REVIEW ARTICLE

Decoding the role of fatty acids and their metabolites in lung fibrosis

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KEY WORDS

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ABSTRACT

Idiopathic pulmonary fibrosis (IPF) is a progressive and life-threatening interstitial lung disease of familial or sporadic onset. The incidence and prevalence of IPF range from 0.09 to 1.3 and from 0.33 to 4.51 per 10000 people, respectively. IPF has a poor prognosis, and death usually occurs within 2 to 5 years following the diagnosis due to secondary respiratory failure. Currently, there are 2 drugs available to treat IPF, pirfenidone and nintedanib. Both only slow the disease progression and, in addition, have unfavorable safety profiles. IPF bears the histology of usual interstitial pneumonia, which is characterized by bronchiolization of distal airspaces, honeycombing, fibroblastic foci, and abnormal epithelial hyperplasia. In the last years, alterations in metabolic pathways, in particular those associated with fatty acid (FA) metabolism have been linked with the pathogenesis of lung fibrosis. Changes in FA profiles have been reported in lung tissue, plasma, and bronchoalveolar lavage fluid of IPF patients, and have been found to correlate with the disease progression and outcome. In addition, they have been associated with the development of a profibrotic phenotype of epithelial cells, macrophages, and fibroblasts/myofibroblasts contributing to their (trans)differentiation and production of the disease-relevant mediators. Furthermore, strategies focusing on the correction of FA profiles in experimental models of lung fibrosis brought advances in understanding tissue scarring processes and contributed to the transition of new molecules into clinical development. This review highlights the role of FAs and their metabolites in IPF and provides evidence for therapeutic potential of lipidome manipulations in the treatment of this disease.

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Idiopathic pulmonary fibrosis Idiopathic pulmonary fibrosis (IPF) is a progressive and life-threatening interstitial lung disease (ILD). The adjusted incidence and prevalence of IPF are estimated to be in the range of 0.09 to 1.3 and 0.33 to 4.51 per 10000 people, respectively.[1] IPF is associated with cough, dyspnea, and markedly impaired quality of life. It has a familial or sporadic onset with a poor prognosis, and death usually occurs within 2–5 years following the diagnosis due to secondary respiratory failure.[2] Some IPF patients may experience acute deterioration of respiratory function over a short period of time. This acute exacerbation often causes death of IPF patients. IPF bears the histology of usual interstitial pneumonia (UIP) which is characterized by bronchiolization of distal airspaces, honeycombing, fibroblastic foci, and abnormal epithelial hyperplasia.[3,4] Diagnosis of IPF is based on identification of the UIP pattern on high-resolution computed tomography scan and/or lung biopsy and exclusion of other ILD conditions. The risk factors increasing the odds of IPF development include smoking, exposure to metallic or wood dust, gastroesophageal reflux, viral infections, and traits that are genetically inherited. Currently,

there are 2 antifibrotic drugs that have been approved for the treatment of IPF, pirfenidone and nintedanib.[5] Both nintedanib, a tyrosine kinase inhibitor, which blocks the effects of platelet-derived growth factor (PDGF), vascular endothelial growth factor, and fibroblast growth factor,[6] and pirfenidone, whose mechanism of action is unclear,[7] were shown to decrease IPF progression, as measured by longitudinal changes of forced vital capacity (FVC) over a period of 52 weeks. Although the approval of pirfenidone and nintedanib was a huge step forward in the treatment of IPF patients, these medications only slow the disease progression and do not markedly improve the quality of life. In addition, they have numerous side effects, which limit their clinical use.[8-11] Thus, there is a high unmet clinical need for new IPF therapeutics.

Idiopathic pulmonary fibrosis pathogenesis Despite decades of research unraveling complexity of the mechanisms underlying IPF pathogenesis, the etiology of the disease remains elusive. The current view on IPF pathobiology brings genetically unstable / aging alveolar epithelial cells into focus. Following multiple injuries, these cells undergo either apoptosis or activation ultimately leading to mesenchymal cell proliferation and deposition of extracellular matrix (ECM) proteins in the lung.[12] These processes are accompanied by accumulation of multiple types of inflammatory cells, including monocyte-derived macrophages, dendritic cells, B cells, and T cells, in the fibrotic lung tissue, and finally formation of an irreversible scar. Thus, identification of the signaling pathways driving miscommunication between multiple cell types and the role of ECM in these processes, all in the context of genetic mutations and epigenetic alterations, is central to unraveling the long-lasting mystery of IPF. In the last years, changes in metabolic pathways, in particular those associated with lipid metabolism, have been linked with the pathogenesis of IPF.[13-15] In addition, strategies focusing on the correction of fatty acid (FA) profiles brought advances in understanding tissue scarring and contributed to the transition of new molecules into clinical development. This review highlights the role of FAs and their metabolites in IPF and provides evidence for therapeutic potential of lipidome manipulations in the treatment of this disease.

