Review Article

Pleural mesothelial cells in pleural and lung diseases

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Abstract: During development, the mesoderm maintains a complex relationship with the developing endoderm giving rise to the mature lung. Pleural mesothelial cells (PMCs) derived from the mesoderm play a key role during the development of the lung. The pleural mesothelium differentiates to give rise to the endothelium and smooth muscle cells via epithelial-to-mesenchymal transition (EMT). An aberrant recapitulation of such developmental pathways can play an important role in the pathogenesis of disease processes such as idiopathic pulmonary fibrosis (IPF). The PMC is the central component of the immune responses of the pleura. When exposed to noxious stimuli, it demonstrates innate immune responses such as Toll-like receptor (TLR) recognition of pathogen associated molecular patterns as well as causes the release of several cytokines to activate adaptive immune responses. Development of pleural effusions occurs due to an imbalance in the dynamic interaction between junctional proteins, n-cadherin and β-catenin, and phosphorylation of adherens junctions between PMCs, which is caused in part by vascular endothelial growth factor (VEGF) released by PMCs. PMCs play an important role in defense mechanisms against bacterial and mycobacterial pleural infections, and in pathogenesis of malignant pleural effusion, asbestos related pleural disease and malignant pleural mesothelioma. PMCs also play a key role in the resolution of inflammation, which can occur with or without fibrosis. Fibrosis occurs as a result of disordered fibrin turnover and due to the effects of cytokines such as transforming growth factor-β, platelet-derived growth factor (PDGF), and basic fibroblast growth factor; which are released by PMCs. Recent studies have demonstrated a role for PMCs in the pathogenesis of IPF suggesting their potential as a cellular biomarker of disease activity and as a possible therapeutic target. Pleural-based therapies targeting PMCs for treatment of IPF and other lung diseases need further exploration.

Keywords: Pleural mesothelium1; pleural mesothelial cells (PMCs); idiopathic pulmonary fibrosis (IPF); Wilms tumor-1 (WT1); epithelial-mesenchymal transition (EMT)

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Introduction

The pleural mesothelium, derived from the embryonic mesoderm, is a monolayer of mesothelial cells that blanket the chest wall and lungs on the parietal and visceral surfaces, respectively. The normal mesothelial cell layer appears smooth, glistening, and semi-transparent. On light microscopy, the appearance of the mesothelial cells may vary from a row of flattened and elongated ovoid nuclei widely separated by cytoplasm to cuboidal or columnar cells with round basal nuclei and a cuboidal and fuzzy luminal surface (1). These pavement-like cells are similar in cytologic characteristics to mesothelial cells that line other body cavities such as the peritoneum (2).

The pleural mesothelial cell (PMC) is the most common cell in the pleural space and is the primary cell that initiates responses to noxious stimuli (3). PMCs are metabolically active cells that maintain a dynamic state of homeostasis in the pleural space. As a response to injury, mesothelial cells respond by proliferation and chemotaxis to cover areas of denuded extracellular matrix. This response is mediated by an autocrine signaling due to the production of chemokines.

Juxtacrine and paracrine communications between cells allow for a rapid response during inflammation (4). The cytoplasm of PMCs contains abundant organelles and glycogen granules. PMCs are phagocytic and produce several cytokines and adhesion molecules (5). Mesothelial cells have microvilli and multiple intercellular adherens junctions as well as focal adhesions that anchor the mesothelial cell onto the extracellular membrane via integrins. The size and shape, as well as the number of microvilli and the amount of organelles in a PMC may reflect its functionality.

Pleural mesothelial cells in development and disease

Interactions between the developing endoderm and mesoderm

The complex interplay of signaling pathways between the developing endoderm and mesoderm is essential for development (6). The lung mesoderm plays a key role in regulating the morphogenesis of the lung during all stages of the development of the anterior foregut endoderm (7). It continuously interacts with the lung endoderm to generate various cell lineages within the lung (8) serving as an important source of signaling molecules such as Fibroblast growth factor 10 (Fgf10) and Wnt2 (9-12) that are essential for processes like patterning of early endoderm progenitors, epithelial proliferation, and differentiation. Additionally, several mesodermal derived cells, including airway smooth muscle, vascular smooth muscle, endothelial and mesothelial cells, pericytes, alveolar fibroblasts, and lipofibroblasts are present in the mature lung (8).

The specification of the respiratory system in the anterior foregut endoderm during development depends on Wnt/β-catenin signaling specifying Nkx2.1+ respiratory endoderm progenitors (8). Active bone morphogenetic protein (Bmp) signaling is necessary to repress the transcription factor Sox2 to allow the expression of Nkx2.1. Interestingly, loss of Bmp signaling leads to tracheal agenesis with retention of the branching region of the lungs (13). Branching morphogenesis relies upon active signaling between the developing mesoderm and endoderm and the loss of Fgf10 signaling to Fgfr2 in the developing endoderm can lead to disruption of branching (11,14). Fgf10 expression is in turn regulated by a complex interplay of signaling molecules such as Bone morphogenetic protein 4 (Bmp4) and sonic hedgehog (Shh) (9,10,15).

Role of PMCs during development

PMCs are mesenchymal in origin but exhibit several characteristics which are typical of epithelial cells, such as a polygonal cell shape, expression of surface microvilli, epithelial cytokeratins and tight junctions (16). A process called epithelial-to-mesenchymal transition (EMT) allows for the differentiation of mesothelium to give rise to the endothelium and vascular smooth muscle cells of the vascular system, heart, liver and gut during development (17-19). Lineage labeling studies in the developing heart show that the surface epicardial mesothelium undergoes EMT and migrates into the myocardium where it differentiates into various cell types, including endothelium, smooth muscle cells, and cardiomyocytes (20-23). Moreover, it has also been shown that the serosal mesothelium of the gut contributes the majority of vascular smooth muscle cells (24,25). The hepatocyte growth factor (HGF) is a well-known cytokine produced by cells of mesenchymal origin and plays an important role in EMT during organogenesis and in regulation of lung morphogenesis (26,27).

Wilms tumor-1 (Wt1), a zinc finger transcription factor, discovered as a tumor suppressor gene in Wt of the kidney (28), is expressed in certain mesoderm-derived tissues including the pleura (29). Wt1 regulates many functional properties of the developing mesothelium (30,31). Wt1 can function either as a tumor suppressor (32) or as an oncogene (33-35) and has the potential to induce EMT (36-38). It confers oncogenic properties in cells of hemopoietic origin and regulates transforming growth factor-β1 (TGF-β1) in the kidney, demonstrating its tissue specific responses (39). PMCs express the Wt1 gene, encoding for a 49-52 kDa protein with an N-terminal domain that is involved in protein-RNA interactions critical for its transcriptional regulatory function (40). In lineage labeling studies, using Wt1 as a marker, PMCs were found to track into the lung parenchyma and undergo mesothelial-mesenchymal transition (MMT) to form smooth muscle cells of the vascular wall, as well as other cells of the lung mesenchyme during development (7,20,41). Another lineage tracing study in the mouse embryo showed PMCs readily migrate into the lung parenchyma and express α-smooth muscle actin (α-SMA) (42). A study employing $Wt1^{CreERT2/+}$ mice visualized Wt1+ mesothelial cell entry into the lung by live imaging, and by lineage tagging identified their progenies in subpopulations of bronchial smooth muscle cells, vascular smooth muscle cells and desmin + fibroblasts (43). These studies establish the quintessential role of the

mesothelium during development and organogenesis and suggest the possibility that re-activation of such developmental pathways may modulate lung injury-repair and play a role in the pathogenesis of disease processes in the post-natal period.

Pleural mesothelial cells are pluripotent

Although limited, there is evidence suggesting the existence of a population of progenitor-like mesothelial cells, with the capacity to differentiate into cells of different phenotypes (44). It has been demonstrated that the embryonic and adult mesothelium represents a common lineage to trunk fibroblasts, smooth muscle cells and vasculature (45). In one study, primary rat and human mesothelial cells were maintained in osteogenic or adipogenic media, and changes in mRNA expression of these cells suggested that these cells could differentiate into osteoblast- and adipocyte-like cells via EMT (46). The transduction of the rat peritoneum and pleura with an adenovirus expressing TGF-β1 causes mesothelial cells to undergo EMT with subsequent fibrotic changes (47,48). In response to TGF-β1 and platelet derived growth factor (PDGF), the mesothelial cells retain the ability to produce mesenchyme, including smooth muscle cells (25,49) and have been shown to adopt a myofibroblast phenotype *in vitro* (50). PMCs respond with haptotactic migration to a gradient of TGF-β1, which is dependent on smad-2 signaling, suggesting that PMCs may be a possible source of myofibroblasts in idiopathic pulmonary fibrosis (IPF) (51). Another study demonstrated TGF-β1 treated PMCs to traffic into the lung and differentiate into myofibroblasts (52). Taken together these results suggest a role for PMCs in the pathogenesis of IPF.

Pleural mesothelial cell defense mechanisms

PMC is a central component of the pathophysiologic processes affecting the pleural space and is essential in maintaining its normal homeostasis (4). There exists a harmonious cross talk between PMCs and immune cells of adaptive immunity. Upon pleural infection, the PMCs initiate pro-inflammatory responses by recruiting and activating immune cells, which in turn modulate mesothelial cell responses (53).

Innate immunity

The innate immune response of the pleura is ignited within the first few hours following an insult to the pleural space (54). This response is primarily driven by the PMCs that recognize the offending agent and initiate the inflammatory cascade, which differs according to the invading agent.

Glycoconjugates, which consist of PMC-associated sialomucins, cover the free surface of the mesothelium (55). These mesothelial cell-associated sialomucins are strong anionic sites that coat the pleural surface with a negative charge and repulse abnormal cells, organisms, and particles. These glycoproteins also provide a second level of mechanical repulsion to invading cells, microbes, etc. (56,57). In addition, mesothelial cells produce fibronectin, a large glycoprotein that prevents adherence of organisms such as *Pseudomonas aeruginosa* (55).

