level increases with age (cut-off 55 years of age) (Simpson et al. 2013)
Long-term cigarette smoking (for 30 years and 31 pack-years) stimulates sputum cell expressions of MMP-9 and MMP-12, although this effect was not observed at their protein levels (Chaudhuri et al. 2012, 2013)
Active smoking increases the elastase activity and the protein concentrations of MMP-1 and MMP-9 (D'Armiento et al. 2013; Ilumets et al. 2007)
Two inhibitors (TIMP1 and TIMP2) are upregulated in smokers (Chaudhuri et al. 2012)
Circulating MMP/TIMP network
No or mild effect of ageing may be seen when comparing healthy young (aged 18–30 years) with older individuals (aged 40–90 years) (Bonnema et al. 2007; Ilumets et al. 2011)
Short-term smoking (<10 years) does not have to significantly modify circulating MMP-9 or TIMP1 (Ilumets et al. 2011)
Long-term smoking (>10 years) increases both MMP-9 and TIMP1 and strengthens the mild effect of ageing in terms of MMP-9 and TIMP1 elevations (Ilumets et al. 2011, 2007)
In healthy individuals aged 40 years, MMP-9 is 25 % higher in current than never smokers (Snitker et al. 2013)

the light of their frequent co-occurrence and generally require surgical lung biopsy for detailed analysis of the nature of the small airways disease (Burgel et al. 2013). Similarly, current knowledge on the MMP/TIMP network in COPD has been mostly limited to only some MMPs and their inhibitors, especially MMP-9. Given the overlapping enzymatic properties of MMPs, their combined assessments could be more useful for future clinical practice than focussing only on individual measurement of MMP-9 (Paone et al. 2011; Thomsen et al. 2013).

MMP-9 as a Predictive Biomarker of Exacerbations of COPD in Longitudinal Studies

In clinical practice, routine measurement of laboratory biomarkers is justifiable if they are able to predict the future course of disease and thus aid the early initiation of treatment with better clinical outcomes. In COPD, clinical studies aim to identify predictive biomarkers of frequent acute exacerbations of COPD (ECOPD) that greatly influence disease progression. Here, longitudinal prospective studies may bring new clinically relevant information. The longitudinal prospective observations, which use less invasive biological materials such as induced sputum and peripheral blood, would be most likely to be utilised clinically.

Regarding sputum MMPs, two longitudinal analyses of paired data obtained from relatively small numbers (8–12 subjects) of COPD patients showed increased concentrations of MMP-8 and MMP-9 during ECOPD compared to stable COPD (Ilumets et al. 2008; Mercer et al. 2005). Assuming that ECOPD-associated MMP increases may, to some extent, persist in the subgroup of stable COPD patients with worse prognosis such as those with frequent ECOPD events, sputum MMP-8 and MMP-9 could be two interesting candidates for longitudinal perspective studies. So far, however, no sputum MMP has been a subject of longitudinal analysis focused on the prediction of frequent ECOPD.

Regarding systemic MMPs, circulating MMP-9 has been investigated in two longitudinal prospective studies and suggested to be a predictive biomarker of frequent ECOPD in COPD (Pinto-Plata et al. 2007) and emphysema patients with A1AT deficiency (AATD) (Omachi et al. 2011). In emphysema patients with AATD, Omachi et al. (2011) measured plasma MMP-9 at 0 (baseline), 3- and 6-month time points to study the correlation with the subsequent frequency of ECOPD. Increased concentrations of plasma MMP-9 predicted an annualised average of 0.27 additional COPD exacerbations. Importantly, the MMP-9 level retained its role as a predictor of subsequent COPD exacerbations when controlled for the presence/absence of prior COPD exacerbations. Although this association must be confirmed in independent cohorts, Omachi et al. (2011) were likely able to achieve more stable and relatively reliable findings due to repeated measurements of MMP-9 over the study timeline. We can speculate that this approach may reduce the impact of some difficult to control confounders and thus provides new avenues in longitudinal investigation searching for clinically predictive biomarkers (Fig. 2).

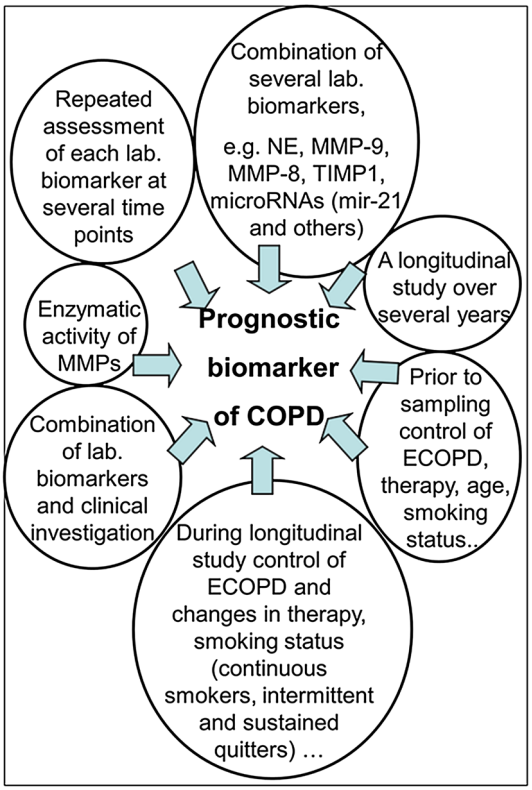


Fig. 2 Schematic recommendations for future longitudinal studies based on current cross-sectional studies and pilot longitudinal observations on MMPs and their inhibitors in COPD. Taking into consideration the complex character of the MMP/TIMP network, the combined assessments of several MMPs (e.g. MMP-9 with MMP-8, MMP-12 and other proteases) and their ratios to TIMP inhibitors (e.g. TIMP4) or alternatively direct assessment of proteolytic activities could provide additional information on COPD pathogenesis. The repeated measurements throughout the time of any study may reduce those confounders that are otherwise difficult to control and thus provide new avenues for longitudinal investigations aiming at identifying clinically predictive biomarkers. The longitudinal studies over several years that would continuously record any changes in smoking habits, therapeutic effects and exacerbations (ECOPD) are highly desirable in COPD

MMP-9 as a Predictive Biomarker of COPD Progression in Longitudinal Studies

Regarding COPD progression, concentrations of MMP-1, MMP-9 and MMP-12 are reported to increase with increased extent of emphysema and progressive airflow obstruction (Ishii et al. 2014). This relationship is based on

several reports from correlation analyses in COPD patients with different disease stages (Ishii et al. 2014; Navratilova et al. 2012; Pinto-Plata et al. 2007).