Fatty acid biology FAs are a large group of molecules with a basic structure that contains a carboxylic acid group attached to a chain of carbon and hydrogen atoms. FAs can be either saturated or unsaturated. Saturated FAs have only single bonds between carbons (eg, stearic acid, palmitic acid, arachidic acid), whereas unsaturated FAs contain 1 or more double bonds between carbons (eg, linoleic acid, oleic acid, arachidonic acid [AA]), and can be either *cis* or *trans* isomers. Based on the length of the carbon chain, FAs can be divided into 4 groups: short-chain fatty acids (SCFAs) with the chain of 5 or fewer carbons (eg, butyric acid), medium-chain fatty acids (MCFAs) with the carbon tail of 6 to 12 carbons (eg, caprylic acid), long-chain fatty acids with the carbon chain of 13 to 21 carbons (eg, palmitic acid, stearic acid, or arachidic acid), and very long chain fatty acids with the carbon tail of 22 or more carbons (eg, lignoceric acid).[16]

FAs serve as an energy source and building blocks of cell membranes. In addition, FA-derived lipid mediators regulate a plethora of biological processes including signal transduction, cell cycle regulation, apoptosis, and cell differentiation.[16] FAs can either be produced in the cytoplasm from the tricarboxylic acid cycle intermediate, citrate, or taken up from the extracellular space via the FA transporter, CD36 protein.[17] Once entering the intracellular pool, FAs can be esterified with glycerol or sterol backbones and stored in the form of triglycerides (TGs) in lipid droplets or utilized for energy production in β-oxidation. In β-oxidation, FAs attached to coenzyme A (CoA) are transported via carnitine shuttle into the mitochondria, where their intermediates are oxidized to generate adenosine triphosphate.

Among biologically active fatty acids, AA and its metabolites are the best characterized ones. AA is a 20 carbon-chain FA with 4 methylene-interrupted *cis* double bonds and a biochemical nomenclature of all-*cis*-5,8,11,14-eicosatetraenoic acid. AA can be either obtained from food or synthetized in vivo from linoleic acid. AA incorporated into cellular phospholipids ensures flexibility, fluidity, and permeability of biological membranes and thus influences the function of membrane proteins and plays a fundamental role in the maintenance of cell and organelle integrity. Following release from biological membranes by phospholipase A2, AA is rapidly converted into active metabolites known as prostaglandins (PGs), prostacyclins, thromboxanes (TXs), leukotrienes (LTs), lipoxins, and epoxyeicosatrienoic acids (EETs). The physiological activities of these metabolites are widespread and diverse. They are involved in the regulation of the inflammatory responses, pain perception, hemostasis, and cell proliferation. In addition, PGs and prostacyclins are known as strong vasodilators, TXs as potent vasoconstrictors, LTs as bronchoconstrictors, and lipoxins as effective anti-inflammatory and pro-resolving agents.[18,19]

Altered profiles of fatty acids, their derivatives, and metabolites in lung fibrosis Changes in lipidome have been reported in lung tissue, plasma / serum, bronchoalveolar lavage fluid (BALF) of IPF patients, and preclinical models of pulmonary fibrosis.[20] In the lungs of IPF patients, elevated levels of caproate, caprylate, myristate, and palmitoleate and decreased levels of stearic acid, carnitine, and medium-chain acyl-carnitines have been determined.[13,21,22] These changes imply a decrease in β-oxidation in fibrotic lungs. Indeed, reduced β-oxidation was described in alveolar epithelial