Mesothelial cells release various mediators of inflammation such as PDGF, interleukin-8 (IL-8), monocyte chemotactic peptide (MCP-1), collagen, antioxidant enzymes and the plasminogen activation inhibitor (PAI) (58). Activation of proteinase-activated receptor-2 (PAR-2) present on PMCs has been shown to potently induce the release of inflammatory cytokines such as macrophage inflammatory protein (MIP)-2 and tumor necrosis factor (TNF)-β and cause neutrophil recruitment into the pleural cavity (59).

Another innate response of the PMCs is the release of reactive oxygen species and the nitric oxide (NO) radical. PMCs produce large quantities of NO radicals in response to the stimulation by cytokines, lipopolysaccharide (LPS), and other signaling molecules (3,60). Inducible NO synthase may contribute to the control of infections in the pleural space and may be involved in pleural inflammation from other insults (55).

Infectious pathogens express pathogen-associated molecular patterns (PAMPs) that are composed of proteins, carbohydrates, lipids, or nucleic acids and may be intracellular or surface bound (61). PAMPs include LPS, bacterial lipoproteins, lipoteichoic acids of gram-positive bacteria, bacterial cell wall peptidoglycans (PGNs), and fungal and mycobacterial cell wall components (62). The mesothelial cells recognize PAMPs and initiate multi-level defense mechanisms (63). Some of the pattern recognition receptors including CD14, integrins, the mannose receptor, and the Toll-like receptors (TLRs) (64) bind to PAMPs to identify the pathogen and initiate downstream signaling with production of various peptides with antimicrobial activity, chemokines, and cytokines such as TNF-α, IL-1, IL-6, and IL-8 (62). Murine primary PMCs constitutively express TLR-1 through TLR-9 and activation with staphylococcal PGN, which is a gram-positive bacterial cell

wall component and a TLR-2 agonist, results in significant increase in TLR-2 and the antimicrobial peptide beta-defensin-2 (mBD-2) expression (65).

Acquired immunity

Acquired immunity involves the T- and B-cell lymphocytes and the expression of distinct antigenic receptors (66,67). PMCs release chemokines such as IL-1, IL-6 and interferons (IFNs), which co-stimulate T cells, and contribute to the cytokine networks that allow for undifferentiated T lymphocytes to become T-helper (Th)-1 or Th2-type cells that subsequently direct different inflammatory responses in the pleural space (3).

Defensins are small cationic peptides with antimicrobial function. In addition to innate immune responses, as noted above, human β-defensin-2 also promotes adaptive immune responses by recruiting dendritic cells and T lymphocytes and attracting neutrophils to sites of microbial invasion (68,69). Pleural fluids from patients with empyema contain elevated levels of human β-defensin-2 (70). PMCs have also contribute to kallikrein-kinin system (KKS)-mediated inflammation in pleural disease via a heat shock protein 90 (HSP90)-dependent mechanism (71).

Pleural permeability and formation of pleural effusion

PMCs are linked together by adherens junctions. Malignant cells, bacteria, or cytokine mediated activation of the pleural mesothelial monolayer results in altered shape and gap formation, leakage of protein and fluids, and movement of phagocytic cells into the pleural space, causing a breach in the integrity of the pleura.

Cadherins and catenins are transmembrane adherens junction proteins that allow for a change in permeability via the contraction of the intracellular actin cytoskeletal filaments and gap formation between mesothelial cells (72). Neural cadherin (n-cadherin) on PMCs loses tyrosine phosphorylation and combines with plakoglobin and actin in tightly confluent cells when adherens junctions are stabilized (73). However, n-cadherin is heavily phosphorylated in tyrosine and there is decreased expression of β-catenin in weakened junctions (74). The opening up of adherens junctions is reversible, functioning as a "zipper", with mesothelial cells returning to their normal shape with closure of junctions within 15 min after stimulation *in vitro* (75).

Vascular endothelial growth factor (VEGF), a 35- to 45-kDa dimeric polypeptide, is a permeability and angiogenic factor mediating neovascularization (76). Its expression is upregulated in activated PMCs (77) and it is produced in large quantities in inflammatory and malignant effusions (76,78,79). VEGF dependent tyrosine phosphorylation of adherens junction proteins and the dynamic interaction between n-cadherin and β-catenin, are key determinants of mesothelial paracellular permeability. Upon exposure to noxious stimuli, the interaction of surface ligands with intercellular molecules expressed on mesothelial cells can cause cell migration and leakage of high molecular weight proteins across the pleural membrane, leading to the formation of a pleural effusion.

Parapneumonic effusion and Empyema

A characteristic feature of parapneumonic effusions is the accumulation of neutrophils and mononuclear phagocytes. Pleural fluid from patients with uncomplicated parapneumonic effusions and empyemas contains higher levels of IL-8 (released by PMCs) than pleural effusions from patients with malignancy, tuberculosis, or heart failure (80). Interestingly, PMCs produce IL-8 in a polar manner during pleural inflammation, and thereby regulate the influx of neutrophils into the pleural space (81). Moreover, antibodies to IL-8 can mediate inhibition of neutrophil entry into the pleural space (82). PMCs have also been shown to release Hsp72 [an isoform of Heat shock protein 70 (HSP70)] in response to bacterial infection and levels of Hsp72 are significantly increased in infectious pleural effusions, as compared to non-infectious effusion (83). The role of Hsp2 in the pathogenesis of pleural infection needs to be further explored.

Recently, a novel murine model of pneumonia-associated empyema revealed that *S. pneumoniae* crossed mesothelial layers by translocation through cells rather than by a paracellular route (84). Pleural infection by bacteria, such as *Staphylococcus aureus*, induces the PMCs to release VEGF which alters mesothelial permeability, leading to protein exudation in empyema (78). *S. aureus* activates the early response genes c-fos and c-jun and activator protein-1 (AP-1) in primary mouse PMCs, which may contribute to the activation of pro-apoptotic genes Bak and Bad and release of cytochrome-c and caspase-3, thereby, resulting in apoptosis of PMCs (85). Interestingly, *S. aureus*-activated PMCs appear to extend the life span of recruited polymorphonuclear leukocytes by modulating Bcl-xL and Bak gene expression and activity of active caspases during acute inflammation and empyema (86).

Tuberculous pleural effusion

Early during the course of granulomatous inflammation,

there is a neutrophil-predominant response (87). Subsequently, mononuclear phagocytes engulf mycobacteria resulting in coalescence of mononuclear cells into granulomas. Bacillus Calmette-Guérin (BCG) infection has been shown to induce chemokine expression and increase the production of MIP-1 alpha and MCP-1 (CCL2) by mouse PMCs (88), which is inhibited by IL-4, suggesting that Th1 and Th2 cytokines may regulate the C-C chemokine expression in PMCs and play an important role in mononuclear cell recruitment to the pleural space (89). BCG infection has also been shown to down regulate beta-catenin (an adherens junction protein) expression, decrease electrical resistance across the PMC monolayer, enhance the release of VEGF from PMCs, and increase permeability across the mesothelial monolayer (90). In tuberculous pleuritis, PMCs express intercellular adhesion molecule (ICAM)-1 and facilitate monocyte transmigration across a chemotactic gradient generated by MIP 1-alpha or MCP-1 (91).

Pleural fluids of patients with granulomatous inflammation also contain interferon-γ (IFN-γ), a critical cytokine for the recruitment of mononuclear cells (92). IFN-γ augments cytokine and chemokine production by local cells and causes a significant increase in MCP-1 and MIP-1 production by mesothelial cells (88). IFN-γ also upregulates antimicrobial, phagocytic and T-cell-activating functions, and NO release by PMCs (60).

Malignant pleural effusion

Metastases from cancers of the lung, breast, stomach, and ovary are seen in greater frequency in the pleural space than metastases from other malignancies. Malignant cells can overcome the pleural defense mechanisms by means of various mechanisms (93). For example, the sialomucin complex (SMC) on the PMCs acts as a defense lawyer, and its removal by sialidase (as expressed by ovarian cancer cells HTB-77) increases the susceptibility of the PMC layer to the adherence of malignant cells and to increased metastasis (57).

PMCs produce significant quantities of hyaluronan, which is a ligand for CD44 receptors (94,95). Malignant cells internalize the CD44-hyaluronan complex and hydrolyze it to several low-molecular-weight oligosaccharides. These oligosaccharides are angiogenic and also chemotactic for malignant cells and increase the permeability of the mesothelial monolayer. Low-molecular-weight hyaluronan also induces malignant mesothelioma cell proliferation and haptotaxis via interaction of the CD44 receptor (96).

VEGF and basic fibroblast growth factor (bFGF) released by malignant cells increase the permeability of the surrounding tissues to allow for neovascularization of the pleural surface. Angiogenesis develops an environment surrounded by blood vessels through which the malignant cells can be nourished and is crucial for their growth. Cancer cells can also induce PMCs to release VEGF, increase the permeability of the monolayer, and can also produce autocrine growth factors (97).

Endostatin, released by normal cells and tissues, induces cell cycle arrest and apoptosis, inhibits endothelial cell migration, inhibits angiogenesis and reduces tumor growth (98). It is a potential defense mechanism of PMCs against invading malignant cells. The pleural fluids from patients with malignant pleural effusions contain significantly lower levels of endostatin when compared with fluids from patients with congestive heart failure (99). Interestingly, talc insufflation has been noted to induce PMCs to release endostatin (100).

Asbestos related pleural disease

PMCs have been shown to initiate the inflammatory response to asbestos by releasing chemotaxins for neutrophils in the presence of crocidolite (101). Asbestos directly stimulates PMCs to synthesize IL-8, which may play an important role in mediating asbestos induced pleural inflammation (102). Crocidolite asbestos has been shown to induce PDGF mediated fibroblast proliferation in the pleura (98). Moreover, PMCs actively phagocytose asbestos fibers, which seem to stimulate PMC fibronectin synthesis that may play a role in the induction of pleural fibrosis (103).

Carbon nanotubes (CNT) have recently been shown to cause a length-dependent, asbestos-like inflammatory response via a significant release of acute phase cytokines such as IL-1β, TNFα, IL-6 and IL-8 from the macrophages. When treated with conditioned media from CNT-treated macrophages, mesothelial cells cause a dramatic release of cytokines, which can potentiate the pro-inflammatory response of macrophages that can lead to fiber, related pleural disease (104).