Several pilot studies have attempted to extend these baseline observations and monitored disease progression with time (Fig. 2). Progressive airflow obstruction was determined as a rate of decline in lung function by quantifying the difference in FEV1 between baseline and follow-up respiratory measurements. Local lung concentrations of neither MMP-9, MMP-1 nor MMP-12 or their inhibitor TIMP1 could predict decline in lung function after 0.5–2 years (D'Armiento et al. 2013; Paone et al. 2011). Circulating MMP-9 was useful for predicting pulmonary parenchymal destruction in one study (Omachi et al. 2011). To predict airflow obstruction, three studies investigated circulating MMP-9 with conflicting observations (D'Armiento et al. 2013; Higashimoto et al. 2009; Omachi et al. 2011). In the prognostic study by D'Armiento et al. (2013), plasma MMP-1 and TIMP1 levels correlated poorly with change in FEV1 at 6, 9 and 18 months' follow-up. A broadly discussed question is whether the length of these study periods and the number of recruited patients were sufficient to reliably assess a decline in lung function because FEV1 data generally show a slow decline with high variability within and across patients with COPD (Omachi et al. 2011). Therefore, there is a demand for a longitudinal study consecutively monitoring the decline in lung function in an adequately sized cohort of COPD patients for several years.

Park et al. (2013) suggested that the total duration of a longitudinal study should include a longer run in phase before spirometry data or sample collection to eliminate the possibly confounding effects of smoking cessation and the impact of newly introduced therapy (Tashkin et al. 2008) (Fig. 2). They have even attempted to define smoking status for 2 years before spirometry. Taking this extra time into account is important because a newly diagnosed COPD patient is likely to quit cigarette smoking and at the same time, new therapy is usually prescribed by his or her physician. For a short period, both smoking cessation and the first therapeutic intervention improve FEV1 and thus, baseline spirometry measurement may not reflect the real situation. As a consequence, COPD course, determined by the decline in FEV1 between baseline and follow-up respiratory measurements, may show excessively accelerated progression of airflow obstruction that does not represent the actual extent of progression in COPD patients.

To control the temporary effects of smoking cessation on lung function, D'Armiento et al. (2013) enrolled only emphysema patients who had abstained from tobacco use for at least 6 months. In addition to the effect of cigarette smoking on lung function, the increased levels of MMP-9 in active smokers were shown to persist for up to 6 months after smoking cessation (Louhelainen et al. 2010). The long-term lingering effect of active smoking may, therefore, affect both MMP-9 concentration and the baseline spirometry measurement.

Another underestimated aspect in longitudinal observation may be that some COPD patients, probably those who suffer from relatively mild respiratory difficulties, re-start cigarette smoking during the study period. Long-term monitoring of smoking habits before spirometry and sample collections could increase the probability of consistent smoking behaviour for the whole study period and thus reduce the effect of active cigarette smoking on both MMP production and lung function (Louhelainen et al. 2010) (Fig. 2).

Variability in Local Response

To be able to predict the course of COPD, an ideal prognostic biomarker must exhibit great consistency over time and should be minimally affected by factors other than COPD progression (Sturgeon et al. 2010). Taking into consideration the great variability of sputum content, Aaron et al. (2010) conducted a longitudinal study where sputum samples were collected at two-week intervals at three time points. Over this time period, sputum TIMP1 showed good stability among the putative sputum biomarkers available for stable COPD (Aaron et al. 2010); however, this was not confirmed by other studies (D'Armiento et al. 2013; Ropcke et al. 2012). One explanation for this inconsistency may be variability due to the presence/absence of airway infection. The existence of specific expression profiles associated with bacterial or viral acute ECOPD has been well described (Bafadhel et al. 2011; Gao et al. 2013). Also, in stable COPD patients without acute ECOPD symptoms, these infections may induce inflammatory responses similar to acute ECOPD (Beasley et al. 2012; Hoenderdos and Condliffe 2013; Millares et al. 2012; Sethi et al. 2006). The presence/absence of airway infection might, therefore, be another source of inter-subject variability in stable COPD and may even contribute to poor intra-subject repeatability of sputum biomarker measurements during the course of a year (Bafadhel et al. 2011; Beasley et al. 2012; Jenkins et al. 2012; Millares et al. 2012; Ropcke et al. 2012).

Enzymatic Activities of Circulating MMPs

The pathological relevance of up-regulated MMPs requires functional study as protein levels do not seem to reflect overall enzymatic activity (Lowrey et al. 2008). Table 1 shows some MMPs whose enzymatic activity has been measured in COPD lung. However, there is limited

information on direct assessment of specific enzymatic activity at the systemic level (Higashimoto et al. 2005). In particular, Higashimoto et al. (2009) measured proteolytic activity of MMP-9 in serum and failed to find any difference between COPD patients and controls. Circulating inhibitors of MMPs (TIMP1 and TIMP4) could, therefore, be speculated to compensate for increased serum concentration of MMPs in COPD (Loza et al. 2012; Navratilova et al. 2012) (Table 2).

On the other hand, in patients with airflow obstruction, an insufficient inhibition of systemic MMPs may be hypothesised based on the increased ratios of MMPs to the inhibitor TIMP1 (Ilumets et al. 2007; Olafsdottir et al. 2010). The presence of MMP-degraded substrates also supports an increased proteolytic activity of MMPs at the systemic level in COPD. In particular, elastin fragments, known to be generated by MMP-9 and MMP-12, have been shown to be increased in the serum of patients with COPD (Skjot-Arkil et al. 2012). In addition to the serum and lungs, degraded products of elastin and collagen have also been demonstrated in increased concentrations in the urine (Devenport et al. 2011; Leeming et al. 2012; Ma et al. 2007). Furthermore, cutaneous expression of MMP-9 correlating with increased degradation of skin elastin may result from increased activity of MMP-9 at the systemic level in COPD patients (Maclay et al. 2012).

The direct assessment of enzymatic activity in the circulation could provide additional information on COPD pathogenesis and explain some inconsistent observations in the prevailing reports showing up-regulated MMP-9 concentrations in COPD (Dickens et al. 2011; Pinto-Plata et al. 2012).

Relationship between Local and Systemic Responses

Several members of the MMP/TIMP network and other proteases have been reported to be present at increased concentrations in local lung compartments and similar up-regulation was also observed at the systemic level in COPD (Finlay et al. 1997; Ilumets et al. 2007; Navratilova et al. 2012; Pinto-Plata et al. 2007). Based on these separate observations, it was attractive to speculate that clinical usage of non-invasive peripheral blood samples could yield highly desirable information on local inflammation ongoing in the less accessible lower airways or the lung tissue. Ropcke et al. (2012), therefore, evaluated the direct relationship between the local lung and circulating MMP/TIMP networks in the same COPD patients: MMP-9 and its ratio to TIMP1 were correlated when the two lung compartments (induced sputum and BAL samples) were compared but not when lung compartments were compared with serum concentrations. The poor correlation between the serum and lung MMPs/TIMP networks is in line with the relationship observed for other inflammatory biomarkers and supports the existence of an independent systematic component in COPD (Wouters et al. 2009).