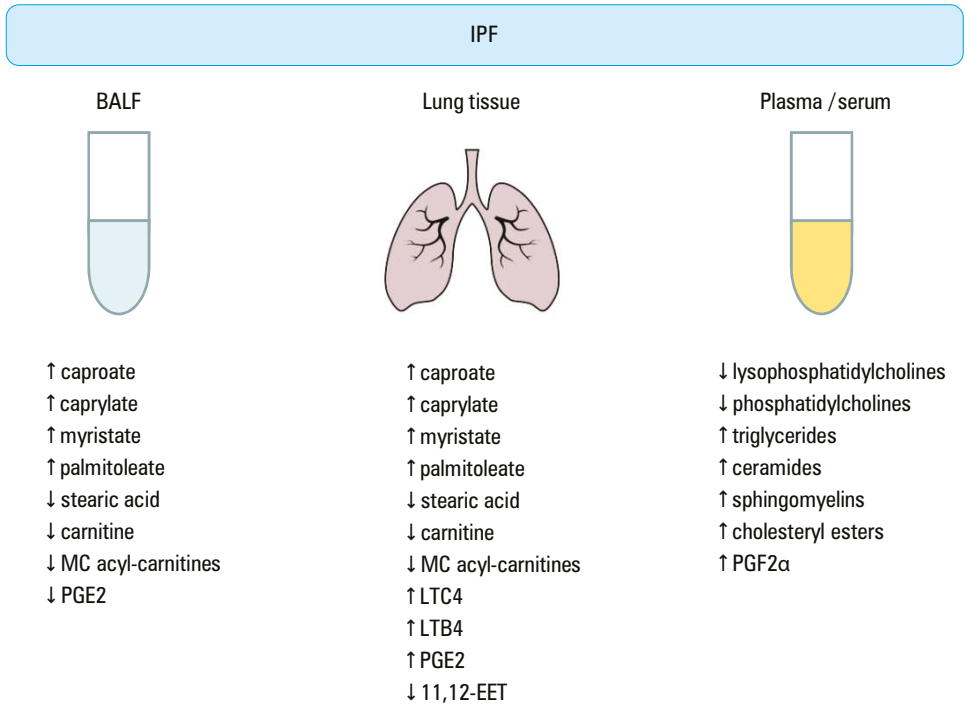

FIGURE 1 Fatty acids, their derivatives, and metabolites in the lung tissue and biological fluids of IPF patients
Abbreviations: BALF, bronchoalveolar lavage fluid; EET, epoxyeicosatrienoic acid; IPF, idiopathic pulmonary fibrosis; LT, leukotriene; MC, medium-chain; PG, prostaglandin

cells of IPF patients. Interestingly, an opposite phenomenon was reported in alveolar macrophages, suggesting that these cells undergo metabolic reprogramming during fibrogenesis, and they support maladaptive remodeling of the lung tissue using FAs as an energy source.[23] In line with these observations, changes in the FA composition of surfactants in IPF patients were described, partially explaining impaired gas exchange in these individuals.[24] In the serum of IPF patients, cholesteryl esters were the most markedly upregulated metabolites.[25] Furthermore, higher levels of TGs and phosphatidylcholines were determined in the IPF progressors, as compared with stable IPF patients. These findings are supported by previous reports highlighting increased plasma levels of glycerophospholipids, glycerolipids, sterol lipids, and sphingolipids in IPF patients in comparison with controls.[20] Building on these observations, Lyu et al[26] identified 5 FA metabolism-related genes (γ-glutamyltransferase 5, acyl-CoA oxidase, leukotriene-B4 Ω-hydroxylase, 3-hydroxyacyl-CoA dehydratase 4, and ornithine decarboxylase) associated with IPF survival. Accordingly, the FA metabolism-related gene expression signature was proposed as a biomarker for predicting IPF clinical outcome.[26] Moreover, an imbalance in the production of AA metabolites has been described in IPF, with increased levels of LTC4, LTB4, and PGE2, and reduced levels of 11,12-EET in the lung tissue,[27,28] decreased amounts of PGE2 in BALF,[29] and elevated levels of PGF2α in the plasma. The increased levels of PGF2α in the plasma of IPF patients negatively correlated with forced expiratory volume, FVC, diffusion lung capacity for carbon monoxide and 6-minute walk distance. In addition, IPF patients with high PGF2α levels had increased risk of mortality, which strongly suggested a detrimental role of this eicosanoid in the disease progression.[30] Finally, areas of increased expression of 15-prostaglandin dehydrogenase, an enzyme responsible for the inactivation of PGs, have been reported in IPF-affected lungs[31] (FIGURE 1). Our unpublished data demonstrated massive accumulation of lipids / lipid-loaded cells in semifibrotic regions of IPF-affected lungs, suggesting contribution of these agents to tissue scarring processes (FIGURE 2).