Malignant pleural mesothelioma (MPM)

The Eph transmembrane tyrosine kinases constitute the largest family of receptor tyrosine kinases. The Eph receptors are capable of recognizing signals from the cell microenvironment and influencing cell-cell interaction and migration. EphA2 overexpression has been implicated in tumor growth, angiogenesis, and metastasis, and has been noted in aggressive malignancies (105-108). Overexpression

of EphA2, as seen in malignant mesothelioma cell lines, significantly increases the haptotactic migration of the malignant mesothelioma cells while downregulation of EphA2 expression causes inhibition of cell proliferation and haptotactic migration, and induction of apoptosis through caspase-9 activation (109). Moreover, activation of the EphA2 receptor by its ligand ephrinA1 downregulates total EphA2 expression via phosphorylation and suppresses the growth of MPM cells via ERK1/2 signaling (110). Receptor EphA2 inhibition has been suggested as an approach for inhibiting MPM growth as it has been shown to induce both extrinsic and intrinsic apoptotic pathways in MPM Cells (111).

High levels of activated HGF and c-Met have been observed in mesothelioma and these correlate with disease relapse and poor prognosis (112-114). Inhibition of HGF signaling can block phosphorylation of downstream signaling molecules, cell growth, migration and invasion in mesothelioma (115-117). HGF has also been implicated in dissemination of mesothelioma by inducing mesothelial cells to round up, separate and detach from the serosal surface and then stimulate invasion of adhering tumour cells (118-121).

IL-8, a proinflammatory and angiogenic cytokine, has been described to function as an autocrine growth factor and plays an important role in tumor-related neovascularization (122). Antibody treatment against IL-8 has been shown to decrease human MPM progression (123).

A term mesodermoma has been suggested to define neoplasms arising from undifferentiated and multipotential mesoderm (124). The different tissue types seen in malignant mesothelioma and other serosal pathologies may be a result of mesothelial cell differentiation. The expression of CD26 has been shown to be increased in various cancers (125) and it has been demonstrated that CD26 upregulates periostin secretion by malignant pleural mesothelioma (MPM) cells (126). In a study of a retrospective cohort of 352 patients, immunohistochemistry of a tissue microarray showed that the activation of periostin-triggered EMT is associated with the sarcomatoid histotype of malignant mesothelioma and has an impact on shorter survival of patients (127).

Recently, in an orthotopic model of human pleural malignancy, intrapleural chimeric antigen receptor (CAR) T cell therapy caused antigen-induced T cell activation and a robust CAR T cell expansion and effector differentiation, resulting in increased antitumor efficacy. Interestingly, a significant finding of this study was that regional T cell administration also promoted elimination of extrathoracic tumor sites (104).

Resolution of pleural inflammation

The resolution of pleural inflammation is primarily dependent upon the resolution of the inciting process, for example, eradication of the pathogenic microbe and microbial products from the pleural space in case of pleural infection. Inflammation of the pleural surface may resolve without fibrosis with regeneration of a normal mesothelial surface, or with fibrosis that involves the production and proliferation of fibroblasts.

Repair of injured pleura without fibrosis not only requires a re-establishment of the normal pleural mesothelial monolayer but also a downregulation of the inflammatory response, including inhibition of fibroblast proliferation and collagen synthesis. Rat PMCs exhibit chemotaxis and proliferation in response to thrombin in a dose-dependent manner, suggesting that thrombin may play an important role in the regulation of pleural repair without fibrosis and the re-establishment of the mesothelial monolayer (128). PMCs produce prostaglandin E2 (PGE2), the release of which is completely blocked by anti-thrombin 3 and indomethacin suggesting its role in the repair process of pleural injury (129). In addition, MCP-1 induces proliferative and haptotactic responses in PMCs, which may play a crucial role in the regeneration of the mesothelium and re-epithelialization of the denuded basement membrane at the site of pleural injury during the process of pleural repair (130).

Pleural fibrosis

Pleural fibrosis can be a result of various inflammatory processes such as rheumatoid pleurisy, bacterial empyema, asbestos exposure, malignancy, retained hemothorax, and medications (6). PMCs play a pivotal role in the initiation and maintenance of pleural inflammation that is driven by cytokines and a large number of inflammatory cells that are recruited to the pleural space.

Transforming growth factor-β (TGF-β)

A key abnormality in most fibrotic diseases is the overproduction of TGF-β, a family of multifunctional growth-modulating cytokines. PMCs express receptors for TGF-β, and elevated levels of TGF-β have been found in pleural effusions and pleural tissues during disease processes (131). TGF-β regulates cell proliferation, cell migration, cell differentiation, and extracellular matrix production and is a potent chemo-attractant for fibroblasts (1). Mesothelial cell stimulation by TGF-β leads to increased synthesis of collagen, matrix

proteins, matrix metalloproteinase-1 and -9, and tissue inhibitor of matrix metalloproteinases-2 (132,133). The presence of high levels of TGF-β in empyema, tuberculous pleuritis, and asbestos-related pleural effusions suggests a role in pleural fibrosis (134-136). In animal models, intrapleural administration of TGF-β induces pleural scarring and mediates pleurodesis (137,138).

Platelet-derived growth factor (PDGF)

Mesothelial cells produce PDGF (139), a mitogenic cytokine for mesothelial cells (140), that stimulates hyaluronan production in mesothelial cells and fibroblasts and promotes the growth of fibroblasts (141,142). Moreover, PDGF stimulates collagen production by mesothelial cells. In mouse models, PDGF mediates fibroblast proliferation in the pleura in response to inhaled crocidolite asbestos fibers, whereas antibodies against PDGF inhibit fibroblast proliferation (143). Furthermore, PDGF also induces the expression of TGF-β, thereby potentiating the fibrotic response (144).

Basic fibroblast growth factor (bFGF)

bFGF, also called fibroblast growth factor-2 (FGF-2), stimulates mesothelial cell proliferation *in vitro* and *in vivo* (145). This angiogenic factor is mitogenic for fibroblasts, smooth muscle cells, and endothelial cells (146-148) and is present in pleural effusions of various etiologies (149,150). In one study, bFGF levels were higher in the pleural fluid of patients who underwent successful talc pleurodesis compared to those who failed talc pleurodesis (151). Moreover, mesothelial cells stimulated with talc were noted to release higher amounts of bFGF when compared to controls (151).

Hepatocyte growth factor (HGF)

The role of HGF may be opposite to that of TGF-β or b-FGF in pleural fibrosis (118). Elevated HGF levels have been reported in serosal (pleural and peritoneal) fluids, serum, and bronchoalveolar lavage (BAL) and pulmonary edema fluid of patients with various diseases (152-156). HGF stimulates proliferation, migration and collagen production in mesothelial cells (156-158). Increasing HGF levels in the lung and other organs enhances repair and slows the progression of fibrosis (159-166), while inhibition of HGF by neutralizing antibodies increases fibrosis (167). The role of HGF in repair has been described for various tissues, but its role in the pleura is not well established.

Disordered fibrin turnover

Disordered fibrin turnover plays an important role in the pathogenesis of pleural fibrosis (168). Formation of a transitional fibrin neomatrix contributes to tissue organization and fibrotic repair during the process of wound healing. With ongoing remodeling, collagen deposition occurs and leads to progressive scarring and fibrotic repair (168).

Tissue factor is expressed by mesothelial cells, macrophages, and fibroblasts (169-171) and is detectable in the pleural fluid (172). The concurrent expression of tissue factor pathway inhibitor (TFPI) by PMCs regulates the process of coagulation in the pleural space (172,173). In the setting of pleural injury, the level of intrapleural tissue factor appears to exceed that of TFPI and intrapleural coagulation is upregulated in patients with exudative effusions compared to patients with effusions due to congestive heart failure (172).

The PMCs and recruited inflammatory cells can produce components of both the fibrinolytic system and inhibitors of the fibrinolytic system including tissue plasminogen activator, urokinase, urokinase receptor, and plasminogen activator inhibitor-1 (PAI-1) and are hypothesized to be involved in the pathogenesis of pleural injury and fibrosis (172).

Human PMCs secrete urokinase and tissue plasminogen activator, which are detectable in pleural effusions in a free form and complexed to PAI-1 and PAI-2 (169,172). Both urokinase and tissue plasminogen activator can activate plasminogen present in pleural fluids with the subsequent generation of plasmin. PMCs, macrophages, and lung fibroblasts also express urokinase receptors (174-176). Both urokinase and urokinase receptor are involved in the regulation of cytokine-mediated cellular signaling and cell trafficking (177). Moreover, urokinase is a chemotaxin and a mitogen for mesothelial cells and lung fibroblasts (174,178). Tissue plasminogen activator is mainly responsible for intravascular thrombolysis while urokinase is mainly involved in extravascular proteolysis and tissue remodeling (179). TGF-β increases mesothelial cell production PAI-1 and PAI-2, which are the major inhibitors of urokinase mediated intrapleural fibrin clearance and can lead to accelerated pleural connective tissue matrix organization and pleural fibrosis (169,180). The complex interplay of urokinase, urokinase receptor, and PAI responses determines the local fibrinolytic activity and influence the processes of pleural inflammation and repair, and development of pleural fibrosis.

Pleurodesis

Pleurodesis is the process of obliteration of the pleural space and absence of defining surfaces between the parietal and visceral pleura. Talc mediates pleurodesis by stimulating PMCs to release chemokines such as IL-8 and MCP-1,

causing increased chemotactic activity for neutrophils and monocytes, and enhancing the expression of ICAM-1 (181). Talc has also been shown to induce PMCs to release bFGF and PDGF (151). In one study, pleural fluids collected after talc insufflation and conditioned media from talc-activated PMCs were noted to induce apoptosis in human umbilical vein endothelial cells. Thus, talc appears to alter the angiogenic balance in the pleural space from a biologically active and angiogenic environment to a more angiostatic milieu (100).

Tetracyclines cause pleurodesis by stimulating the PMCs to produce a growth-factor-like activity for fibroblasts (182). Intrapleural administration of TGF-β has been shown to induce pleurodesis in animal models (137,138). It has been suggested that unlike talc and tetracycline, TGF-β can induce collagen synthesis without stimulating PMCs to release IL-8 and provoking pleural inflammation (183). It is noteworthy that TGF-β can induce transient production of large pleural effusions possibly due to increased production of VEGF from PMCs (184). Interestingly, in case of significantly advanced malignant pleural disease, where talc or another sclerosing agent may have little interaction with normal PMCs, the fibrotic response has been found to be attenuated, emphasizing the role of PMCs in pleural fibrosis (4).

