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Review Article

A brief review on Pleiotropic effects of Pirfenidone - novel and ongoing outcomes

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ABSTRACT: Presently, there is a huge hype and excitement in the field of synthetic biologics and engineering regarding growing cases of their implication in various fields including health care systems. Furthermore, despite its large body of suggestive and fascinating accomplishments, the synthetic area is always been subject of much more prejudice and debate. However, over a couple of years, the generation of researchers had tested one of such disease modifying compound Pirfenidone (Esbriet®), to unleash their potential in the different disciplines of interventional pharmacology and therapy. In this pipeline, depending on its success of multiple missions, in context to advancing the therapy for different diseases, it became the first prescribed medicine to treat the people with one characteristic lung disorder called idiopathic pulmonary fibrosis (IPF). This review discusses the different therapeutic strategies beyond its well-known anti-fibrotic activity in several well-characterized animals, cell-based and human models and also regarding facts of Pirfenidone (PFD) as anti-inflammatory, anti-fibrinogenic, anti-oxidants including in the treatment of diabetic neuropathy, liver cirrhosis etc. This review also contains current investigations, focusing mainly on the novel findings and their outcomes in improving the quality of life of patients with different conditions and also suggests their implication on the basis of fundamental existential evidences to break the major impediment in transforming this disease-modifying drug into a personalized medicine.

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INTRODUCTION

The intension of this review is to draw an outline on the current knowledge and recent findings about the use of PFD in several treatment modalities such as fibrosis of the lung, kidney, heart and some inflammatory, oxidative stresses including their use in the treatment of diabetic nephropathy, liver cirrhosis etc [1] and also suggests that use of which to improve the quality of life (QOL) of the patients with multiple diseases and comorbid conditions. QOL is itself a complex concept, deals with various functional limitations in mental, physical and social life etc. It reflects many parameters such as symptoms like pain, dyspnea, fatigue, edema etc. QOL is also influenced by many other factors such as age, gender, marital status, education, socioeconomic status, lifestyle habits such as smoking, drinking, personal views on life, desires, expectations, the correspondence between activities of daily living and the patient's wishes [2]. PFD is a low molecular weight, small non-peptide molecule of 185.2 Dalton with the chemical name of 5-methyl-1-phenyl-2-(1H)-pyridon [3]. PFD is the first drug in its pharmacological class and has been placed in the WHOCC (World Health Organization Collaborating Centers) ATC (Anatomical Therapeutic Chemical Classification) drug class of other immune-suppressants' and has been designated an ATC drug code of L04AX05 [4]. PFD was first approved for the treatment of IPF in Japan in 2008. PFD has since been approved for the indication of 'mild to moderate idiopathic pulmonary fibrosis' in the EU (February 2011) and Canada (October 2012), and for the treatment of idiopathic pulmonary fibrosis in the US (October 2014) and Switzerland (September 2015) [4].

The major impediment in transforming any drug into personalized medicine is to redefine the common diseases. We often define diseases by the characteristic signs and symptoms. But as we move into the new millennium, the biomedical sciences are now entering into the new phase of their development. Paradoxically, these personalized medicines are also passing through a phase of increasing uncertainty. On the contrary PFD was initially identified for its anti-inflammatory activity in animal model and thus evaluated as an anti-inflammatory drug. Later discoveries found that they also possess Anti-fibrotic property. The accidental or unexpected discovery of which as an anti-fibrotic, redefined the interest of compound in further researches. Subsequently, PFD has been shown to attenuate fibrosis in numerous animal models, including fibrosis of the lung, liver, heart and kidney [5]. Organ function impairment and destruction of organ architecture is the pathological scaring process in fibrosis. Chronic dysfunction of parenchymal organs such as heart, intestine, kidney, liver, and lung can lead to progressive decline in overall organ function, poor quality of life, and, ultimately early death, thus accounting for an estimated one third of natural deaths worldwide [6].