Lipid perturbations, with increased FA, phosphatidylcholine, and phosphatidylethanolamine levels were also reported in the lungs of bleomycin- or silica-treated mice. Weckerle et al[32] described upregulation of FA acyl-carnitines and downregulation of triacylglycerols, as well as enhanced β-oxidation in the lungs of bleomycin-treated animals irrespective of their age. These changes seem to reflect high-energy demand of the fibrotic tissue associated with increased cell proliferation, production of ECM proteins, and inflammation. In silicosis, PGD2 and TXA2 were found to be significantly upregulated.[33]

Mechanistic insights into the role of lipid mediators in lung fibrosis

Fatty acids Lipids comprise diverse classes of molecules, which play critical roles in the lungs as structural components, energy storage, pulmonary surfactant constituents, and signaling mediators.[34] Their potential role in IPF pathogenesis remains, however, unclear.

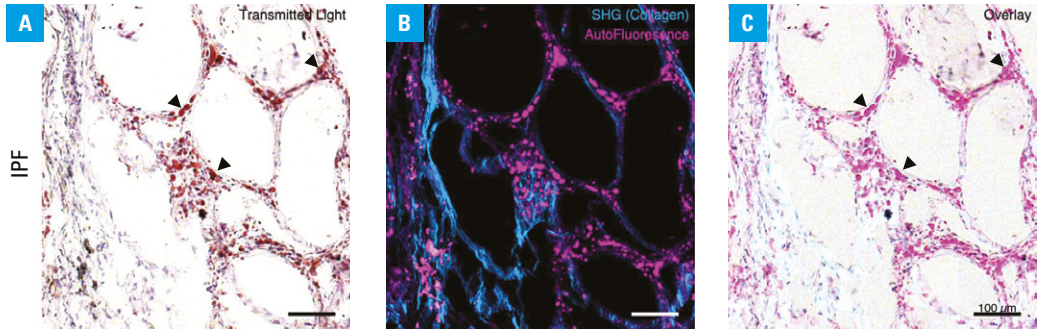

FIGURE 2 Lipids/lipid-loaded cells in semifibrotic regions in an IPF-affected lung; **A** – overlay scan of confocal transmitted lasers (670 nm, 540 nm, and 476 nm). Lipids were detected using Sudan IV dye (reddish color indicated by arrow heads); **B** – second harmonic generation (SHG) and autofluorescence (500–700 nm), derived from 2-photon excitation at 860 nm. Collagen fibers were detected using SHG (cyan spots). Autofluorescence is depicted in magenta. **C** – overlay of **A** and **B**. Arrow heads indicate overlap of lipid staining and autofluorescence.
Abbreviations: see **FIGURE 1**

Substantial alterations in the metabolism of FAs have been reported in IPF patients. These changes have been associated with the development of a profibrotic phenotype of alveolar epithelial cells, macrophages, and fibroblasts / myofibroblasts contributing to fibrogenesis in multiple ways.[35,36] Firstly, accumulation of TGs in the form of lipid droplets in alveolar epithelial cells was found to induce endoplasmic reticulum (ER) stress.[37] Accordingly, exposure of alveolar epithelial cells to palmitic acid triggered ER stress and cell death. These effects were diminished following genetic ablation or pharmacologic inhibition of CD36. In line with these findings, increased ER stress and aggravated lung fibrosis were observed in bleomycin-exposed mice, fed a high-fat diet rich in palmitic acid.[22] Interestingly, ER stress was not only described in alveolar epithelium of mice but also in humans with IPF, and it was associated with maladaptive remodeling of the lung tissue.[38] Secondly, high rates of lipid peroxidation, a process which mainly affects polyunsaturated fatty acids (PUFAs), were reported to produce oxidative stress, cell damage, and inflammation in experimental lung fibrosis.[39,40] Furthermore, elevated levels of lipid peroxides and their protein adducts were measured in the lung tissue and BALF of IPF patients.[41] Consequently, restoration of lipid peroxide balance and suppression of oxidative stress, for example, by overexpression of carnitine palmityl transferase 1A, alleviated pulmonary fibrosis in the rat bleomycin model.[42] Thirdly, Sunaga et al[43] reported that exposure of alveolar epithelial cells to palmitic acid induced transforming growth factor (TGF)-β1 expression and may lead to apoptosis of these cells. Interestingly, opposite results were reported for stearic acid and butyrate. Kim et al[21] demonstrated that stearic acid decreases TGF-β1–induced α-smooth muscle actin, collagen I expression, and reactive oxygen species (ROS) production in fibroblasts, inhibits TGF-β1–triggered epithelial mesenchymal transition (EMT) in the epithelial cells, and reduces hydroxyproline levels in the mouse bleomycin model. Also, Lee et al[44] showed that butyrate reduces TGF-β1–triggered expression of profibrotic factors in the fibroblasts and linked these changes with stabilization of the mitochondrial function. Hence, application of butyrate attenuated bleomycin-induced lung fibrosis in mice and rats.[45] Finally, FAs were found to stimulate macrophage polarization into M2 phenotype, which is known to propagate profibrotic processes.[36]