Involvement of miRNAs (miRNome) in the Regulation of the MMP/TIMP Network in COPD

Cigarette smoking is assumed to induce and potentiate the release of MMPs in the lungs of smokers (Ilumets et al. 2007; Louhelainen et al. 2010). By contrast, cigarette smoking elicits the opposite effect on the miRNome, with the reduction being more pronounced in heavy smokers compared to light smokers (Graff et al. 2012). This inverse association with cigarette smoking is in line with the biological role of the miRNome, composed of numerous small single-stranded (20–24 nucleotides long) non-coding RNAs termed miRNAs that bind to complementary sequences within targeted mRNAs and thus inhibit their translation (Graff et al. 2012). In this context, it has been reported that almost half of the up-regulated proteins in smokers could be explained by the cigarette smoking-induced down-regulation of miRNAs resulting in translational de-repression.

The computational prediction of miRNA targets using TarBase, TargetScan and other databases showed several specific miRNAs to be potential inhibitors of MMP expression (Mu and Zhang 2012; Vergoulis et al. 2012). Some of these were also identified in experiments with tumour and other cell-lines in vitro (Van Pottelberge et al. 2011; Xu et al. 2012). This review addresses specific miRNAs involved in silencing MMP transcripts with emphasis on those that are deregulated in COPD or in smokers (Table 4).

Graff et al. (2012) compared the expression profiles of miRNAs between healthy smokers and non-smokers and reported miR-452 targeting MMP-12 in a human acute monocytic leukaemia cell line as the most down-regulated miRNA in smokers as compared with non-smokers. Importantly, expression of miR-452 was negatively correlated with MMP-12 expression in alveolar macrophages (Graff et al. 2012). Thus, miR-452 represents the first in vivo validated miRNA involved in silencing MMP gene expression. In COPD patients, the expression profile of other miRNAs in alveolar macrophages has not been investigated to date. Among other biological samples, COPD studies on lung tissue and blood samples have reported a distinct profile of down-regulated miRNAs (Ezzie et al. 2012; Leidinger et al. 2011) that did not include miR-452. However, even if down-regulation of miR-452 was not present in the lung tissues or blood of COPD patients, miR-452 may still play a role in COPD

Table 4 MiRNA targeting MMPs/TIMPs that are deregulated in smokers or COPD patients

miR	Up- or down-regulation	Cellular source	References	Relationship between microRNA and the MMP/TIMP network	MMP/TIMP	Cell line used for validation of targets	References
miR-216b	↑	Blood cells	Leidinger et al. (2011)	↓	MMP14	Murine pancreatic tumour cell line	Ali et al. (2012)
miR-92a	↑	Blood cells	Leidinger et al. (2011)	↓	MMP1	Normal dermal fibroblasts	Sing et al. (2012)
	–	Bronchoalveolar cells	Molina-Pinelo et al. (2014)				
miR-221 (/222)	↑×	Alveolar macrophages	Graff et al. (2012)	↓	TIMP3 and PTEN and MMP1	Breast carcinomas and megakaryoblastic leukaemia cell line and tongue squamous cell carcinoma cell line	Garofalo et al. (2009), Liu et al. (2009), Lu et al. (2011)
	↓	Induced sputum supernatant	Van Pottelberge et al. (2011)				
	↓×	Induced sputum supernatant	Van Pottelberge et al. (2011)				
	–	Bronchoalveolar cells	Molina-Pinelo et al. (2014)				
miR-200a	↓×	Alveolar macrophages	Graff et al. (2012)	↑	MMP13 and TIMP1	Human stellate cell line	Murakami et al. (2011)
miR-452	↓×	Alveolar macrophages	Graff et al. (2012)	↓	MMP12	Alveolar macrophages, human acute monocytic leukaemia cell line	Graff et al. (2012)
miR-21	↑	Lung tissue	Ezzie et al. (2012)	↓	TIMP3, RECK and PTEN	Glioma cell line	Fata et al. (2012), Gabriely et al. (2008)
	↓	EBC	Pinkerton et al. (2013)				
miR-126*	↑	Lung tissue	Ezzie et al. (2012)	↓	ADAM9	Pancreatic cancer cell line	Hamada et al. (2012)
	–	Bronchoalveolar cells	Molina-Pinelo et al. (2014)				
miR-146 (a/b-5p)	↓	Induced sputum supernatant	Van Pottelberge et al. (2011)	↓	MMP13, ADAMTS-5 and collagen II	Human articular chondrocytes and synoviocytes	Li et al. (2011)
	↓	Bronchoalveolar cells	Molina-Pinelo et al. (2014)				
	↓×	Bronchial airway epithelium	Schembri et al. (2009)				
miR-203	↓	Induced sputum supernatant	Van Pottelberge et al. (2011)	↑	MMP1	Synovial fibroblasts	Stanczyk et al. (2011)
	↓×	Induced sputum supernatant	Van Pottelberge et al. (2011)				

↑ up-regulation, ↓ down-regulation, × smokers vs. non-smokers, *RECK* reversion-inducing cysteine-rich protein with Kazal motifs, *PTEN* phosphatase and tensin homologue

pathogenesis because of the tissue/cell-specific character of miRNA expression (Van Pottelberge et al. 2011). Without evidence from further studies, it is impossible to conclude whether miR-452 deregulation in alveolar macrophages is associated with COPD pathogenesis or only with cigarette smoking per se.

As shown in Table 4, some miRNAs are up-regulated in COPD, in contrast with the general trend of the down-regulated global miRNome (Ezzie et al. 2012). Interestingly, one of the upregulated miRNAs, miR-21, has been suggested to silence three inhibitors of the MMP network—TIMP3, RECK and PTEN (Gabriely et al. 2008; Ma

et al. 2011; Meng et al. 2007). The extracellular matrix-bound TIMP3, cellular membrane-anchored RECK and intracellular phosphatase PTEN can collectively inhibit both expression and enzymatic activity of MMPs. Thus, the abundance of miR-21 in COPD lung tissue may contribute to the increased activity of MMPs in COPD due to insufficient inhibition at multiple levels (Gabriely et al. 2008; Ma et al. 2011; Meng et al. 2007).

With current progress in the understanding of post-transcriptional regulation, however, it is important to note that RNA-binding proteins can also regulate MMP mRNA expression in addition to the impact of several miRNAs (Ivanov and Anderson 2013; Ma et al. 2011). Their additive, multiple or competitive effects on the final MMP/TIMP network represent an important area for future investigation.