Especially sarcadosis and pulmonary fibrosis are associated with numerous diffuse parenchymal lung inflammations and diseases. The conditions like atrial fibrillations, chronic heart failure, acute myocardial infarction and cardiac remodeling attributes to cardiac fibrosis [7]. Multiple forms of chronic kidney diseases and impaired kidney functions attributes to renal fibrosis. And non-alcoholic steato-hepatitis, a chronic hepatitis C and B viral infection causes hepatic fibrosis [8]. Much more current researches apart from different fibrotic diseases regarding the use of PFD had also been identified such as use of which for liver cirrhosis, Diabetic nephropathy [9] etc. In this review we discuss the recent outcomes and ongoing experience with PFD investigation in animal, cell- based and clinical trials. Also provides some evidence-based studies to suggest the incorporation of which into treatment algorithm of patients with cardiac failure, kidney failure, liver injury, allergy etc.

Newly identified *In vivo* and *In vitro* targets of PFD

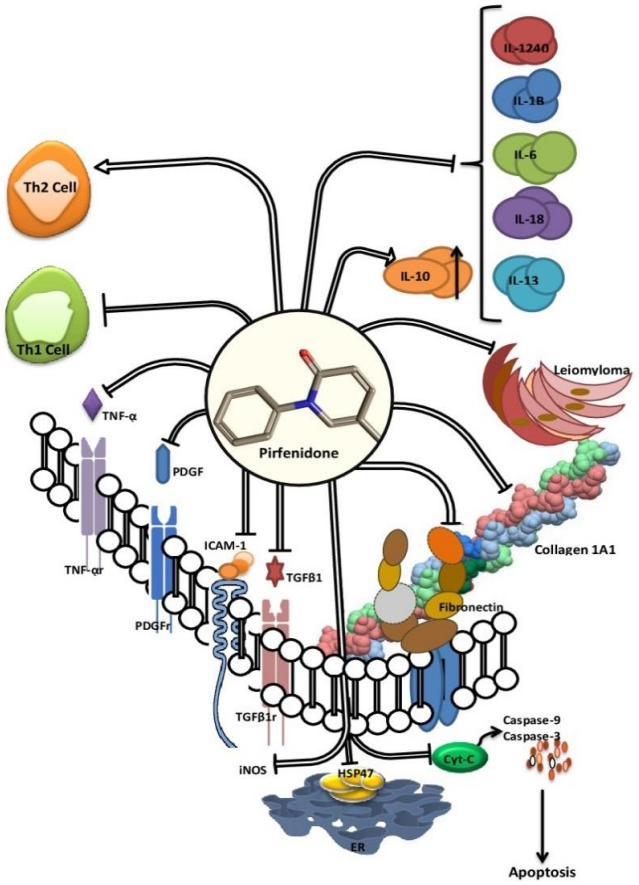

Figure 1: The most prominent being inhibitors of TNF-ά and TGF-β1. However, it has also been showed that PFD has either direct or indirect action on other molecules such as Collagen I, PDGF. IL-6, Il-1β, IL-13, IL-12p40, Fibronectin, HSP447 and ICAM-1

Table 1: -Chemical Characteristics of PFD molecule.

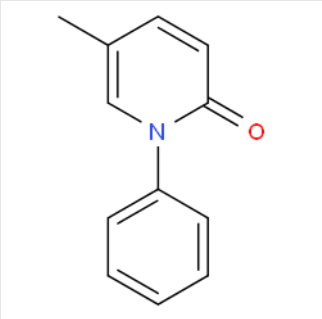

Figure 2: 2-D Structure of PFD

IUPAC Name: 5-methyl-1-phenylpyridin-2-one	
Molecular Formula	**$C_{12}H_{11}NO$**
Molecular weight	**185.22184 [g/mol]**
Exact Mass	185.084064
Mono Isotopic Mass	185.084064
H-Bond Donor	0
H-Bond Acceptor	1
XLogP3-AA	1.9
Rotatable Bond Count	1
Topological polar Surface Area	20.3
Heavy Atom Count	14
Complexity	285
Covalently Bonded Unit Count	1

Structure activity relationship of PFD molecule

As previously described, PFD is now one of the two commercially available, broad spectrum drug of its class belongs to pyridone group. It is a multi-target drug act on TGF-β, TWF-Aβ, PDGF, TNF-α etc. Few studies showed that presence of methyl group on 5 position of PFD causes it to metabolize rapidly into carboxylic acid in *invivo*. Therefore, it becomes necessary to understand the SAR of the drug to modify its property to prevent presystemic metabolism *invivo*. To screen *invitro* De-fibrotic activity, NIH3T3 mouse fibroblast cell line was performed by MTT assay.