These findings prompt repurposing / development of agents regulating lipidome profiles for the treatment of pulmonary fibrosis. In this respect, PBI-4050, a synthetic analogue of a MCFA that displays agonist and antagonist activity toward the G-protein coupled receptors GPR40 and GPR84, respectively, attenuated lung fibrosis in the mouse bleomycin model.[46] This agent was already evaluated in a 12-week, open-label, phase 2 clinical study of IPF and demonstrated no safety concerns when used alone or in combination with nintedanib or pirfenidone. Furthermore, the stability of FVC between baseline and week 12 was encouraging for PBI-4050 alone and in combination with nintedanib.[47] Metformin, an activator of adenosine monophosphate–activated protein kinase α and an inhibitor of lipid synthesis, accelerated resolution of lung fibrosis upon bleomycin administration in mice,[48,49] and fenofibrate and rosiglitazone, 2 drugs known to decrease circulating lipid levels, reduced bleomycin-induced lung fibrosis in rats.[50] Valproic acid, known from the treatment of epilepsy and mental disorders, reduced EMT and pulmonary fibrosis in the murine bleomycin model,[51] and α-lipoic acid, a SCFA approved in Germany for the treatment of diabetic neuropathy, reduced oxidative stress and collagen levels in the bleomycin and silica models of lung fibrosis, in rats and mice, respectively. In the silica model, α-lipoic acid also reduced hyperglycemia.[52] Finally, treatment of rats with methyl palmitate attenuated silica-induced lung inflammation and fibrosis. Mechanistically, it reduced lactate dehydrogenase and glutathione activity, diminished overproduction of pulmonary nitrite / nitrate and malondialdehyde content, and increased superoxide dismutase activity in

the lung tissue. These observations suggest that methyl palmitate may counteract the inflammatory and fibrotic processes by decreasing ROS generation in the injured organ.[53]

Taken together, there is strong preclinical evidence of therapeutic power of correcting FA profiles in lung fibrosis. Further efforts should focus on the role of various FAs in the lung hemostasis and diseases and translation of basic research findings into clinical practice.