Idiopathic pulmonary fibrosis (IPF)

IPF is a rapidly progressive lung disease of unknown etiology, with limited therapeutic options and a median survival of 3-5 years (185). Fibrotic remodeling in IPF occurs by mesenchymal cell proliferation and the differentiation of progenitor cells into myofibroblasts, which secrete excessive amounts of extracellular matrix resulting in scarring and destruction of the lung architecture (46,186,187). It begins in the distal sub-pleural regions and progresses proximally into the lung parenchyma, the reasons for which are poorly understood (188). High-resolution computed tomography (189) and 3-dimensional (3D) morphometric analysis (190) of the IPF lung suggest a complex and highly interconnected reticulum of fibrous tissue extending from the pleura into the underlying parenchyma. These findings suggest an intrinsic factor of the pleura as the culprit for IPF.

The extent of fibroblastic foci present on lung biopsy predicts survival in IPF patients (191,192). The mechanisms involved in the formation of fibroblastic foci and the origin of myofibroblasts are poorly understood (193). Also, there is no clear explanation for the histopathological pattern of usual interstitial pneumonia (UIP) and its peripheral localization (194). The reasons for association of IPF with ageing and aberrant epithelial activation are also unknown, but there is some evidence to suggest that an abnormal recapitulation of developmental pathways may play a role (188).

Pleural mesothelial cells in IPF

The embryonic mesoderm plays a critical role in lung-branching morphogenesis, vasculogenesis, and alveologenesis, the latter involving septation by alveolar fibroblasts (195). In response to airway-alveolar injury, the pleural mesothelium may mobilize reparative cells in a process that replicates features of embryonic development (16,75,196-198). PMCs respond to their microenvironment and have the capacity to differentiate into adipocytes, endothelial cells, and osteoblasts, suggesting remarkable plasticity (46,51,75,85). EMT seems to play a role in liver, kidney, and lung fibrosis (199). Some studies have suggested a role for EMT in the generation of myofibroblasts in lung parenchyma (200-202), although other studies appear to contradict this in injury-provoked lung fibrosis (203).

Wt1 expressing cells, including PMCs, have the capacity to switch between a mesenchymal and epithelial state (204). A balance between the epithelial and mesenchymal states of cells is essential for normal development and for maintenance of adult tissue homeostasis (205). Wt1 is necessary for the morphologic integrity of pleural membrane and loss of Wt1 contributes to IPF via MMT of PMCs into a myofibroblast phenotype (206). Wt1 expressing PMCs have been shown to migrate into the lung parenchyma and differentiate into subpopulations of bronchial smooth muscle cells, vascular smooth muscle cells, fibroblasts, and also myofibroblasts supporting the hypothesis that IPF may be an altered recapitulation of development (42,43,207). Other studies have demonstrated the differentiation of PMCs into myofibroblasts in response to transforming growth factor TGF-β1 (51,208). In response to TGF-β1, PMCs lose their polarity and cell-cell junctional complexes, migrate into lung parenchyma, and undergo phenotypic transition into myofibroblasts via smad-2 signaling (51,208). The demonstration of TGF-β1 induced PMC trafficking into the lung and differentiation into myofibroblasts supports a role for PMCs in the pathogenesis of IPF and suggests a potential role for pleural-based therapies to modulate pleural mesothelial activation and parenchymal fibrosis progression (52).

Pleural mesothelial cell as a potential therapeutic target

Recent studies show that in IPF patients, PMCs are present

in the explanted lung tissue parenchyma (52,208). Moreover, the number of calretinin-positive cells correlate with the degree of fibrotic change seen in the parenchyma (208), as measured by the Ashcroft score (the histo-pathological grading of pulmonary fibrosis (209-212). The finding that PMCs migrate into the lung parenchyma and transform into myofibroblasts provides a rational explanation for the spatio-temporal distribution of fibrosis in IPF and invokes a novel, alternative hypothesis for the origin and source of the myofibroblasts. PMCs not only seem to play a role in the tissue remodeling responses seen in patients with IPF, but may also represent a novel cellular biomarker of disease activity and a potential therapeutic target.

Intra-pleural delivery of compounds is an innovative therapeutic modality that can be refined to deliver drugs targeting the lungs. Direct delivery of the small molecule inhibitors to the pleura can potentially provide a direct and efficient way to deliver a high concentration of the compound to target the pro-fibrogenic activities of PMCs, thereby increasing its efficacy and minimizing systemic toxicity. Intra-pleural delivery may result in higher, sustained drug levels in the BAL fluid when compared with serum levels (208). For example, intrapleural CAR T cell therapy was found to vastly outperform systemically infused T cells even when accumulated at equivalent numbers in the pleural tumor (104).

Several methods such as liposomal drug delivery, nanoparticle (NP) delivery of proteins, and gene therapy have been explored for site-directed delivery of therapeutic agents (213-215). For example, biodegradable fluorescein isothiocyanate (FITC) labeled PLGA (poly-lactic-co-glycolic acid) NPs (which can carry therapeutic compounds conjugated to PLGA) can be coated with antibody targeted to mesothelin (a PMC marker) to allow them to target the pleural surface and potentially diffuse into the lung parenchyma. Intra-pleural delivery of molecules to the lung is feasible and appears to be safe, however, delivery techniques will need to be refined to minimize lung injury.

Conclusions

The PMCs are the most common cells in the pleural space and are quintessential for maintenance of a dynamic state of homeostasis in the pleural space. PMCs are mesenchymal in origin and via the process of EMT, give rise to the endothelium and vascular smooth muscle cells heart, liver and gut during development. In response to TGF-β1 and PDGF, PMCs have been shown to produce mesenchyme, adopt a myofibroblast phenotype *in vitro*, and undergo EMT with subsequent fibrotic changes; suggesting pluripotency of PMCs and their importance in the diseases of lung and the pleura.

PMCs exhibit various innate and acquired immune mechanisms and form the central component of pleural defense mechanisms. These mechanisms include functions such as providing a mechanical barrier to invasion as well as a sophisticated, multilayered, and coordinated system of cytokines and inflammatory cell recruitment. For example, TLRs on PMCs recognize pathogens via PAMPs such as LPS, bacterial lipoproteins, cell wall PGNs, and bacterial and viral nucleic acids; and initiate downstream signaling with production of various peptides with antimicrobial activity, chemokines, and cytokines such as TNF-α, IL-1, IL-6, and IL-8. Transmembrane adherens junction proteins, Cadherins and catenins, and VEGF allow PMCs to regulate pleural permeability and upon exposure to noxious stimuli, the interaction of surface ligands for intercellular molecules expressed on PMCs causes changes in the permeability of the pleural membrane, leading to the formation of a pleural effusion.

Metastases to the pleura are seen in greater frequency, from cancers of lung, breast, stomach, and ovary than from other malignancies. Malignant cells can overcome the pleural defense mechanisms by means of various mechanisms such as removal of SMC by sialidase, hydrolysis of CD44-hyaluronan complex, suppression of endostatin release by PMCs, and by VEGF and bFGF mediated increase in permeability and neovascularization. Overexpression of EphA2 (a member of the Eph transmembrane tyrosine kinase family), as seen in malignant mesothelioma cell lines, significantly increases the haptotactic migration of the malignant mesothelioma cells while downregulation of EphA2 expression causes inhibition of cell proliferation and induction of apoptosis. High levels of activated HGF and c-Met have been observed in mesothelioma and inhibition of HGF signaling can block phosphorylation of downstream signaling molecules, cell growth, migration and invasion in mesothelioma.

Resolution of pleural inflammation may occur without fibrosis with regeneration of a normal mesothelial surface, or with fibrosis. PMCs play a pivotal role in the process of pleural fibrosis via release of TGF-β, PDGF, bFGF and HGF, and by a disordered state of fibrin turnover; resulting in the production and proliferation of fibroblasts. PMCs also migrate into the lung parenchyma and differentiate into subpopulations of bronchial smooth muscle cells, vascular

smooth muscle cells, fibroblasts, and also myofibroblasts suggesting that IPF may be an altered recapitulation of developmental pathways. Moreover, PMCs may represent a novel cellular biomarker of disease activity in IPF and a potential therapeutic target.