1. Addition of trichloro methyl group at 5 position of pyridone ring was more favorable than methyl or chloro group substitution

2. Conjugation of pyridone ring of PFD with aryl ring of flurofenidone (which is an analogue of PFD) did not alters the affectivity of the compound

3. Substitution of derivatives at *p*-position was more superior that *m*-position.

4. Hydrophobic group substitution at 1 or 3 position of pyridone ring would provide better potent anti-fibrotic activity [10,11].

Current Highlights and Evidence based Meta-analysis on PFD

Cell-Based models

In cell-based assays, several activities of PFD which are ideally relevant to its anti-fibrotic activity have reported here, which in addition to its fibrotic mediator's modulation to animal models. The *in vitro* anti fibrotic activities of PFD basically fall into three general categories:

- Reduction of myofibroblast proliferation and fibroblast
- Inhibition of synthesis and deposition of extracellular; and
- Reductions in fibrotic and related bio-markers

Each of these activities is relevant to the attenuation of chronic fibrosis, is consistent with the effects reported for PFD in animal [12].

Anti-fibrotic effects of PFD and Rapamycine in human alveolar epithelial cells and primary IPF fibroblasts;

In the presence or absence of transforming growth factor β1 (TGF−β), PFD and rapamycine was treated with human alveolar epithelial cells (A549) and Primary lung fibroblasts which was taken from the IPF patients. The markers involved with lung fibrosis, smooth muscle actin and collagens, gene expression and extracellular matrix were analyzed. And it was observed that there was significant reduction in fibronectin and tenacsin-c of fibrotic fibroblast and other fibrotic biomarkers. Though the treatment failed to revert the transition pathway of mesenchyme cell from epithelium which was supposed to be activated by the TGF- β. However, it was successful enough to significantly abrogate the fibroblast to myofibroblasts. PFD in combination with rapamycine in fact down the line actually potentiated the inhibitory effect on fibroblast migration in the scratch-wound assay. Hence these novel findings thus indicated the use of PFD in combination with rapamycine widens the probability of fibroblast migration and inhibitory range of fibrogenic markers [13]. Modulatory properties of PFD and Nintedanib on fibroblasts and myofibroblasts in IPF;

This study describes treatment using PFD and Nintedanib (PFD-NTNB) with stromal cells, which was collected and cultured from Positive IPF patients or the control patients [14]. The extent of inhibitory properties of these combinations (PFD-NTNB) were analyzed by measuring the cell proliferation, fibronectin expression (by western analysis, immuno-electron microscope, ultra-structural properties by transmission electron microscope [TEM]), invasion assay and functional properties by collagen gel contraction techniques. It was observed that PFD-NTNB combination shows monotonic inhibitory response and it was said that the inhibitory effect of one enhances the other. Although the level of inhibition is however cell line dependent but both of them were significantly reduced the amount of myofibroblastic cells and smooth muscle actin (α-SMA) appearance. The same was further experimentally confirmed by the specific function assay and thus found it was variable too [15,16].

Inhibition of Leiomyoma Cell Proliferation and Collagen Production:

Leiomyomas is characterized by increased tissue fibrosis and cell proliferation for which an effective and long-term drug therapies are yet not available. Treatment of PFD on collagen expression and cell-proliferations in cultured leiomyoma smooth muscle cell and myometrial smooth muscle cells was examined and effect of the PFD on the same was thus measured using tritiated thymidine incorporation assays. The changes in the actual cell number before and after the treatment were observed. Generation of collagen I and III was assessed by northern blotting techniques. The doses of PFD used in this experiment are 0.01 to 1.0 mg/ml. Study showed dose-dependent inhibitory effect of PFD on Leiomyoma cells and possible cytotoxic effects were measured using tryphan blue exclusion and lactate dehydrogenase assays thus showed no cytotoxicity of PFD at the concentration that inhibited collagen production and cell proliferation [17].

Preclinical studies on PFD using several well characterized animal models:

Effects in *in vivo*-invasive models of cardiac fibrosis

Cardiac fibrosis (CF) develops in response to increased diastolic stiffness, chronic hypertension and increased risk of arrhythmias associated with loss of overall cardiac function [5]. CF is a significant factor in several disorders, including congestive heart failure [18], chronic hypertension [19] which contributes to morbidity and cardiac dysfunction following myocardial infarction [20,21]. In particularly, the Areas of heart in which there are fibrotic tissue compromise overall cardiac function by interfering with coordinated electrical conduction and increase vulnerability to arrhythmias.