Arachidonic acid metabolites As far as AA metabolites are concerned, it was demonstrated that LTs exert proinflammatory effects and can promote fibroblast migration, proliferation, and the production of ECM,[27] whereas PGE2 can support the survival of alveolar epithelial cells, reduce fibroblasts proliferation and ECM expression, as well as increase the sensitivity of fibroblasts / myofibroblasts to apoptosis.[1] In addition, treprostinil, a synthetic prostacyclin analogue, prevented PDGF-BB– and TGF-β1–induced human lung fibroblast proliferation and deposition of ECM proteins.[54] These antifibrotic effects were also observed following administration of treprostinil prodrug (hexadecyl-treprostinil), in a therapeutic dosing paradigm, to the lungs of bleomycin-treated rats.[55] Noteworthy, treprostinil inhalations are approved in the United States, Argentina, and Israel for the treatment of World Health Organization (WHO) group 1, and in the United States for WHO group 3 of pulmonary hypertension (PH).[56] The antifibrotic effects were also observed for another prostacyclin analogue, iloprost, which decreased expression of proinflammatory and profibrotic cytokines (tumor necrosis factor-α, interleukin-6, and TGF-β1) and increased expression of antifibrotic mediators (interferon-γ and CXCL10) in the mouse bleomycin model.[57] Together, these findings implicate that overproduction of LTs and PGE2 deficiency promote and sustain lung fibrosis and suggest that strategies aiming to elevate PGE2 / prostacyclin levels may offer an attractive therapeutic option for IPF. Hence, a synthetic PGE2 analogue (16,16-dimethyl-PGE2) attenuated bleomycin-induced lung fibrosis in mice.[58] Similarly, local pulmonary delivery of PGE2 containing liposomes or PGE2 nanostructured lipid carriers protected mice against bleomycin-induced inflammation, weight loss, fibrosis, and mortality. Of note, PGE2 containing liposomes and PGE2 nanostructured lipid carriers were delivered during the acute phase of lung injury triggered by bleomycin.[59,60] Strategies aiming to inhibit PG or EET metabolism by SW03329 (a small-molecule inhibitor of 15-hydroxyprostaglandin dehydrogenase) or TPPU (an inhibitor of soluble epoxide hydrolase), respectively, also turned out to have beneficial effects on lung inflammation and fibrosis in the mouse bleomycin model.[28,31,61] Likewise, attenuation of lung fibrosis, with diminished accumulation of collagen in the affected organ and improved survival, was observed following repeated administration of ONO-1301, a long-acting prostacyclin agonist with TX synthase inhibitory activity, to bleomycin-treated mice.[62] Significant alleviation of silica-induced pulmonary inflammation and fibrosis was also observed upon administration of ramatroban, a clinical antagonist of both PGD2 and TXA2 receptors.[33]

In contrast with these findings, PGE2 worsened pulmonary fibrosis induced by adenoviral overexpression of active TGF-β1 (AdTGF-β1) in mice, and failed to attenuate *Streptococcus pneumoniae*–exacerbated lung fibrosis in the same animal model.[63] In addition, it had poor therapeutic effect when administered 2 weeks after instillation of bleomycin to mice.[64] These observations suggest that the role of PGE2 as the pro- and / or antifibrotic mediator depends on the experimental model employed, therapeutic window, and a secondary hit. In this regard, the bleomycin model of lung fibrosis triggers acute lung injury, which is followed by a fibrosing-healing phase, whereas AdTGF-β1–induced fibrosis lacks the phase of acute lung injury and thus more closely reflects human IPF. Therefore, differences between lung fibrosis models and treatment modalities have to be carefully evaluated before making general conclusions about therapeutic potential of molecules that are supposed to enter clinical development programs.

Regarding strategies focusing on the impairment of LT synthesis, inhibition of LTB4 biosynthesis by histone deacetylase inhibitors, such as suberanilohydroxamic acid or its analogue 4-(dimethylamino)-N-[7-(hydroxyamino)-7-oxoheptyl]benzamide diminished lung inflammation and fibrosis following bleomycin administration.[65] Strikingly, treatment of bleomycin-receiving mice with senolytics decreased LT levels and attenuated lung fibrosis, thus highlighting LT as a part of a senescence-associated secretory phenotype and pointing toward senescence cells as a source of these eicosanoids.[66] These results are in line with previously published reports demonstrating that pharmacological inhibition of LT activity, either by a 5-lipoxygenase inhibitor, zileuton, or a cys-leukotriene receptor antagonist, MK-571, attenuates inflammation and fibrosis in the lungs of bleomycin-treated mice.[67] **TABLE 1** summarizes studies evaluating FAs / FA metabolites as drug candidates in animal models of lung fibrosis.

Taken together, an accumulating body of evidence suggests that eicosanoids, next to their inflammatory properties, may also control profibrotic processes (**FIGURE 3**). Hence, modulating the eicosanoid levels in the fibrotic lungs may have therapeutic potential. However, it requires further research focusing on their dynamics and causal flows in the ligand-receptor interaction networks.