Current and Prospective Therapies for COPD in the Context of MMPs

The mainstay of COPD therapy is short- and long-acting bronchodilators, especially inhaled β2-adrenergic receptor agonists and inhaled anticholinergics. Their single-dose effects last from 4–8 h in case of short-acting bronchodilators (fenoterol, salbutamol, terbutaline and ipratropium) to 12–24 h in case of long-acting bronchodilators (formoterol, salmeterol and tiotropium). Persistent COPD symptoms are treated with long-acting bronchodilators that are proven to provide long-term improvements in lung function, quality of life and frequency of exacerbations in patients with COPD (Tashkin et al. 2008). To reach the maximum benefit, long-acting bronchodilators are usually prescribed in combination with short-acting bronchodilators (van Noord et al. 2000). As a potential prognostic biomarker, MMPs should be modulated by the COPD therapeutic interventions. To the best of our knowledge, the MMP/TIMP network has not yet been investigated in a long-term clinical study of COPD patients treated with bronchodilator therapy. Regarding animal studies, the short-acting bronchodilator salbutamol was ineffective in modulating MMP-9 release and cell influx in the BAL fluid from mice (Lagente et al. 2004). The long-acting bronchodilators suppressed in vitro productions of MMP-2 and MMP-9 in primary lung fibroblasts and alveolar macrophages established from non-COPD lung but this suppression was not observed in primary airway smooth muscle cells from COPD lung (Asano et al. 2008; Lambers et al. 2014; Perng et al. 2012).

In advanced COPD with frequent acute ECOPD, long-acting bronchodilators are recommended in combination with inhaled glucocorticosteroids (Vestbo et al. 2013). Release of MMPs from COPD lung and circulating cells in vitro and in vivo was shown to be resistant to corticosteroid therapy (Chaudhuri et al. 2012; Higashimoto et al. 2009; Milara et al. 2014; Vlahos et al. 2012). In agreement with these observations, a 4-week intervention study by Culpitt et al. (1999) failed to show any effect of the inhaled corticosteroid fluticasone on the sputum MMP/TIMP network in COPD. However, some response of local lung MMP/TIMP network to the inhaled corticosteroid may contribute to the reasons of inconsistent observations among the studies on COPD progression and their poor replicability discussed above (Pinto-Plata et al. 2007, 2012). In particular, Perng et al. (2009) showed decreased MMP-9 and IL-8 levels after a 3-month combined treatment of fluticasone with salmeterol that is assumed to be described in more advanced stages, compared to monotherapy with tiotropium alone that is usually taken in less advanced stage, but there were no differences when compared to tiotropium combined with fluticasone.

Roflumilast, an inhibitor of phosphodiesterase 4, is an anti-inflammatory treatment option in patients with COPD GOLD stages III–IV (C + D). It provides a sustained and significant improvement in lung function and a reduction in the frequency of acute ECOPD (Wedzicha et al. 2013). The benefit of the compound results from the reduction of the COPD inflammatory response. The particular inhibitory effect of roflumilast on the MMP network has been described in vitro (Growcott et al. 2006; Jones et al. 2005) but was not confirmed in vivo in a mouse model (Le Quement et al. 2008). In a clinical study by Grootendorst et al. (2007), short-term roflumilast therapy decreased sputum neutrophils and neutrophil elastase and, therefore, a similar effect was assumed for other products of neutrophils such as MMPs (Hoenderdos and Condliffe 2013). In line with this hypothesis, 1 year later, Milara et al. (2014) showed an active metabolite of roflumilast, roflumilast-N-oxide, strongly reduced MMP-9 release from neutrophils from COPD patients.

To date, however, neither the MMP/TIMP network nor other inflammatory mediators have been a primary subject of longitudinal prospective clinical investigations of the COPD response to roflumilast. The main reason is that no reliable biomarker, including CRP generally accepted as a clinical marker of systemic inflammation in various diseases, has yet been confirmed to be suitable for monitoring COPD inflammation and progression with adequate specificity and sensitivity (Park et al. 2013). Thus, the assessment of inflammatory mediators or MMPs/TIMPs does not seem to be as useful for the evaluation of a new therapeutic approach compared to the use of clinical indices such as lung function tests or acute ECOPD history. However, an international clinical, 16-week, placebo-controlled, parallel-group study was started in May 2014 to explore the cellular mechanisms underlying the anti-

inflammatory effects of roflumilast and identify potentially important biomarkers (Barnes et al. 2014).

Several studies in animal models have indicated that therapies primarily based on the inhibition of MMPs, e.g. MMP-9 and MMP-12, might be appropriate goals (Churg et al. 2007; Ganesan et al. 2010; Le Quement et al. 2008; Wu et al. 2012). In humans, a selective MMP-12 inhibitor (V85546, formerly AS111793) (Le Quement et al. 2008) has successfully completed phase I clinical testing and is about to be tested in a phase II programme (http://www.vernalis.com). Similar to V85546, another selective MMP-9 and MMP-12 inhibitor (AZD1236) was generally well tolerated over 6 weeks in patients with moderate-to-severe COPD. AZD1236 showed no clinical efficacy in the short-term. Regarding local and systemic inflammation, AZD1236 did not affect any pro-inflammatory biomarkers including sputum expression of MMP-8 and MMP-9 proteins and the activity of MMPs. It is important to note that these data should be interpreted with caution given the multiple analyses conducted and lack of adjustment for multiple comparisons (Dahl et al. 2012).

Conclusion and Future Directions

As documented in this review, among the members of the MMP/TIMP network, MMP-1, MMP-8, MMP-9, MMP-12 and TIMP1 have been repeatedly associated with COPD in cross-sectional studies. Only local expression of MMP-9 relates to COPD pathogenesis in case–control studies in which smoking status was closely matched. Sampling both upper and lower airways resulted in consistent observations of MMP-9 expression in sputum and BAL macrophages in vivo and in vitro (Paone et al. 2011; Russell et al. 2002). Prognostic potency of circulating MMP-9 was suggested for a specific COPD phenotype (Higashimoto et al. 2009; Omachi et al. 2011). However, other members of the MMP/TIMP network also deserve a similar prospective approach in newly planned research projects. The most robust future analyses may even include coincident measurement of several MMPs (e.g. MMP-8, MMP-9, MMP-12 and other MMPs or proteases) with their inhibitors (e.g. TIMP4) that could achieve greater reliability when measured repeatedly at several time points over the course of several years. Apart from the measurements of MMP concentrations, determination of their enzymatic activities may have clinical relevance in terms of therapeutic intervention.