At a molecular and cellular level, there are similarities between fibrosis in other organs including persistent myofibroblasts cardiac fibrosis, elevated fibrogenic mediators (PDGF, TGF-b and connective tissue growth factor) and cardiac fibrosis [22].

In Canine model of heart failure-In vivo

Ventricular tachypacing was performed to induce Congestive heart failure (CHF) in dogs which was outfitted with single chambered pacemaker with dietary administration of PFD of 800mg thrice a day for over a 3- week period. The study showed, treatment with PFD significantly reduced the left atrial fibrosis induced CHF by 50% and also reduced the heterogenecity of the left arterial conduction and susceptibility to atrial fibrillation [23].

In Murine Invasive models of heart failure

By transverse aortic constriction (TAC) in mouse models of myocardial Fibrosis and pressure overload was induced and thus found that the left ventricle fibrosis and contractile dysfunction was progressed further from 4 to 8 week after TAC. On contrary, the treatment group prevented the risk of myocardial fibrosis beginning from 4th week after TAC. PFD treatment showed cell-type specific effect on isolated vascular endothelial cells and cardiac fibroblasts from the left ventricles of adult male mice. Study showed significant suppression of collagen 1 and cldn5 expression level, resulting in reduced fibrosis, serum albumin leakage into the interstitial space during the chronic phase of heart failure in TAC hearts and also prevents vascular permeability [24, 25].

Similarly**,** the effect of PFD on hypertensive rat models was also studied by the next set of researchers, to identify their potential to be an anti-hypertensive drug. The hypertension was induced by removal of left kidney with either 1% NaCl in drinking water and subcutaneous injections of deoxy-corticosterone acetate or no further treatment (control rats). Treatment with PFD (0.4% in feed) was instigated after 2 weeks of the surgery and treatment was continued till conclusion of the study at 4th week. PFD treatment attenuated cardiac fibrosis, diastolic stiffness and ventricular myocyte hypertrophy. Cardiac fibrosis and diastolic stiffness were reduced to a level, slightly below those observed in parallel animals at initiation of treatment (week 2). Hence suggesting, PFD may reverse nascent fibrosis in this model. Collectively, these studies demonstrate that PFD can exert anti-fibrotic effects from the contemporary lung fibrosis and investigation also indicated the fibrotic pathways targeted by PFD are not tissue specific [19].

Effect in Invitro models of cardiac fibrosis

Pleiotropic effects of PFD on cardiac fibroblast (CFs) was investigated their potential benefit in cardiac remodeling with this they observed, to which extent the PFD can directly influence the cardiac fibroblast modulation. CFs behaviors are important in cardiac remodeling such as myofibroblast differentiation, proliferation, cytokine secretion and migration. To determine the effect of PFD on fibroblast function, bioassay was performed and isolated fibroblasts from neonatal rats' heart were analyzed. Study thus showed treatment with PFD on CFs showed significant reduction in proliferation, attenuated the fibroblasts of smooth muscle actin expression and collagen contractility. Inhibitory effects of migration could possibly by decreasing ratio of matrix mettaloproteinase-9 to tissue inhibitors of metalloproteinase-1. Furthermore, PFD attenuated both synthesis and secretion of TGF-b1 but showed elevation in the levels of interluikin-10 [26].

Effect in *in vivo*-invasive models of lung fibrosis

Experimental bleomycine models of lung fibrosis was designed to assess the molecular mechanism, clinical safety and efficacy of the drug when administer to treat lung diseases including interstitial pulmonary fibrosis and several models of acute lung injury [27]. These groups of disorders are characterized by deep scarring of the lung tissue, loss of functional alveoli and shortness of breath, thus limiting overall oxygen exchange capacity of the lung [28]. Etiology includes inhalation of gases, vapours, fumes, organic and inorganic dusts, radiation exposure, use of medications (quite often) and development of diseases like silicosis, byssinosis (occurs after exposure to cotton dust), or hypersensitivity pneumonitis, coal worker pneumoconiosis and among others [29].