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References

1. Antony VB, Sahn SA, Mossman B, et al. NHLBI workshop summaries. Pleural cell biology in health and disease. Am Rev Respir Dis 1992;145:1236-9.
2. Jones JS. The pleura in health and disease. Lung 2001;179:397-413.
3. Owens MW, Grisham MB. Nitric oxide synthesis by rat pleural mesothelial cells: induction by cytokines and lipopolysaccharide. Am J Physiol 1993;265:L110-6.
4. Jantz MA, Antony VB. Pathophysiology of the pleura. Respiration 2008;75:121-33.
5. Jonjić N, Peri G, Bernasconi S, et al. Expression of adhesion molecules and chemotactic cytokines in cultured human mesothelial cells. J Exp Med 1992;176:1165-74.
6. Jantz MA, Antony VB. Pleural fibrosis. Clin Chest Med 2006;27:181-91.
7. Que J, Wilm B, Hasegawa H, et al. Mesothelium contributes to vascular smooth muscle and mesenchyme during lung development. Proc Natl Acad Sci U S A 2008;105:16626-30.
8. Herriges M, Morrisey EE. Lung development: orchestrating the generation and regeneration of a complex organ. Development 2014;141:502-13.
9. Weaver M, Dunn NR, Hogan BL. Bmp4 and Fgf10 play opposing roles during lung bud morphogenesis. Development 2000;127:2695-704.
10. Bellusci S, Furuta Y, Rush MG, et al. Involvement of Sonic hedgehog (Shh) in mouse embryonic lung growth and morphogenesis. Development 1997;124:53-63.
11. Sekine K, Ohuchi H, Fujiwara M, et al. Fgf10 is essential for limb and lung formation. Nat Genet 1999;21:138-41.
12. Goss AM, Tian Y, Tsukiyama T, et al. Wnt2/2b and beta-catenin signaling are necessary and sufficient to specify lung progenitors in the foregut. Dev Cell 2009;17:290-8.
13. Domyan ET, Ferretti E, Throckmorton K, et al. Signaling through BMP receptors promotes respiratory identity in the foregut via repression of Sox2. Development 2011;138:971-81.
14. Ohuchi H, Hori Y, Yamasaki M, et al. FGF10 acts as a major ligand for FGF receptor 2 IIIb in mouse multi-organ development. Biochem Biophys Res Commun 2000;277:643-9.
15. Pepicelli CV, Lewis PM, McMahon AP. Sonic hedgehog regulates branching morphogenesis in the mammalian lung. Curr Biol 1998;8:1083-6.
16. Mutsaers SE. Mesothelial cells: their structure, function and role in serosal repair. Respirology 2002;7:171-91.
17. Muñoz-Chápuli R, Pérez-Pomares JM, Macías D, et al. Differentiation of hemangioblasts from embryonic mesothelial cells? A model on the origin of the vertebrate cardiovascular system. Differentiation 1999;64:133-41.
18. Pérez-Pomares JM, Carmona R, González-Iriarte M, et al. Origin of coronary endothelial cells from epicardial mesothelium in avian embryos. Int J Dev Biol 2002;46:1005-13.
19. Pérez-Pomares JM, Carmona R, González-Iriarte M, et al. Contribution of mesothelium-derived cells to liver sinusoids in avian embryos. Dev Dyn 2004;229:465-74.
20. Zhou B, von Gise A, Ma Q, et al. Genetic fate mapping demonstrates contribution of epicardium-derived cells to the annulus fibrosis of the mammalian heart. Dev Biol 2010;338:251-61.
21. Cai CL, Martin JC, Sun Y, et al. A myocardial lineage derives from Tbx18 epicardial cells. Nature 2008;454:104-8.
22. Dettman RW, Denetclaw W Jr, Ordahl CP, et al. Common epicardial origin of coronary vascular smooth muscle, perivascular fibroblasts, and intermyocardial fibroblasts in the avian heart. Dev Biol 1998;193:169-81.
23. Mikawa T, Gourdie RG. Pericardial mesoderm generates a population of coronary smooth muscle cells migrating into the heart along with ingrowth of the epicardial organ. Dev Biol 1996;174:221-32.
24. Wilm B, Ipenberg A, Hastie ND, et al. The serosal mesothelium is a major source of smooth muscle cells of the gut vasculature. Development 2005;132:5317-28.
25. Kawaguchi M, Bader DM, Wilm B. Serosal mesothelium retains vasculogenic potential. Dev Dyn 2007;236:2973-9.
26. Ohmichi H, Koshimizu U, Matsumoto K, et al. Hepatocyte growth factor (HGF) acts as a mesenchyme-derived morphogenic factor during fetal lung development. Development 1998;125:1315-24.
27. Padela S, Cabacungan J, Shek S, et al. Hepatocyte growth factor is required for alveologenesis in the neonatal rat. Am J Respir Crit Care Med 2005;172:907-14.
28. Haber DA, Buckler AJ, Glaser T, et al. An internal

deletion within an 11p13 zinc finger gene contributes to the development of Wilms' tumor. Cell 1990;61:1257-69.
29. Park S, Schalling M, Bernard A, et al. The Wilms tumour gene WT1 is expressed in murine mesoderm-derived tissues and mutated in a human mesothelioma. Nat Genet 1993;4:415-20.
30. Jomgeow T, Oji Y, Tsuji N, et al. Wilms' tumor gene WT1 17AA(-)/KTS(-) isoform induces morphological changes and promotes cell migration and invasion in vitro. Cancer Sci 2006;97:259-70.
31. Ito K, Oji Y, Tatsumi N, et al. Antiapoptotic function of 17AA(+)WT1 (Wilms' tumor gene) isoforms on the intrinsic apoptosis pathway. Oncogene 2006;25:4217-29.
32. Zhang TF, Yu SQ, Guan LS, et al. Inhibition of breast cancer cell growth by the Wilms' tumor suppressor WT1 is associated with a destabilization of beta-catenin. Anticancer Res 2003;23:3575-84.
33. Loeb DM, Evron E, Patel CB, et al. Wilms' tumor suppressor gene (WT1) is expressed in primary breast tumors despite tumor-specific promoter methylation. Cancer Res 2001;61:921-5.
34. Oji Y, Miyoshi S, Maeda H, et al. Overexpression of the Wilms' tumor gene WT1 in de novo lung cancers. Int J Cancer 2002;100:297-303.
35. Ueda T, Oji Y, Naka N, et al. Overexpression of the Wilms' tumor gene WT1 in human bone and soft-tissue sarcomas. Cancer Sci 2003;94:271-6.
36. Burwell EA, McCarty GP, Simpson LA, et al. Isoforms of Wilms' tumor suppressor gene (WT1) have distinct effects on mammary epithelial cells. Oncogene 2007;26:3423-30.
37. Bax NA, van Oorschot AA, Maas S, et al. In vitro epithelial-to-mesenchymal transformation in human adult epicardial cells is regulated by TGFβ-signaling and WT1. Basic Res Cardiol 2011;106:829-47.
38. Bax NA, Pijnappels DA, van Oorschot AA, et al. Epithelial-to-mesenchymal transformation alters electrical conductivity of human epicardial cells. J Cell Mol Med 2011;15:2675-83.
39. Licciulli S, Kissil JL. WT1: a weak spot in KRAS-induced transformation. J Clin Invest 2010;120:3804-7.
40. Call KM, Glaser T, Ito CY, et al. Isolation and characterization of a zinc finger polypeptide gene at the human chromosome 11 Wilms' tumor locus. Cell 1990;60:509-20.
41. Zhou B, Honor LB, He H, et al. Adult mouse epicardium modulates myocardial injury by secreting paracrine factors. J Clin Invest 2011;121:1894-904.
42. Colvin JS, White AC, Pratt SJ, et al. Lung hypoplasia and neonatal death in Fgf9-null mice identify this gene as an essential regulator of lung mesenchyme. Development 2001;128:2095-106.
43. Dixit R, Ai X, Fine A. Derivation of lung mesenchymal lineages from the fetal mesothelium requires hedgehog signaling for mesothelial cell entry. Development 2013;140:4398-406.
44. Herrick SE, Mutsaers SE. Mesothelial progenitor cells and their potential in tissue engineering. Int J Biochem Cell Biol 2004;36:621-42.
45. Rinkevich Y, Mori T, Sahoo D, et al. Identification and prospective isolation of a mesothelial precursor lineage giving rise to smooth muscle cells and fibroblasts for mammalian internal organs, and their vasculature. Nat Cell Biol 2012;14:1251-60.
46. Lansley SM, Searles RG, Hoi A, et al. Mesothelial cell differentiation into osteoblast- and adipocyte-like cells. J Cell Mol Med 2011;15:2095-105.
47. Decologne N, Kolb M, Margetts PJ, et al. TGF-beta1 induces progressive pleural scarring and subpleural fibrosis. J Immunol 2007;179:6043-51.
48. Margetts PJ, Bonniaud P, Liu L, et al. Transient overexpression of TGF-{beta}1 induces epithelial mesenchymal transition in the rodent peritoneum. J Am Soc Nephrol 2005;16:425-36.
49. Wada AM, Smith TK, Osler ME, et al. Epicardial/Mesothelial cell line retains vasculogenic potential of embryonic epicardium. Circ Res 2003;92:525-31.
50. Yang AH, Chen JY, Lin JK. Myofibroblastic conversion of mesothelial cells. Kidney Int 2003;63:1530-9.
51. Nasreen N, Mohammed KA, Mubarak KK, et al. Pleural mesothelial cell transformation into myofibroblasts and haptotactic migration in response to TGF-beta1 in vitro. Am J Physiol Lung Cell Mol Physiol 2009;297:L115-24.
52. Zolak JS, Jagirdar R, Surolia R, et al. Pleural mesothelial cell differentiation and invasion in fibrogenic lung injury. Am J Pathol 2013;182:1239-47.
53. Hage CA, abdul-Mohammed K, Antony VB. Pathogenesis of pleural infection. Respirology 2004;9:12-5.
54. Medzhitov R, Janeway CA Jr. Innate immunity: impact on the adaptive immune response. Curr Opin Immunol 1997;9:4-9.
55. Ohtsuka A, Yamana S, Murakami T. Localization of membrane-associated sialomucin on the free surface of mesothelial cells of the pleura, pericardium, and peritoneum. Histochem Cell Biol 1997;107:441-7.
56. Sassetti C, Van Zante A, Rosen SD. Identification of endoglycan, a member of the CD34/podocalyxin family of