Fatty acid metabolites in clinical studies on idiopathic pulmonary fibrosis The promising results of preclinical studies led to the initiation of several clinical trials evaluating the safety and efficacy

TABLE 1 Summary of studies evaluating fatty acids/fatty acid metabolites as drug candidates in animal models of lung fibrosis

Drug candidate	Model of lung fibrosis	Main findings	Outcome	Reference
PBI-4050	Mouse, bleomycin	Reduced fibrotic lesion score	Favorable	[46]
Butyrate	Rat or mouse, bleomycin	Decreased levels of inflammatory mediators and collagen, diminished oxidative stress	Favorable	[45,86]
Valproic acid	Mouse, bleomycin	Decreased EMT and fibrotic lesion score	Favorable	[51]
α-Lipoic acid	Rat, bleomycin	Decreased oxidative stress, collagen levels, and ameliorated MMP-1/TIMP-1 ratio	Favorable	[87]
Methyl palmitate	Rat, silica	Reduced lactate dehydrogenase and glutathione activity, diminished overproduction of nitrite/nitrate and malondialdehyde, increased SOD activity	Favorable	[53]
Treprostinil prodrug	Rat, bleomycin	Decreased collagen content and fibrotic lesion score	Favorable	[55]
PGE2	Mouse, bleomycin	Decreased inflammation, fibrosis, and mortality	Favorable	[58-60]
PGE2	Mouse, AdTGF-β1	Increased fibrosis and expression of collagen and α-SMA in alveolar type II cells isolated from affected mice	Harmful	[63]
PGE2	Mouse, AdTGF-β1 + *S. pneumoniae*	No change in comparison to animals treated with AdTGF-β1 only (see above)	No change	[63]
PGE2	Mouse, bleomycin	No change in fibrotic lesion score and collagen content	No change	[64]
ONO-1301	Mouse, bleomycin	Decreased inflammation and fibrosis, improved survival	Favorable	[62]
Iloprost	Mouse, bleomycin	Decreased expression of proinflammatory and profibrotic cytokines, increased expression of antifibrotic mediators	Favorable	[57]
Iloprost	Mouse, bleomycin	No change in the number of inflammatory cells in the lung and collagen content	No change	[64]

Abbreviations: AdTGF-β1, adenoviral overexpression of active TGF-β1; α-SMA, α-smooth muscle actin; EMT, epithelial-mesenchymal transition; MMP-1, matrix metalloproteinase-1; SOD, superoxide dismutase; TGF-β1, transforming growth factor-β1; TIMP-1, tissue inhibitors of metalloproteinase-1; others, see FIGURE 1

of FAs / FA metabolites in IPF. These include: a 12-week, phase 2, single-arm, open-label study exploring safety, efficacy, and pharmacokinetics of oral PBI-4050 in IPF patients (NCT02538536)[47]; a 12-week, double-blind, multicenter, trial of inhaled iloprost in IPF patients with PH[68]; and a phase 3, double-blind, multicenter, placebo--controlled trial of inhaled treprostinil in patients with ILD and associated PH (PH-ILD) (INCREASE; NCT02630316).[69]

Daily oral doses of PBI-4050 alone and in combination with approved antifibrotics demonstrated no safety concerns. Furthermore, the stability of FVC between baseline and week 12 looked promising for PBI-4050 alone and in combination with nintedanib.[47] Inhaled iloprost was well--tolerated by IPF patients but it did not meet secondary efficacy end points.[68] Pulmonary delivery of treprostinil was associated with a reduction in the N-terminal pro–B-type natriuretic peptide levels, improvement in the 6-minute walk distance, lower risk of clinical worsening, and fewer exacerbations of underlying lung disease, over the 16-week treatment period. Strikingly, these differences were most evident in patients with idiopathic interstitial pneumonia, particularly IPF, suggesting that inhaled treprostinil might be a promising therapy for IPF that warrants further investigation.[69] Indeed, the prospectively designed double-blind, placebo-controlled, 52-week, TETON 1 and 2 studies (NCT04708782, NCT05255991) will evaluate the safety and efficacy of inhaled treprostinil for treatment of IPF alone, irrespective of the antifibrotic therapy and the presence of PH. The primary efficacy end point of TETON 1 and 2 studies is a change in the absolute FVC from baseline to week 52. The TETON clinical program is the first study evaluating inhaled therapy for IPF. This local drug delivery can provide a more targeted and efficient drug supply option than systemic administration, and thus may confer additional benefits with potentially fewer adverse effects. Future will show whether treprostinil has a chance to enter the clinic.[70]