In Bleomycine induced lung fibrosis-Invivo

Bleomycin is produced by "*Streptomyces verticillus*" which is a chemotherapeutic antibiotic, plays an important role in the treatment of squamous cell carcinoma, lymphoma, germ cell tumor and malignant pleural effusion. It is believed that bleomycin interrupts the cell cycle by acting through the breakage of single and double stranded DNA in tumor cells. This kind of DNA cleavage happens by metal ion chelation and the reaction formed psuedoenzyme with oxygen, leads to production of hydroxide and superoxide free radicles [30]. Over production of reactive oxygen species (ROS) leads to increased pulmonary toxicity by inflammatory response then activation of fibroblast and subsequent fibrosis. This experimental lung fibrosis was first described in dogs [31] and later in murine models [32,33]. Several studies demonstrated the effect of PFD on bleomycin models of pulmonary fibrosis, over the course of 42 days (repeated intravenous administration of bleomycin) followed by oral gavage administration of PFD (30/100 mg/kg in three divided doses) to minimize early lung pathology, edema and reduced accumulation of hydroxyproline. It was thus observed that treatment with PFD normalized pulmonary expression of fibrogenic and pro-inflammatory proteins [34].

In Transplant-induced pulmonary fibrosis-Invasive-Invasive

PFD is also operative in reducing dysfunction in preclinical lung allograft models and post-transplant pulmonary fibrosis. The Prophylactic properties of PFD regimen in transplant models of lung between Lewis rat and Sprague-Dawley was evaluated. And it was observed that treatment with PFD significantly reduced peri-bronchial and interstitial fibrosis, reduced allograft induced rejections, preserved interstitial and alveolar structures and in addition, PFD ameliorated increases in peak airway pressure in the transplanted lung [27]. Rejection of subcutaneous tracheal transplant with the inflammatory cell infiltration, loss of respiratory epithelium, fibrosis and intraluminal granulations observed, when transplanted between BALB/c donor to a C57BL/6 recipient.

The resultant of these fibrotic components of allograft rejection slowly developed and became rampant at 28th day. Prophylactic treatment with PFD for 28 days (0.5% in feed), prevented the loss of epithelial cells, collagen deposition, and intraluminal degranulation. 67% of untreated allografts (complete occlusion of airway with granulation tissue) were compared with only 7% of PFD treated group. Collectively, these studies specify the anti-fibrotic activities of PFD in pulmonary models are not unique to fibrosis that results from a chemical insult of bleomycin [35].

Effect in Invitro models of lung fibrosis

Recently, it has been suggested that lung fibrosis is caused in part by inflammation and chronic oxidative stress. Augmented production of reactive oxygen species (ROS), leads to pro-inflammatory factors activation, oxidation of proteins and DNA lipid peroxidation, has been pragmatic in several *in vitro* lung models; PFD has been shown to diminish these pathological stress states. For example, PFD suppressed chemokine secretion, alveolar macrophage cytokine and Hermansky-Pudlak syndrome (HPS)-1, *in vitro* in a dose-dependent manner. Both *in vitro* and *in vivo*, PFD were found to inhibit the proliferation index of both CD4 and CD8 and the responder frequency of T-cell rearrangement (TCR)-stimulated CD4 cell total proliferation. Additionally, PFD inhibited TCR-induced production of multiple pro-inflammatory chemokine's and cytokines. Remarkably, PFD had no effect on the suppressive properties of naturally occurring regulatory T cells and there was no change in TGF-b production by purified T cells. In addition, the anti-fibrotic properties of PFD may be mediated through inhibition of collagen-specific molecular chaperone or heat shock protein 47 (HSP 47) with a resultant reduction in collagen synthesis in lung fibrosis [36].

Effects in *In vivo-invasive* models of renal fibrosis

Renal fibrosis is an interactive and dynamic process initiated by cellular damage, characterized by mediators of molecular & cellular fibrosis and loss of cellular differentiation phenotypes, are common to fibrosis in other organs including the presence of elevated fibro-genic mediators like TGF-b, PDGF, interleukin (IL)-1b and basic (b) FGF, myofibroblasts, persistent fibroblasts and an imbalance of metalloproteinases and their inhibitors [1,5].

PFD has been shown to significantly diminish renal fibrosis in several pilot studies. This had been established in a several studies, including models driven by partial unilateral ureteral obstruction (UUO), nephrectomy, and Diabetic neuropathy [9]. Limited subsets of these studies are summarized below.