sialomucins. J Biol Chem 2000;275:9001-10.
57. Sharma RK, Mohammed KA, Nasreen N, et al. Defensive role of pleural mesothelial cell sialomucins in tumor metastasis. Chest 2003;124:682-7.
58. Kroegel C, Antony VB. Immunobiology of pleural inflammation: potential implications for pathogenesis, diagnosis and therapy. Eur Respir J 1997;10:2411-8.
59. Lee YC, Knight DA, Lane KB, et al. Activation of proteinase-activated receptor-2 in mesothelial cells induces pleural inflammation. Am J Physiol Lung Cell Mol Physiol 2005;288:L734-40.
60. Owens MW, Milligan SA, Grisham MB. Nitric oxide synthesis by rat pleural mesothelial cells: induction by growth factors and lipopolysaccharide. Exp Lung Res 1995;21:731-42.
61. Janeway CA Jr, Medzhitov R. Innate immune recognition. Annu Rev Immunol 2002;20:197-216.
62. Tosi MF. Innate immune responses to infection. J Allergy Clin Immunol 2005;116:241-9; quiz 250.
63. Heine H, Ulmer AJ. Recognition of bacterial products by toll-like receptors. Chem Immunol Allergy 2005;86:99-119.
64. Schnare M, Rollinghoff M, Qureshi S. Toll-like receptors: sentinels of host defence against bacterial infection. Int Arch Allergy Immunol 2006;139:75-85.
65. Hussain T, Nasreen N, Lai Y, et al. Innate immune responses in murine pleural mesothelial cells: Toll-like receptor-2 dependent induction of beta-defensin-2 by staphylococcal peptidoglycan. Am J Physiol Lung Cell Mol Physiol 2008;295:L461-70.
66. Delves PJ, Roitt IM. The immune system. Second of two parts. N Engl J Med 2000;343:108-17.
67. Delves PJ, Roitt IM. The immune system. First of two parts. N Engl J Med 2000;343:37-49.
68. Pazgier M, Hoover DM, Yang D, et al. Human beta-defensins. Cell Mol Life Sci 2006;63:1294-313.
69. Niyonsaba F, Ogawa H, Nagaoka I. Human beta-defensin-2 functions as a chemotactic agent for tumour necrosis factor-alpha-treated human neutrophils. Immunology 2004;111:273-81.
70. Ashitani J, Mukae H, Nakazato M, et al. Elevated pleural fluid levels of defensins in patients with empyema. Chest 1998;113:788-94.
71. Varano Della Vergiliana JF, Lansley S, Tan AL, Creaney J, et al. Mesothelial cells activate the plasma kallikrein-kinin system during pleural inflammation. Biol Chem 2011;392:633-42.
72. Corada M, Liao F, Lindgren M, et al. Monoclonal antibodies directed to different regions of vascular endothelial cadherin extracellular domain affect adhesion and clustering of the protein and modulate endothelial permeability. Blood 2001;97:1679-84.
73. Dejana E. Endothelial adherens junctions: implications in the control of vascular permeability and angiogenesis. J Clin Invest 1996;98:1949-53.
74. Blankesteijn WM, van Gijn ME, Essers-Janssen YP, et al. Beta-catenin, an inducer of uncontrolled cell proliferation and migration in malignancies, is localized in the cytoplasm of vascular endothelium during neovascularization after myocardial infarction. Am J Pathol 2000;157:877-83.
75. Antony VB. Immunological mechanisms in pleural disease. Eur Respir J 2003;21:539-44.
76. Thickett DR, Armstrong L, Millar AB. Vascular endothelial growth factor (VEGF) in inflammatory and malignant pleural effusions. Thorax 1999;54:707-10.
77. Becker PM, Alcasabas A, Yu AY, Oxygen-independent upregulation of vascular endothelial growth factor and vascular barrier dysfunction during ventilated pulmonary ischemia in isolated ferret lungs. Am J Respir Cell Mol Biol 2000;22:272-9.
78. Mohammed KA, Nasreen N, Hardwick J, et al. Bacterial induction of pleural mesothelial monolayer barrier dysfunction. Am J Physiol Lung Cell Mol Physiol 2001;281:L119-25.
79. Ishimoto O, Saijo Y, Narumi K, et al. High level of vascular endothelial growth factor in hemorrhagic pleural effusion of cancer. Oncology 2002;63:70-5.
80. Strieter RM, Koch AE, Antony VB, et al. The immunopathology of chemotactic cytokines: the role of interleukin-8 and monocyte chemoattractant protein-1. J Lab Clin Med 1994;123:183-97.
81. Nasreen N, Mohammed KA, Hardwick J, et al. Polar production of interleukin-8 by mesothelial cells promotes the transmesothelial migration of neutrophils: role of intercellular adhesion molecule-1. J Infect Dis 2001;183:1638-45.
82. Broaddus VC, Boylan AM, Hoeffel JM, et al. Neutralization of IL-8 inhibits neutrophil influx in a rabbit model of endotoxin-induced pleurisy. J Immunol 1994;152:2960-7.
83. Varano Della Vergiliana JF, Lansley SM, Porcel JM, et al. Bacterial infection elicits heat shock protein 72 release from pleural mesothelial cells. PLoS One 2013;8:e63873.
84. Wilkosz S, Edwards LA, Bielsa S, et al. Characterization of a new mouse model of empyema and the mechanisms of pleural invasion by Streptococcus pneumoniae. Am J Respir Cell Mol Biol 2012;46:180-7.

85. Mohammed KA, Nasreen N, Antony VB. Bacterial induction of early response genes and activation of proapoptotic factors in pleural mesothelial cells. Lung 2007;185:355-65.
86. Nasreen N, Mohammed KA, Sanders KL, et al. Pleural mesothelial cells modulate polymorphonuclear leukocyte apoptosis in empyema. J Clin Immunol 2003;23:1-10.
87. Antony VB, Sahn SA, Antony AC, et al. Bacillus Calmette-Guérin-stimulated neutrophils release chemotaxins for monocytes in rabbit pleural spaces and in vitro. J Clin Invest 1985;76:1514-21.
88. Mohammed KA, Nasreen N, Ward MJ, et al. Mycobacterium-mediated chemokine expression in pleural mesothelial cells: role of C-C chemokines in tuberculous pleurisy. J Infect Dis 1998;178:1450-6.
89. Mohammed KA, Nasreen N, Ward MJ, et al. Helper T cell type 1 and 2 cytokines regulate C-C chemokine expression in mouse pleural mesothelial cells. Am J Respir Crit Care Med 1999;159:1653-9.
90. Mohammed KA, Nasreen N, Hardwick J, et al. Mycobacteria induces pleural mesothelial permeability by down-regulating beta-catenin expression. Lung 2003;181:57-66.
91. Nasreen N, Mohammed KA, Ward MJ, et al. Mycobacterium-induced transmesothelial migration of monocytes into pleural space: role of intercellular adhesion molecule-1 in tuberculous pleurisy. J Infect Dis 1999;180:1616-23.
92. Ellner JJ, Barnes PF, Wallis RS, et al. The immunology of tuberculous pleurisy. Semin Respir Infect 1988;3:335-42.
93. Antony VB. Pathogenesis of malignant pleural effusions and talc pleurodesis. Pneumologie 1999;53:493-8.
94. Bourguignon LY, Lokeshwar VB, Chen X, et al. Hyaluronic acid-induced lymphocyte signal transduction and HA receptor (GP85/CD44)-cytoskeleton interaction. J Immunol 1993;151:6634-44.
95. Ponta H, Wainwright D, Herrlich P. The CD44 protein family. Int J Biochem Cell Biol 1998;30:299-305.
96. Nasreen N, Mohammed KA, Hardwick J, et al. Low molecular weight hyaluronan induces malignant mesothelioma cell (MMC) proliferation and haptotaxis: role of CD44 receptor in MMC proliferation and haptotaxis. Oncol Res 2002;13:71-8.
97. Sriram PS, Mohammed KA, Nasreen N, et al. Adherence of ovarian cancer cells induces pleural mesothelial cell (PMC) permeability. Oncol Res 2002;13:79-85.
98. O'Reilly MS, Boehm T, Shing Y, et al. Endostatin: an endogenous inhibitor of angiogenesis and tumor growth. Cell 1997;88:277-85.
99. Nasreen N, Mohammed KA, Sanders K, et al. Pleural mesothelial cell (PMC) defense mechanisms against malignancy. Oncol Res 2003;14:155-61.
100. Nasreen N, Mohammed KA, Brown S, et al. Talc mediates angiostasis in malignant pleural effusions via endostatin induction. Eur Respir J 2007;29:761-9.
101. Antony VB, Owen CL, Hadley KJ. Pleural mesothelial cells stimulated by asbestos release chemotactic activity for neutrophils in vitro. Am Rev Respir Dis 1989;139:199-206.
102. Boylan AM, Rüegg C, Kim KJ, et al. Evidence of a role for mesothelial cell-derived interleukin 8 in the pathogenesis of asbestos-induced pleurisy in rabbits. J Clin Invest 1992;89:1257-67.
103. Kuwahara M, Kuwahara M, Verma K, et al. Asbestos exposure stimulates pleural mesothelial cells to secrete the fibroblast chemoattractant, fibronectin. Am J Respir Cell Mol Biol 1994;10:167-76.
104. Adusumilli PS, Cherkassky L, Villena-Vargas J, et al. Regional delivery of mesothelin-targeted CAR T cell therapy generates potent and long-lasting CD4-dependent tumor immunity. Sci Transl Med 2014;6:261ra151.
105. Xu F, Zhong W, Li J, et al. Predictive value of EphA2 and EphrinA-1 expression in oesophageal squamous cell carcinoma. Anticancer Res 2005;25:2943-50.
106. Thaker PH, Deavers M, Celestino J, et al. EphA2 expression is associated with aggressive features in ovarian carcinoma. Clin Cancer Res 2004;10:5145-50.
107. Kinch MS, Moore MB, Harpole DH Jr. Predictive value of the EphA2 receptor tyrosine kinase in lung cancer recurrence and survival. Clin Cancer Res 2003;9:613-8.
108. Easty DJ, Herlyn M, Bennett DC. Abnormal protein tyrosine kinase gene expression during melanoma progression and metastasis. Int J Cancer 1995;60:129-36.
109. Nasreen N, Mohammed KA, Antony VB. Silencing the receptor EphA2 suppresses the growth and haptotaxis of malignant mesothelioma cells. Cancer 2006;107:2425-35.
110. Nasreen N, Mohammed KA, Lai Y, et al. Receptor EphA2 activation with ephrinA1 suppresses growth of malignant mesothelioma (MM). Cancer Lett 2007;258:215-22.
111. Mohammed KA, Wang X, Goldberg EP, et al. Silencing receptor EphA2 induces apoptosis and attenuates tumor growth in malignant mesothelioma. Am J Cancer Res 2011;1:419-31.
112. Harvey P, Warn A, Dobbin S, et al. Expression of HGF/SF in mesothelioma cell lines and its effects on cell motility, proliferation and morphology. Br J Cancer 1998;77:1052-9.
113. Harvey P, Warn A, Newman P, et al. Immunoreactivity