To improve prediction of COPD prognosis, laboratory data obtained with the most current techniques (such as multiplex analyses) will have to be combined with clinical factors including possible changes in therapy and smoking cessation (Park et al. 2013). The effect of continuous clinical changes will be important in prospective studies that will involve following COPD patients for several years. The prognostic value of MMPs may differ among COPD patients according to the presence of emphysema and AATD and, more broadly, precise clinical phenotyping of COPD patients and clear definitions of inclusion/exclusion criteria are also essential.

Development of pharmacological therapy inhibiting MMPs is at an early stage. In addition to MMPs/TIMPs, newer biomarkers for COPD, including novel molecules exemplified here by miRNAs, have been intensively sought. As current investigations of the miRNome focus on potential mRNA targets, therefore, the longitudinal prospective investigation of miRNA in COPD patients still awaits its prime time. However, considering the relatively good stability of the miRNome and miRNAs in accessible body fluids (plasma, serum, BAL fluid and urine), it is only a matter of time before they enter the arena and, due to the intrinsic relationship of some miRNAs to MMP/TIMP regulation, will be applied in the context of current knowledge. The inclusion of miRNome data into the investigations will definitely bring new data and insights into the complex regulation of COPD inflammation by MMPs and their inhibitors and may open new avenues for identification of biomarkers for diagnosis and management of patients with this important group of conditions.

Acknowledgments This work was supported by the project CZ.1.07/2.3.00/30.0004 and IGA PU LF 2015 020.

References

Aaron SD, Vandemheen KL, Ramsay T et al (2010) Multi analyte profiling and variability of inflammatory markers in blood and induced sputum in patients with stable COPD. Respir Res 11:41

Ali S, Banerjee S, Logna F et al (2012) Inactivation of Ink4a/Arf leads to deregulated expression of miRNAs in K-Ras transgenic mouse model of pancreatic cancer. J Cell Physiol 227:3373–3380

Asano K, Shikama Y, Shibuya Y et al (2008) Suppressive activity of tiotropium bromide on matrix metalloproteinase production from lung fibroblasts in vitro. Int J Chron Obstruct Pulmon Dis 3:781–789

Bafadhel M, McKenna S, Terry S et al (2011) Acute exacerbations of chronic obstructive pulmonary disease: identification of biologic clusters and their biomarkers. Am J Respir Crit Care Med 184:662–671

Baines KJ, Simpson JL, Gibson PG (2011) Innate immune responses are increased in chronic obstructive pulmonary disease. PLoS ONE 6:e18426

Barnes NC, Saetta M, Rabe KF (2014) Implementing lessons learned from previous bronchial biopsy trials in a new randomized controlled COPD biopsy trial with roflumilast. BMC Pulm Med 14:9

Beasley V, Joshi PV, Singanayagam A et al (2012) Lung microbiology and exacerbations in COPD. Int J Chron Obstr Pulmon Dis 7:555–569

Bolton CE, Stone MD, Edwards PH et al (2009) Circulating matrix metalloproteinase-9 and osteoporosis in patients with chronic obstructive pulmonary disease. Chron Respir Dis 6:81–87

Bonnema DD, Webb CS, Pennington WR et al (2007) Effects of age on plasma matrix metalloproteinases (MMPs) and tissue inhibitor of metalloproteinases (TIMPs). J Card Fail 13:530–540

Boosani CS, Agrawal DK (2013) PTEN modulators: a patent review. Expert Opin Ther Pat 23:569–580

Brajer B, Batura-Gabryel H, Nowicka A et al (2008) Concentration of matrix metalloproteinase-9 in serum of patients with chronic obstructive pulmonary disease and a degree of airway obstruction and disease progression. J Physiol Pharmacol 59(Suppl 6):145–152

Brusselle GG, Joos GF, Bracke KR (2011) New insights into the immunology of chronic obstructive pulmonary disease Lancet 378:1015–1026

Burgel PR, Bergeron A, de Blic J et al (2013) Small airways diseases, excluding asthma and COPD: an overview. Eur Respir Rev 22:131–147

Cataldo D, Munaut C, Noel A et al (2000) MMP-2- and MMP-9-linked gelatinolytic activity in the sputum from patients with asthma and chronic obstructive pulmonary disease. Int Arch Allergy Immunol 123:259–267

Cataldo D, Munaut C, Noel A et al (2001) Matrix metalloproteinases and TIMP-1 production by peripheral blood granulocytes from COPD patients and asthmatics. Allergy 56:145–151

Chaudhuri R, McSharry C, Brady J et al (2012) Sputum matrix metalloproteinase-12 in patients with chronic obstructive pulmonary disease and asthma: relationship to disease severity. J Allergy Clin Immunol 129(655–663):e658

Chaudhuri R, McSharry C, Spears M et al (2013) Sputum matrix metalloproteinase-9 is associated with the degree of emphysema on computed tomography in COPD. Translational Respir Med 1:11

Chelladurai P, Seeger W, Pullamsetti SS (2012) Matrix metalloproteinases and their inhibitors in pulmonary hypertension. Eur Respir J 40:766–782

Churg A, Wang R, Wang X et al (2007) Effect of an MMP-9/MMP-12 inhibitor on smoke-induced emphysema and airway remodelling in guinea pigs. Thorax 62:706–713

Churg A, Zhou S, Wright JL (2012) Series "matrix metalloproteinases in lung health and disease": matrix metalloproteinases in COPD. Eur Respir J 39:197–209

Culpitt SV, Maziak W, Loukidis S et al (1999) Effect of high dose inhaled steroid on cells, cytokines, and proteases in induced sputum in chronic obstructive pulmonary disease. Am J Respir Crit Care Med 160:1635–1639

Culpitt SV, Rogers DF, Traves SL et al (2005) Sputum matrix metalloproteases: comparison between chronic obstructive pulmonary disease and asthma. Respir Med 99:703–710

Dahl R, Titlestad I, Lindqvist A et al (2012) Effects of an oral MMP-9 and -12 inhibitor, AZD1236, on biomarkers in moderate/severe COPD: a randomised controlled trial. Pulm Pharmacol Ther 25:169–177

D'Armiento JM, Goldklang MP, Hardigan AA et al (2013) Increased matrix metalloproteinase (MMPs) levels do not predict disease severity or progression in emphysema. PLoS ONE 8:e56352

Davey A, McAuley DF, O'Kane CM (2011) Matrix metalloproteinases in acute lung injury: mediators of injury and drivers of repair. Eur Respir J 38:959–970

Demedts IK, Morel-Montero A, Lebecque S et al (2006) Elevated MMP-12 protein levels in induced sputum from patients with COPD. Thorax 61:196–201

Deshmukh HS, McLachlan A, Atkinson JJ et al (2009) Matrix metalloproteinase-14 mediates a phenotypic shift in the airways to increase mucin production. Am J Respir Crit Care Med 180:834–845