In murine models of renal fibrosis

The area of staining positive for renal fibrosis was reduced by 80% with prophylactic treatment with PFD (1% in feed) with the trends toward improvement of blood urea nitrogen levels and proteinuria as well as creatinine. Similarly, 5/6 th of the nephrectomy model was also evaluated by PFD prophylaxis.

In this study, PFD treatment (0.6–0.9% in feed) prevented 60% of collagen accumulation following nephrectomy and also reduced expression of collagen mRNAs and TGF-b. These effects were concomitant with improved clearance of creatinine.

Overall, PFD showed a significant anti-fibrotic effect in this well-characterized model of chronic kidney disease [37,38].

In unilateral ureteral obstruction (UUO) method

Apart from these two models, there are one more well characterized invasive models through unilateral ureteral obstruction (UUO) method. The study was carried out with two implementations. In the primary implementations, with the continuous ureteral obstruction for over 3 weeks, the fibrosis was developed. Then prophylactic treatment with PFD (0.6-0.9% in feed) showed, 50% reduction in expression of collagen and TGF-b mRNA as well as deposition of collagen to significant levels. In the later implementation of the UUO model, after first week, the obstruction was unconfined and at the time when obstruction was released, the levels of hydroxyproline was noted in parallel animals and despite removal of obstruction, the fibrosis was continued to develop over the next 5 subsequent weeks. Together, these interventions showed that it can ameliorate renal fibrosis, terminate ongoing renal fibrosis and reduce expression of collagen mRNA [37].

In mouse model of human non-alcoholic steato-hepatitis (NASH)

Study was developed with an experimental mouse model of NASH; melanocortin 4 receptor-deficient (MC4R-KO) mice was fed a western diet (WD) or high fat diet to develop a liver condition similar to human NASH, being associated with dyslipidemia, insulin resistance and obesity. Using this model, unique histological structure termed 'hepatic crown-like structures' (hCLS) in the NASH-like liver was discovered, where dead or dying hepatocytes with large lipid droplets were surrounded by macrophages was observed. In hCLS, macrophages are considered to interact with dead parenchymal hepatocytes and fibrogenic cells, thereby accelerating inflammation and fibrosis in the liver. Treatment with PFD remarkably attenuates liver fibrosis in the murine model of human NASH in parallel with significant reduction of and hCLS formation and hepatocyte apoptosis. Study also highlighted the use of which to inhibit fibrogenic responses and TNF-α induced liver injury in *in vivo*, and indeed inhibits directly the TNF-α induced hepatocyte apoptosis in *in vitro* [6].

Effect in models of liver fibrosis

Hepatic fibrosis occurs in patients with chronic liver disease associated with alcoholism, viral hepatitis, and autoimmune liver disease. Hepatic fibrosis is driven by many of the same fibrogenic mediators as in other organs and hepatic stellate cells (HSC), which, like myofibroblasts, are hyper-activated with respect to ECM deposition and production of growth factors [29, 6]. Chronic intoxication of rats with carbon tetrachloride (CCl4) results in hepatic fibrosis and elevated liver enzymes. Treatment with PFD at 200 mg/kg oral administration decreased collagen I mRNA expression and liver fibrosis by 40%.

Markers of liver damage (alanine aminotransferase (ALT), aspartate aminotransferase, total bilirubin and direct bilirubin) were significantly reduced in treated animals relative to controls, demonstrating a functional benefit of PFD treatment.

PFD treatment also reduced markers of oxidative stress, including malondialdehyde and nitrites in the liver; these changes were accompanied by reductions in expression of catalase and superoxide dismutase mRNAs [39].

Similarly, Bile duct ligation model (Invasive) and Dimethylnitrosamine (DMN)-induced liver fibrosis is another commonly used experimental model in rats [39,40].

In vitro models of liver fibrosis; PFD at 1000 µM inhibited PDGF-induced HSC proliferation in rat hepatic stellate cells (HSC), without any toxic effects. It also did not affect HSC viability and did not induce apoptosis. The inhibition in cell proliferation was not associated either with the activation of extracellular signal-related kinase (ERK) or variations in PDGF receptor autophosphorylation, but PFD was able to inhibit PDGF-induced protein kinase C activation, type I collagen accumulation, pro-collagen mRNA expression and activation of the NA+H+ exchanger involved in PDGF induced HSC proliferation [40].