for hepatocyte growth factor/scatter factor and its receptor, met, in human lung carcinomas and malignant mesotheliomas. J Pathol 1996;180:389-94.
114. Tolnay E, Kuhnen C, Wiethege T, et al. Hepatocyte growth factor/scatter factor and its receptor c-Met are overexpressed and associated with an increased microvessel density in malignant pleural mesothelioma. J Cancer Res Clin Oncol 1998;124:291-6.
115. Altomare DA, You H, Xiao GH, et al. Human and mouse mesotheliomas exhibit elevated AKT/PKB activity, which can be targeted pharmacologically to inhibit tumor cell growth. Oncogene 2005;24:6080-9.
116. Jagadeeswaran R, Ma PC, Seiwert TY, et al. Functional analysis of c-Met/hepatocyte growth factor pathway in malignant pleural mesothelioma. Cancer Res 2006;66:352-61.
117. Mukohara T, Civiello G, Davis IJ, et al. Inhibition of the met receptor in mesothelioma. Clin Cancer Res 2005;11:8122-30.
118. Mutsaers SE, Kalomenidis I, Wilson NA, et al. Growth factors in pleural fibrosis. Curr Opin Pulm Med 2006;12:251-8.
119. Yashiro M, Chung YS, Inoue T, et al. Hepatocyte growth factor (HGF) produced by peritoneal fibroblasts may affect mesothelial cell morphology and promote peritoneal dissemination. Int J Cancer 1996;67:289-93.
120. Harvey P, Clark IM, Jaurand MC, et al. Hepatocyte growth factor/scatter factor enhances the invasion of mesothelioma cell lines and the expression of matrix metalloproteinases. Br J Cancer 2000;83:1147-53.
121. Uchiyama A, Morisaki T, Beppu K, et al. Hepatocyte growth factor and invasion-stimulatory activity are induced in pleural fluid by surgery in lung cancer patients. Br J Cancer 1999;81:721-6.
122. Antony VB, Hott JW, Godbey SW, et al. Angiogenesis in mesotheliomas. Role of mesothelial cell derived IL-8. Chest 1996;109:21S-22S.
123. Galffy G, Mohammed KA, Nasreen N, et al. Inhibition of interleukin-8 reduces human malignant pleural mesothelioma propagation in nude mouse model. Oncol Res 1999;11:187-94.
124. Donna A, Betta PG. Mesodermomas: a new embryological approach to primary tumours of coelomic surfaces. Histopathology 1981;5:31-44.
125. Havre PA, Abe M, Urasaki Y, et al. The role of CD26/dipeptidyl peptidase IV in cancer. Front Biosci 2008;13:1634-45.
126. Komiya E, Ohnuma K, Yamazaki H, et al. CD26-mediated regulation of periostin expression contributes to migration and invasion of malignant pleural mesothelioma cells. Biochem Biophys Res Commun 2014;447:609-15.
127. Schramm A, Opitz I, Thies S, et al. Prognostic significance of epithelial-mesenchymal transition in malignant pleural mesothelioma. Eur J Cardiothorac Surg 2010;37:566-72.
128. Hott JW, Sparks JA, Godbey SW, et al. Mesothelial cell response to pleural injury: thrombin-induced proliferation and chemotaxis of rat pleural mesothelial cells. Am J Respir Cell Mol Biol 1992;6:421-5.
129. Hott JW, Godbey SW, Antony VB. Mesothelial cell modulation of pleural repair: thrombin stimulated mesothelial cells release prostaglandin E2. Prostaglandins Leukot Essent Fatty Acids 1994;51:329-35.
130. Nasreen N, Mohammed KA, Galffy G, et al. MCP-1 in pleural injury: CCR2 mediates haptotaxis of pleural mesothelial cells. Am J Physiol Lung Cell Mol Physiol 2000;278:L591-8.
131. Lee YC, Lane KB. The many faces of transforming growth factor-beta in pleural diseases. Curr Opin Pulm Med 2001;7:173-9.
132. Harvey W, Amlot PL. Collagen production by human mesothelial cells in vitro. J Pathol 1983;139:337-47.
133. Ma C, Tarnuzzer RW, Chegini N. Expression of matrix metalloproteinases and tissue inhibitor of matrix metalloproteinases in mesothelial cells and their regulation by transforming growth factor-beta1. Wound Repair Regen 1999;7:477-85.
134. Maeda J, Ueki N, Ohkawa T, et al. Local production and localization of transforming growth factor-beta in tuberculous pleurisy. Clin Exp Immunol 1993;92:32-8.
135. Jagirdar J, Lee TC, Reibman J, et al. Immunohistochemical localization of transforming growth factor beta isoforms in asbestos-related diseases. Environ Health Perspect 1997;105:1197-203.
136. Sasse SA, Jadus MR, Kukes GD. Pleural fluid transforming growth factor-beta1 correlates with pleural fibrosis in experimental empyema. Am J Respir Crit Care Med 2003;168:700-5.
137. Light RW, Cheng DS, Lee YC, et al. A single intrapleural injection of transforming growth factor-beta(2) produces an excellent pleurodesis in rabbits. Am J Respir Crit Care Med 2000;162:98-104.
138. Lee YC, Lane KB, Parker RE, et al. Transforming growth factor beta(2) (TGF beta(2)) produces effective pleurodesis in sheep with no systemic complications. Thorax 2000;55:1058-62.
139. Waters CM, Chang JY, Glucksberg MR, et al. Mechanical forces alter growth factor release by pleural mesothelial

cells. Am J Physiol 1997;272:L552-7.
140. Owens MW, Milligan SA. Growth factor modulation of rat pleural mesothelial cell mitogenesis and collagen synthesis. Effects of epidermal growth factor and platelet-derived factor. Inflammation 1994;18:77-87.
141. Safi A, Sadmi M, Martinet N, et al. Presence of elevated levels of platelet-derived growth factor (PDGF) in lung adenocarcinoma pleural effusions. Chest 1992;102:204-7.
142. Heldin P, Asplund T, Ytterberg D, et al. Characterization of the molecular mechanism involved in the activation of hyaluronan synthetase by platelet-derived growth factor in human mesothelial cells. Biochem J 1992;283 :165-70.
143. Adamson IY, Prieditis H, Young L. Lung mesothelial cell and fibroblast responses to pleural and alveolar macrophage supernatants and to lavage fluids from crocidolite-exposed rats. Am J Respir Cell Mol Biol 1997;16:650-6.
144. Pierce GF, Mustoe TA, Lingelbach J, et al. Platelet-derived growth factor and transforming growth factor-beta enhance tissue repair activities by unique mechanisms. J Cell Biol 1989;109:429-40.
145. Mutsaers SE, McAnulty RJ, Laurent GJ, et al. Cytokine regulation of mesothelial cell proliferation in vitro and in vivo. Eur J Cell Biol 1997;72:24-9.
146. Bikfalvi A, Klein S, Pintucci G, et al. Biological roles of fibroblast growth factor-2. Endocr Rev 1997;18:26-45.
147. Friesel RE, Maciag T. Molecular mechanisms of angiogenesis: fibroblast growth factor signal transduction. FASEB J 1995;9:919-25.
148. Folkman J, Klagsbrun M, Sasse J, et al. A heparin-binding angiogenic protein--basic fibroblast growth factor--is stored within basement membrane. Am J Pathol 1988;130:393-400.
149. Abramov Y, Anteby SO, Fasouliotis SJ, et al. Markedly elevated levels of vascular endothelial growth factor, fibroblast growth factor, and interleukin 6 in Meigs syndrome. Am J Obstet Gynecol 2001;184:354-5.
150. Strizzi L, Vianale G, Catalano A, et al. Basic fibroblast growth factor in mesothelioma pleural effusions: correlation with patient survival and angiogenesis. Int J Oncol 2001;18:1093-8.
151. Antony VB, Nasreen N, Mohammed KA, et al. Talc pleurodesis: basic fibroblast growth factor mediates pleural fibrosis. Chest 2004;126:1522-8.
152. Eagles G, Warn A, Ball RY, et al. Hepatocyte growth factor/scatter factor is present in most pleural effusion fluids from cancer patients. Br J Cancer 1996;73:377-81.
153. Funakoshi H, Nakamura T. Hepatocyte growth factor: from diagnosis to clinical applications. Clin Chim Acta 2003;327:1-23.
154. Adamson IY, Bakowska J. KGF and HGF are growth factors for mesothelial cells in pleural lavage fluid after intratracheal asbestos. Exp Lung Res 2001;27:605-16.
155. Dikmen E, Kara M, Kisa U, et al. Human hepatocyte growth factor levels in patients undergoing thoracic operations. Eur Respir J 2006;27:73-6.
156. Rampino T, Cancarini G, Gregorini M, et al. Hepatocyte growth factor/scatter factor released during peritonitis is active on mesothelial cells. Am J Pathol 2001;159:1275-85.
157. Adamson IY, Bakowska J, Prieditis H. Proliferation of rat pleural mesothelial cells in response to hepatocyte and keratinocyte growth factors. Am J Respir Cell Mol Biol 2000;23:345-9.
158. Warn R, Harvey P, Warn A, et al. HGF/SF induces mesothelial cell migration and proliferation by autocrine and paracrine pathways. Exp Cell Res 2001;267:258-66.
159. Dworkin LD, Gong R, Tolbert E, et al. Hepatocyte growth factor ameliorates progression of interstitial fibrosis in rats with established renal injury. Kidney Int 2004;65:409-19.
160. Mizuno S, Kurosawa T, Matsumoto K, et al. Hepatocyte growth factor prevents renal fibrosis and dysfunction in a mouse model of chronic renal disease. J Clin Invest 1998;101:1827-34.
161. Mizuno S, Matsumoto K, Li MY, et al. HGF reduces advancing lung fibrosis in mice: a potential role for MMP-dependent myofibroblast apoptosis. FASEB J 2005;19:580-2.
162. Matsuda Y, Matsumoto K, Yamada A, et al. Preventive and therapeutic effects in rats of hepatocyte growth factor infusion on liver fibrosis/cirrhosis. Hepatology 1997;26:81-9.
163. Yaekashiwa M, Nakayama S, Ohnuma K, et al. Simultaneous or delayed administration of hepatocyte growth factor equally represses the fibrotic changes in murine lung injury induced by bleomycin. A morphologic study. Am J Respir Crit Care Med 1997;156:1937-44.
164. Matsuo K, Maeda Y, Naiki Y, et al. Possible effects of hepatocyte growth factor for the prevention of peritoneal fibrosis. Nephron Exp Nephrol 2005;99:e87-94.
165. Watanabe M, Ebina M, Orson FM, et al. Hepatocyte growth factor gene transfer to alveolar septa for effective suppression of lung fibrosis. Mol Ther 2005;12:58-67.
166. Hattori N, Mizuno S, Yoshida Y, et al. The plasminogen activation system reduces fibrosis in the lung by a hepatocyte growth factor-dependent mechanism. Am J Pathol 2004;164:1091-8.
167. Gong R, Rifai A, Tolbert EM, et al. Hepatocyte growth factor ameliorates renal interstitial inflammation in rat