While the potential of FAs and their derivatives in the treatment of IPF only begins to be appreciated by internal medicine, other medical specialties had already understood their therapeutic power. For instance, SCFA profiles were found to characterize disease severity in the patients with inflammatory bowel diseases,[71] and dietary or pure ω/n-3 PUFAs were shown to exert protective effects against cardiovascular diseases.[72-75] Interestingly, many IPF patients suffer from comorbidities, such as hypertension, obesity, hypercholesterolemia, cardiovascular disease, gastroesophageal reflux disease, or diabetes. In addition, the presence of comorbidities in IPF patients is associated with their worse survival, and this is not only observed in naïve patients but also in those receiving antifibrotic therapy.[76,77] Based on these findings, it is worth speculating that treatment of comorbidities may have a clinically significant impact on the overall outcome that is meaningful for IPF patients. This would also apply to the treatment of cardiovascular disease with dietary or pure ω/n-3 PUFAs in patients with IPF.[72-75] Interestingly, similar interventions with dietary ω/n-3 PUFAs constituting, for example, fish oil were already found to have protective

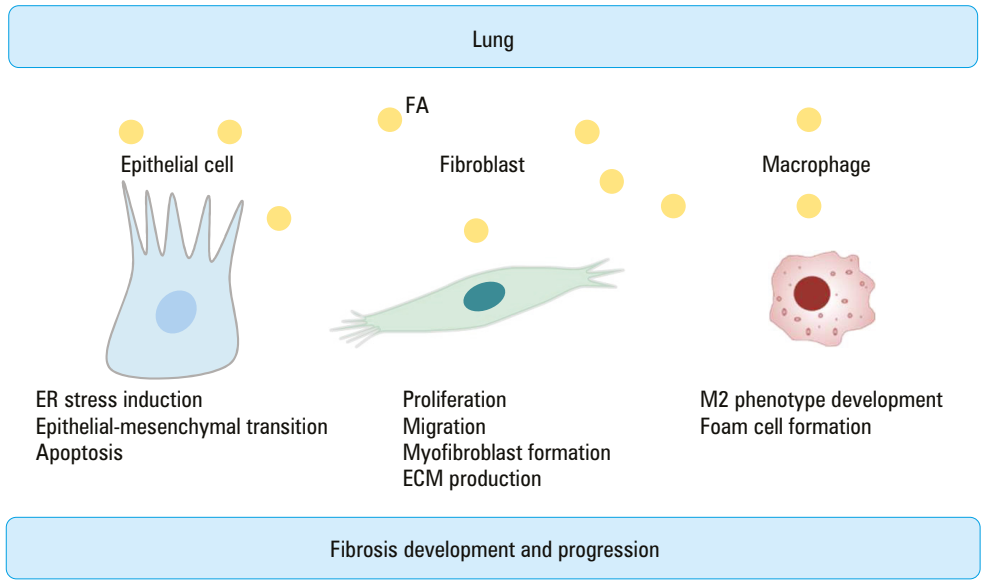

FIGURE 3 Fatty acids as inducers of profibrotic activities of lung epithelial cells, fibroblasts and macrophages
Abbreviations: ECM, extracellular matrix; ER, endoplasmatic reticulum; FA, fatty acid; M, macrophage

effects in asthma,[78,79] at least partly mediated via epigenetic mechanisms.[80,81] Given promising results of preclinical studies[82-84] and some epidemiologic data,[85] oral intake of ω/n-3 PUFA could exert beneficial effects in lung fibrosis as well.

Conclusions Recognition of the emerging role of lipids in the maintenance of lung homeostasis has opened exciting avenues for the development of new therapeutic strategies. FAs and their metabolites are recognized as potent mediators of proinflammatory and profibrotic responses that control phenotypic changes of different lung cell populations during maladaptive remodeling of the lung tissue. The mechanistic insights into the role of lipids in the pathobiology of lung fibrosis are supported by a number of studies demonstrating therapeutic potential of the strategies targeting FAs / FA metabolites in preclinical models of lung injury. Most importantly, the first clinical trial addressing the efficacy of inhaled treprostinil in IPF patients is underway to definitively confirm the intriguing hypothesis of the causative role of FA and their mediators in lung fibrosis. If this clinical study turns out to be successful, it will offer a much needed treatment option for this vulnerable group of patients. Moreover, the design of clinical trials stratified by specific comorbidities, known to be prevalent in IPF patients, may lead to a better understanding of true treatment effects and to better overall outcomes.

ARTICLE INFORMATION

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