Devenport NA, Reynolds JC, Parkash V et al (2011) Determination of free desmosine and isodesmosine as urinary biomarkers of lung disorder using ultra performance liquid chromatography-ion mobility-mass spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci 879:3797–3801

Dickens JA, Miller BE, Edwards LD et al (2011) COPD association and repeatability of blood biomarkers in the ECLIPSE cohort. Respir Res 12:146

Ezzie ME, Crawford M, Cho JH et al (2012) Gene expression networks in COPD: microRNA and mRNA regulation. Thorax 67:122–131

Fata JE, Debnath S, Jenkins EC Jr et al (2012) Nongenomic mechanisms of PTEN regulation. Int J Cell Biol 2012:379685

Finlay GA, O'Driscoll LR, Russell KJ et al (1997) Matrix metalloproteinase expression and production by alveolar macrophages in emphysema. Am J Respir Crit Care Med 156:240–247

Gabriely G, Wurdinger T, Kesari S et al (2008) MicroRNA 21 promotes glioma invasion by targeting matrix metalloproteinase regulators. Mol Cell Biol 28:5369–5380

Ganesan S, Faris AN, Comstock AT et al (2010) Quercetin prevents progression of disease in elastase/LPS-exposed mice by negatively regulating MMP expression. Respir Res 11:131

Gao Z, Ye J (2008) Inhibition of transcriptional activity of c-JUN by SIRT1. Biochem Biophys Res Commun 376:793–796

Gao P, Zhang J, He X et al (2013) Sputum inflammatory cell-based classification of patients with acute exacerbation of chronic obstructive pulmonary disease. PLoS ONE 8:e57678

Garofalo M, Di Leva G, Romano G et al (2009) miR-221&222 regulate TRAIL resistance and enhance tumorigenicity through PTEN and TIMP3 downregulation. Cancer Cell 16:498–509

Global Strategy for the Diagnosis, Management and Prevention of COPD, Global Initiative for Chronic Obstructive Lung Disease (GOLD) (2015) http://www.goldcopd.org/

Gosselink JV, Hayashi S, Elliott WM et al (2010) Differential expression of tissue repair genes in the pathogenesis of chronic obstructive pulmonary disease. Am J Respir Crit Care Med 181:1329–1335

Graff JW, Powers LS, Dickson AM et al (2012) Cigarette smoking decreases global microRNA expression in human alveolar macrophages. PLoS ONE 7:e44066

Grootendorst DC, Gauw SA, Verhoosel RM et al (2007) Reduction in sputum neutrophil and eosinophil numbers by the PDE4 inhibitor roflumilast in patients with COPD. Thorax 62:1081–1087

Growcott EJ, Spink KG, Ren X et al (2006) Phosphodiesterase type 4 expression and anti-proliferative effects in human pulmonary artery smooth muscle cells. Respir Res 7:9

Hamada S, Satoh K, Fujibuchi W et al (2012) MiR-126 acts as a tumor suppressor in pancreatic cancer cells via the regulation of ADAM9. Mol Cancer Res 10:3–10

Harju T, Kinnula VL, Paakko P et al (2010) Variability in the precursor proteins of collagen I and III in different stages of COPD. Respir Res 11:165

Higashimoto Y, Yamagata Y, Iwata T et al (2005) Increased serum concentrations of tissue inhibitor of metalloproteinase-1 in COPD patients. Eur Respir J 25:885–890

Higashimoto Y, Iwata T, Okada MY et al (2009) Serum biomarkers as predictors of lung function decline in chronic obstructive pulmonary disease. Respir Med 103:1231–1238

Hoenderdos K, Condliffe A (2013) The neutrophil in chronic obstructive pulmonary disease. Am J Respir Cell Mol Biol 48:531–539

Hurst JR, Donaldson GC, Perera WR et al (2006) Use of plasma biomarkers at exacerbation of chronic obstructive pulmonary disease. Am J Respir Crit Care Med 174:867–874

Ilumets H, Rytilä P, Demedts I et al (2007) Matrix metalloproteinases -8, -9 and -12 in smokers and patients with stage 0 COPD. Int J Chron Obstruct Pulm Dis 2:369–379

Ilumets H, Rytila PH, Sovijarvi AR et al (2008) Transient elevation of neutrophil proteinases in induced sputum during COPD exacerbation. Scand J Clin Lab Invest 68:618–623

Ilumets H, Mazur W, Toljamo T et al (2011) Ageing and smoking contribute to plasma surfactant proteins and protease imbalance with correlations to airway obstruction. BMC Pulm Med 11:19

Imai K, Dalal SS, Chen ES et al (2001) Human collagenase (matrix metalloproteinase-1) expression in the lungs of patients with emphysema. Am J Respir Crit Care Med 163:786–791

Ishii T, Abboud RT, Wallace AM et al (2014) Alveolar macrophage proteinase/antiproteinase expression in lung function and emphysema. Eur Respir J 43:82–91

Ivanov P, Anderson P (2013) Post-transcriptional regulatory networks in immunity. Immunol Rev 253:253–272

Jenkins CR, Celli B, Anderson JA et al (2012) Seasonality and determinants of moderate and severe COPD exacerbations in the TORCH study. Eur Respir J 39:38–45

Jones NA, Boswell-Smith V, Lever R et al (2005) The effect of selective phosphodiesterase isoenzyme inhibition on neutrophil function in vitro. Pulm Pharmacol Ther 18:93–101

Kanazawa H (2007) Role of vascular endothelial growth factor in the pathogenesis of chronic obstructive pulmonary disease. Med Sci Monit 13:RA189–RA195

Ko FW, Diba C, Roth M et al (2005) A comparison of airway and serum matrix metalloproteinase-9 activity among normal subjects, asthmatic patients, and patients with asthmatic mucus hypersecretion. Chest 127:1919–1927

Kwiatkowska S, Noweta K, Zieba M et al (2012) Enhanced exhalation of matrix metalloproteinase-9 and tissue inhibitor of metalloproteinase-1 in patients with COPD exacerbation: a prospective study. Respiration 84:231–241

Lagente V, Naline E, Guenon I et al (2004) A nitric oxide-releasing salbutamol elicits potent relaxant and anti-inflammatory activities. J Pharmacol Exp Ther 310:367–375

Lambers C, Qi Y, Eleni P et al (2014) Extracellular matrix composition is modified by beta(2)-agonists through cAMP in COPD. Biochem Pharmacol 91:400–408

Le Quement C, Guenon I, Gillon JY et al (2008) The selective MMP-12 inhibitor, AS111793 reduces airway inflammation in mice exposed to cigarette smoke. Br J Pharmacol 154:1206–1215