remnant kidney by modulating tubular expression of macrophage chemoattractant protein-1 and RANTES. J Am Soc Nephrol 2004;15:2868-81.
168. Mutsaers SE, Prele CM, Brody AR, et al. Pathogenesis of pleural fibrosis. Respirology 2004;9:428-40.
169. Idell S, Zwieb C, Kumar A, et al. Pathways of fibrin turnover of human pleural mesothelial cells in vitro. Am J Respir Cell Mol Biol 1992;7:414-26.
170. McGee MP, Rothberger H. Tissue factor in bronchoalveolar lavage fluids. Evidence for an alveolar macrophage source. Am Rev Respir Dis 1985;131:331-6.
171. Idell S, Zwieb C, Boggaram J, et al. Mechanisms of fibrin formation and lysis by human lung fibroblasts: influence of TGF-beta and TNF-alpha. Am J Physiol 1992;263:L487-94.
172. Idell S, Girard W, Koenig KB, et al. Abnormalities of pathways of fibrin turnover in the human pleural space. Am Rev Respir Dis 1991;144:187-94.
173. Bajaj MS, Pendurthi U, Koenig K, et al. Tissue factor pathway inhibitor expression by human pleural mesothelial and mesothelioma cells. Eur Respir J 2000;15:1069-78.
174. Shetty S, Kumar A, Johnson AR, et al. Regulation of mesothelial cell mitogenesis by antisense oligonucleotides for the urokinase receptor. Antisense Res Dev 1995;5:307-14.
175. Sitrin RG, Todd RF 3rd, Albrecht E, et al. The urokinase receptor (CD87) facilitates CD11b/CD18-mediated adhesion of human monocytes. J Clin Invest 1996;97:1942-51.
176. Shetty S, Idell S. A urokinase receptor mRNA binding protein from rabbit lung fibroblasts and mesothelial cells. Am J Physiol 1998;274:L871-82.
177. Chapman HA. Plasminogen activators, integrins, and the coordinated regulation of cell adhesion and migration. Curr Opin Cell Biol 1997;9:714-24.
178. Shetty S, Kumar A, Johnson AR, et al. Differential expression of the urokinase receptor in fibroblasts from normal and fibrotic human lungs. Am J Respir Cell Mol Biol 1996;15:78-87.
179. Vassalli JD, Sappino AP, Belin D. The plasminogen activator/plasmin system. J Clin Invest 1991;88:1067-72.
180. Falk P, Ma C, Chegini N, et al. Differential regulation of mesothelial cell fibrinolysis by transforming growth factor beta 1. Scand J Clin Lab Invest 2000;60:439-47.
181. Nasreen N, Hartman DL, Mohammed KA, et al. Talc-induced expression of C-C and C-X-C chemokines and intercellular adhesion molecule-1 in mesothelial cells. Am J Respir Crit Care Med 1998;158:971-8.
182. Antony VB, Rothfuss KJ, Godbey SW, et al. Mechanism of tetracycline-hydrochloride-induced pleurodesis. Tetracycline-hydrochloride-stimulated mesothelial cells produce a growth-factor-like activity for fibroblasts. Am Rev Respir Dis 1992;146:1009-13.
183. Lee YC, Lane KB, Zoia O, et al. Transforming growth factor-beta induces collagen synthesis without inducing IL-8 production in mesothelial cells. Eur Respir J 2003;22:197-202.
184. Gary Lee YC, Melkerneker D, Thompson PJ, et al. Transforming growth factor beta induces vascular endothelial growth factor elaboration from pleural mesothelial cells in vivo and in vitro. Am J Respir Crit Care Med 2002;165:88-94.
185. Khalil N, O'Connor R. Idiopathic pulmonary fibrosis: current understanding of the pathogenesis and the status of treatment. CMAJ 2004;171:153-60.
186. Bjoraker JA, Ryu JH, Edwin MK, et al. Prognostic significance of histopathologic subsets in idiopathic pulmonary fibrosis. Am J Respir Crit Care Med 1998;157:199-203.
187. Hinz B, Gabbiani G. Fibrosis: recent advances in myofibroblast biology and new therapeutic perspectives. F1000 Biol Rep 2010;2:78.
188. King TE Jr, Pardo A, Selman M. Idiopathic pulmonary fibrosis. Lancet 2011;378:1949-61.
189. Misumi S, Lynch DA. Idiopathic pulmonary fibrosis/usual interstitial pneumonia: imaging diagnosis, spectrum of abnormalities, and temporal progression. Proc Am Thorac Soc 2006;3:307-14.
190. Cool CD, Groshong SD, Rai PR, et al. Fibroblast foci are not discrete sites of lung injury or repair: the fibroblast reticulum. Am J Respir Crit Care Med 2006;174:654-8.
191. King TE Jr, Schwarz MI, Brown K, et al. Idiopathic pulmonary fibrosis: relationship between histopathologic features and mortality. Am J Respir Crit Care Med 2001;164:1025-32.
192. King TE Jr, Tooze JA, Schwarz MI, et al. Predicting survival in idiopathic pulmonary fibrosis: scoring system and survival model. Am J Respir Crit Care Med 2001;164:1171-81.
193. Phan SH. Biology of fibroblasts and myofibroblasts. Proc Am Thorac Soc 2008;5:334-7.
194. Raghu G, Collard HR, Egan JJ, et al. An official ATS/ERS/JRS/ALAT statement: idiopathic pulmonary fibrosis: evidence-based guidelines for diagnosis and management. Am J Respir Crit Care Med 2011;183:788-824.
195. Boström H, Willetts K, Pekny M, et al. PDGF-A signaling is a critical event in lung alveolar myofibroblast development and alveogenesis. Cell 1996;85:863-73.
196. Antony VB, Hott JW, Kunkel SL, et al. Pleural mesothelial

cell expression of C-C (monocyte chemotactic peptide) and C-X-C (interleukin 8) chemokines. Am J Respir Cell Mol Biol 1995;12:581-8.
197. Pace E, Ferraro M, Mody CH, et al. Pleural mesothelial cells express both BLT2 and PPARalpha and mount an integrated response to pleural leukotriene B4. J Immunol 2008;181:7292-9.
198. Glista-Baker EE, Taylor AJ, Sayers BC, et al. Nickel nanoparticles enhance platelet-derived growth factor-induced chemokine expression by mesothelial cells via prolonged mitogen-activated protein kinase activation. Am J Respir Cell Mol Biol 2012;47:552-61.
199. Humphreys BD, Lin SL, Kobayashi A, et al. Fate tracing reveals the pericyte and not epithelial origin of myofibroblasts in kidney fibrosis. Am J Pathol 2010;176:85-97.
200. Borok Z. Role for alpha3 integrin in EMT and pulmonary fibrosis. J Clin Invest 2009;119:7-10.
201. Willis BC, duBois RM, Borok Z. Epithelial origin of myofibroblasts during fibrosis in the lung. Proc Am Thorac Soc 2006;3:377-82.
202. Chilosi M, Poletti V, Zamò A, et al. Aberrant Wnt/beta-catenin pathway activation in idiopathic pulmonary fibrosis. Am J Pathol 2003;162:1495-502.
203. Casey BJ, Somerville LH, Gotlib IH, et al. Behavioral and neural correlates of delay of gratification 40 years later: Proc. Natl. Acad. Sci. U.S.A. 2011, Vol 108 No. 36:14998-5003. Ann Neurosci 2012;19:27-8.
204. Scholz H, Kirschner KM. Oxygen-Dependent Gene Expression in Development and Cancer: Lessons Learned from the Wilms' Tumor Gene, WT1. Front Mol Neurosci 2011;4:4.
205. Miller-Hodges E, Hohenstein P. WT1 in disease: shifting the epithelial-mesenchymal balance. J Pathol 2012;226:229-40.
206. Karki S, Surolia R, Hock TD, et al. Wilms' tumor 1 (Wt1) regulates pleural mesothelial cell plasticity and transition into myofibroblasts in idiopathic pulmonary fibrosis. FASEB J 2014;28:1122-31.
207. Batra H, Antony VB. The pleural mesothelium in development and disease. Front Physiol 2014;5:284.
208. Mubarak KK, Montes-Worboys A, Regev D, et al. Parenchymal trafficking of pleural mesothelial cells in idiopathic pulmonary fibrosis. Eur Respir J 2012;39:133-40.
209. Hagiwara SI, Ishii Y, Kitamura S. Aerosolized administration of N-acetylcysteine attenuates lung fibrosis induced by bleomycin in mice. Am J Respir Crit Care Med 2000;162:225-31.
210. Matsuoka H, Arai T, Mori M, et al. A p38 MAPK inhibitor, FR-167653, ameliorates murine bleomycin-induced pulmonary fibrosis. Am J Physiol Lung Cell Mol Physiol 2002;283:L103-12.
211. Simler NR, Howell DC, Marshall RP, et al. The rapamycin analogue SDZ RAD attenuates bleomycin-induced pulmonary fibrosis in rats. Eur Respir J 2002;19:1124-7.
212. Murakami S, Nagaya N, Itoh T, et al. Prostacyclin agonist with thromboxane synthase inhibitory activity (ONO-1301) attenuates bleomycin-induced pulmonary fibrosis in mice. Am J Physiol Lung Cell Mol Physiol 2006;290:L59-65.
213. Watanabe M, Boyer JL, Crystal RG. AAVrh.10-mediated genetic delivery of bevacizumab to the pleura to provide local anti-VEGF to suppress growth of metastatic lung tumors. Gene Ther 2010;17:1042-51.
214. Perez-Soler R, Shin DM, Siddik ZH, et al. Phase I clinical and pharmacological study of liposome-entrapped NDDP administered intrapleurally in patients with malignant pleural effusions. Clin Cancer Res 1997;3:373-9.
215. Liu J, Wong HL, Moselhy J, et al. Targeting colloidal particulates to thoracic lymph nodes. Lung Cancer 2006;51:377-86.

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