Lee EJ, In KH, Kim JH et al (2009) Proteomic analysis in lung tissue of smokers and COPD patients. Chest 135:344–352

Leeming DJ, Sand JM, Nielsen MJ et al (2012) Serological investigation of the collagen degradation profile of patients with chronic obstructive pulmonary disease or idiopathic pulmonary fibrosis. Biomark Insights 7:119–126

Leidinger P, Keller A, Borries A et al (2011) Specific peripheral miRNA profiles for distinguishing lung cancer from COPD. Lung Cancer 74:41–47

Li X, Gibson G, Kim JS et al (2011) MicroRNA-146a is linked to pain-related pathophysiology of osteoarthritis. Gene 480:34–41

Liu X, Yu J, Jiang L et al (2009) MicroRNA-222 regulates cell invasion by targeting matrix metalloproteinase 1 (MMP1) and manganese superoxide dismutase 2 (SOD2) in tongue squamous cell carcinoma cell lines. Cancer Genomics Proteomics 6:131–139

Loffek S, Schilling O, Franzke CW (2011) Series "matrix metalloproteinases in lung health and disease": biological role of matrix metalloproteinases: a critical balance. Eur Respir J 38:191–208

Louhelainen N, Stark H, Mazur W et al (2010) Elevation of sputum matrix metalloproteinase-9 persists up to 6 months after smoking cessation: a research study. BMC Pulm Med 10:13

Lowrey GE, Henderson N, Blakey JD et al (2008) MMP-9 protein level does not reflect overall MMP activity in the airways of patients with COPD. Respir Med 102:845–851

Loza MJ, Watt R, Baribaud F et al (2012) Systemic inflammatory profile and response to anti-tumor necrosis factor therapy in chronic obstructive pulmonary disease. Respir Res 13:12

Lu Y et al (2011) Anti-microRNA-222 (anti-miR-222) and -181B suppress growth of tamoxifen-resistant xenografts in mouse by targeting TIMP3 protein and modulating mitogenic signal. J Biol Chem 286:42292–42302

Ma S, Lin YY, Turino GM (2007) Measurements of desmosine and isodesmosine by mass spectrometry in COPD. Chest 131:1363–1371

Ma X, Becker Buscaglia LE, Barker JR et al (2011) MicroRNAs in NF-kappaB signaling. J Mol Cell Biol 3:159–166

Maclay JD et al (2012) Systemic elastin degradation in chronic obstructive pulmonary disease. Thorax 67:606–612

Meng F, Henson R, Wehbe-Janek H et al (2007) MicroRNA-21 regulates expression of the PTEN tumor suppressor gene in human hepatocellular cancer. Gastroenterology 133:647–658

Mercer PF, Shute JK, Bhowmik A et al (2005) MMP-9, TIMP-1 and inflammatory cells in sputum from COPD patients during exacerbation. Respir Res 6:151

Milara J, Lluch J, Almudever P et al (2014) Roflumilast N-oxide reverses corticosteroid resistance in neutrophils from patients with chronic obstructive pulmonary disease. J Allergy Clin Immunol 134:314–322

Millares L, Marin A, Garcia-Aymerich J et al (2012) Specific IgA and metalloproteinase activity in bronchial secretions from stable chronic obstructive pulmonary disease patients colonized by Haemophilus influenzae. Respir Res 13:113

Molina-Pinelo S, Pastor MD, Suarez R et al (2014) MicroRNA clusters: dysregulation in lung adenocarcinoma and COPD. Eur Respir J 43:1740–1749

Mu W, Zhang W (2012) Bioinformatic resources of microRNA sequences, gene targets, and genetic variation. Front Genet 3:31

Muller KC, Welker L, Paasch K et al (2006) Lung fibroblasts from patients with emphysema show markers of senescence in vitro. Respir Res 7:32

Murakami Y, Toyoda H, Tanaka M et al (2011) The progression of liver fibrosis is related with overexpression of the miR-199 and 200 families. PLoS ONE 6:e16081

Nakamaru Y, Vuppusetty C, Wada H et al (2009) A protein deacetylase SIRT1 is a negative regulator of metalloproteinase-9. FASEB J 23:2810–2819

Navratilova Z, Zatloukal J, Kriegova E et al (2012) Simultaneous up-regulation of matrix metalloproteinases 1, 2, 3, 7, 8, 9 and tissue inhibitors of metalloproteinases 1, 4 in serum of patients with chronic obstructive pulmonary disease. Respirology 17:1006–1012

Noguera A, Gomez C, Faner R et al (2012) An investigation of the resolution of inflammation (catabasis) in COPD. Respir Res 13:101

Olafsdottir IS, Janson C, Lind L et al (2010) Serum levels of matrix metalloproteinase-9, tissue inhibitors of metalloproteinase-1 and their ratio are associated with impaired lung function in the elderly: a population-based study. Respirology 15:530–535

Omachi TA, Eisner MD, Rames A et al (2011) Matrix metalloproteinase-9 predicts pulmonary status declines in alpha1-antitrypsin deficiency. Respir Res 12:35

Paone G, Conti V, Vestri A et al (2011) Analysis of sputum markers in the evaluation of lung inflammation and functional impairment in symptomatic smokers and COPD patients. Dis Markers 31:91–100

Park HY, Churg A, Wright JL et al (2013) Club cell protein 16 and disease progression in chronic obstructive pulmonary disease (COPD). Am J Respir Crit Care Med 188:1413–1419

Perng DW, Tao CW, Su KC et al (2009) Anti-inflammatory effects of salmeterol/fluticasone, tiotropium/fluticasone or tiotropium in COPD. Eur Respir J 33:778–784

Perng DW, Su KC, Chou KT et al (2012) Long-acting beta2 agonists and corticosteroids restore the reduction of histone deacetylase activity and inhibit H_2O_2-induced mediator release from alveolar macrophages. Pulm Pharmacol Ther 25:312–318

Philibert RA, Sears RA, Powers LS et al (2012) Coordinated DNA methylation and gene expression changes in smoker alveolar macrophages: specific effects on VEGF receptor 1 expression. J Leukoc Biol 92:621–631

Pinkerton M, Chinchilli V, Banta E et al (2013) Differential expression of microRNAs in exhaled breath condensates of patients with asthma, patients with chronic obstructive pulmonary disease, and healthy adults. J Allergy Clin Immunol 132:217–219

Pinto-Plata V, Toso J, Lee K et al (2007) Profiling serum biomarkers in patients with COPD: associations with clinical parameters. Thorax 62:595–601

Pinto-Plata V, Casanova C, Müllerova H et al (2012) Inflammatory and repair serum biomarker pattern. Association to clinical outcomes in COPD. Respir Res 13:71

Ropcke S, Holz O, Lauer G et al (2012) Repeatability of and relationship between potential COPD biomarkers in bronchoalveolar lavage, bronchial biopsies, serum, and induced sputum. PLoS ONE 7:e46207

Russell RE, Culpitt SV, DeMatos C et al (2002) Release and activity of matrix metalloproteinase-9 and tissue inhibitor of metalloproteinase-1 by alveolar macrophages from patients with chronic obstructive pulmonary disease. Am J Respir Cell Mol Biology 26:602–609

Schembri F, Sridhar S, Perdomo C et al (2009) MicroRNAs as modulators of smoking-induced gene expression changes in human airway epithelium. Proc Natl Acad Sci USA 106:2319–2324

Segura-Valdez L, Pardo A, Gaxiola M et al (2000) Upregulation of gelatinases A and B, collagenases 1 and 2, and increased parenchymal cell death in COPD. Chest 117:684–694

Sethi S, Maloney J, Grove L et al (2006) Airway inflammation and bronchial bacterial colonization in chronic obstructive pulmonary disease. Am J Respir Crit Care Med 173:991–998

Shaker SB, von Wachenfeldt KA, Larsson S et al (2008) Identification of patients with chronic obstructive pulmonary disease (COPD) by measurement of plasma biomarkers. Clin Respir J 2:17–25

Shaykhiev R, Otaki F, Bonsu P et al (2011) Cigarette smoking reprograms apical junctional complex molecular architecture in the human airway epithelium in vivo. Cell Mol Life Sci 68:877–892

Simpson JL, McDonald VM, Baines KJ et al (2013) Influence of age, past smoking, and disease severity on TLR2, neutrophilic inflammation, and MMP-9 levels in COPD. Mediators Inflamm 2013:462934

Sing T, Jinnin M, Yamane K et al (2012) microRNA-92a expression in the sera and dermal fibroblasts increases in patients with scleroderma. Rheumatology 51:1550–1556

Skjot-Arkil H, Clausen RE, Nguyen QH et al (2012) Measurement of MMP-9 and -12 degraded elastin (ELM) provides unique information on lung tissue degradation. BMC Pulm Med 12:34

Snitker S, Xie K, Ryan KA et al (2013) Correlation of circulating MMP-9 with white blood cell count in humans: effect of smoking. PLoS ONE 8:e66277

Stanczyk J, Ospelt C, Karouzakis E et al (2011) Altered expression of microRNA-203 in rheumatoid arthritis synovial fibroblasts and its role in fibroblast activation. Arthritis Rheum 63:373–381

Sturgeon C, Hill R, Hortin GL et al (2010) Taking a new biomarker into routine use—a perspective from the routine clinical biochemistry laboratory. Proteomics Clin Appl 4:892–903

Takahashi C, Sheng Z, Horan TP et al (1998) Regulation of matrix metalloproteinase-9 and inhibition of tumor invasion by the membrane-anchored glycoprotein RECK. Proc Natl Acad Sci USA 95:13221–13226

Tashkin DP, Celli B, Senn S et al (2008) A 4-year trial of tiotropium in chronic obstructive pulmonary disease. N Engl J Med 359:1543–1554

Thomsen M, Ingebrigtsen TS, Marott JL et al (2013) Inflammatory biomarkers and exacerbations in chronic obstructive pulmonary disease. JAMA 309:2353–2361

van Noord JA, de Munck DR, Bantje TAAM et al (2000) Long-term treatment of chronic obstructive pulmonary disease with salmeterol and the additive effect of ipratropium. Eur Respir J 15:878–885

Van Pottelberge GR, Mestdagh P, Bracke KR et al (2011) MicroRNA expression in induced sputum of smokers and patients with chronic obstructive pulmonary disease. Am J Respir Crit Care Med 183:898–906

Vergoulis T, Vlachos IS, Alexiou P et al (2012) TarBase 6.0: capturing the exponential growth of miRNA targets with experimental support. Nucleic Acids Res 40:D222–D229

Vernooy JH, Lindeman JH, Jacobs JA et al (2004) Increased activity of matrix metalloproteinase-8 and matrix metalloproteinase-9 in induced sputum from patients with COPD. Chest 126:1802–1810

Vestbo J, Hurd SS, Rodriguez-Roisin R (2012) The 2011 revision of the global strategy for the diagnosis, management and prevention of COPD (GOLD)—why and what? Clin Respir J 6:208–214

Vestbo J, Hurd SS, Agustí AG et al (2013) Global strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease: GOLD executive summary. Am J Respir Crit Care Med 187:347–365

Vignola AM, Riccobono L, Mirabella A et al (1998) Sputum metalloproteinase-9/tissue inhibitor of metalloproteinase-1 ratio correlates with airflow obstruction in asthma and chronic bronchitis. Am J Respir Crit Care Med 158:1945–1950

Vlahos R, Wark PA, Anderson GP et al (2012) Glucocorticosteroids differentially regulate MMP-9 and neutrophil elastase in COPD. PLoS ONE 7:e33277

Vucic EA, Chari R, Thu KL et al (2014) DNA methylation is globally disrupted and associated with expression changes in chronic obstructive pulmonary disease small airways. Am J Respir Cell Mol Biol 50:912–922

Wallace AM, Sandford AJ, English JC et al (2008) Matrix metalloproteinase expression by human alveolar macrophages in relation to emphysema. COPD 5:13–23

Wang IM, Stepaniants S, Boie Y et al (2008) Gene expression profiling in patients with chronic obstructive pulmonary disease and lung cancer. Am J Respir Crit Care Med 177:402–411

Wedzicha JA, Rabe KF, Martinez FJ et al (2013) Efficacy of roflumilast in the COPD frequent exacerbator phenotype. Chest 143:1302–1311

Wouters EF, Reynaert NL, Dentener MA et al (2009) Systemic and local inflammation in asthma and chronic obstructive pulmonary disease: is there a connection? Proc Am Thorac Soc 6:638–647

Wu Y, Li J, Wu J et al (2012) Discovery of potent and selective matrix metalloprotease 12 inhibitors for the potential treatment of chronic obstructive pulmonary disease (COPD). Bioorg Med Chem Lett 22:138–143

Xu N, Zhang L, Meisgen F et al (2012) MicroRNA-125b down-regulates matrix metallopeptidase 13 and inhibits cutaneous squamous cell carcinoma cell proliferation, migration, and invasion. J Biol Chem 287:29899–29908

Yao H, Hwang JW, Sundar IK et al (2013) SIRT1 redresses the imbalance of tissue inhibitor of matrix metalloproteinase-1 and matrix metalloproteinase-9 in the development of mouse emphysema and human COPD. Am J Physiol Lung Cell Mol Physiol 305:L615–L624