PFD was found to be ineffective as a superoxide radical scavenger, in decomposing H_2O_2 and chelating iron in sheep liver microsomes; however, in a deoxyribose degradation assay; PFD was a potent scavenger of hydroxyl radicals, which could be related to its beneficial effects as an anti-oxidant [41].

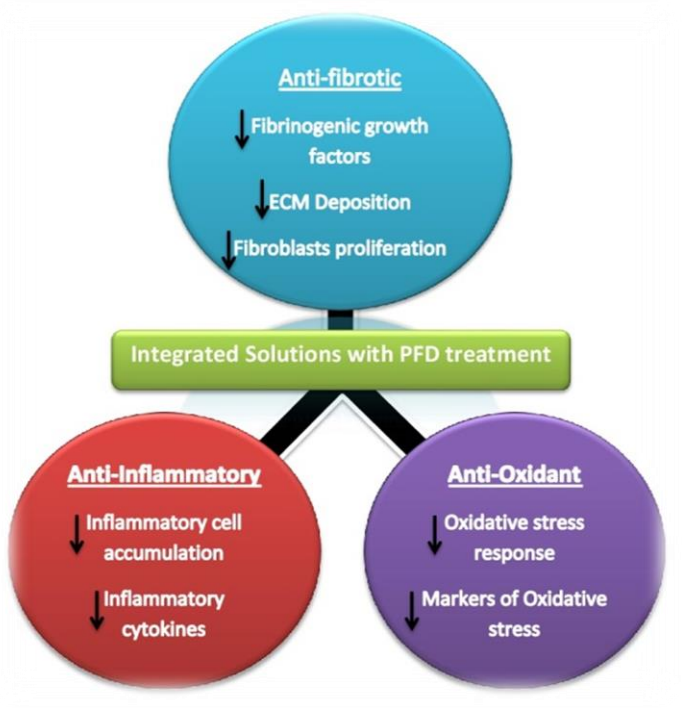

Figure 03: Overview on brief expression of PFD

Table 2: Current and on-going experience with the use of PFD in multiple clinical trials

S. no.	Study designed	Phase	Purpose	Status	References
1	A Bioequivalence Study Comparing Pirfenidone Tablet and Capsule Dosage Forms in Healthy Adult Participants	1	Not available	Completed	[42]
2	Pirfenidone in Treating Young Patients with Neurofibromatosis Type 1 and Plexiform Neurofibromas	1	Prevention	Completed	[43]
3	A Study of Oral Vismodegib in Combination with Pirfenidone in Participants with Idiopathic Pulmonary Fibrosis	1	Treatment	Completed	[44]
4	For Squamous Cell Carcinoma of Lung / Metastatic Lung Cancer / Cancer, Advanced / Lung Cancers / Non-Squamous Non-Small Cell Neoplasm of Lung / Lung Cancer Non-Small Cell Cancer (NSCLC) Treatment	1	Treatment	Recruiting	[45]
5	Graft Versus Host Disease (GVHD) / Obliterative Bronchiolitis Treatment	1	Treatment	Recruiting	[46]
6	Chronic Kidney Disease (CKD) / Fibrosis Treatment	1/2	Treatment	Completed	[47]
7	Diabetic Nephropathies / Diabetes Mellitus (DM) Treatment	1/2	Treatment	Completed	[48]
8	Albinism, Oculocutaneous / Pulmonary Fibrosis / Inborn Errors of Metabolism / Albinism / Platelet Storage Pool Deficiency Treatment	2	Treatment	Completed	[49]
9	Chronic Hepatitis C Virus (HCV) Infection Treatment	2	Treatment	Completed	[50]
10	Hypertrophic Cardiomyopathy Treatment	2	Treatment	Completed	[51]

CONCLUSION

PFD is a multi-purpose broad spectrum, non-peptide, orally bioavailable, small molecule used for the treatment of IPF. Clinical investigations on PFD, based on the models suggested in preclinical studies. These suggested animal models, ideally reflects detailed characteristics of human diseases including progressive tissue scarring, aberrant epithelial repair, and inflammation with the induction of fibrotic foci.

These models are highly consistent, reproducible, easy to perform, access and inexpensive to maintain. Several studies also demonstrated the relationship between improvements in related functional-endpoint and reduction in fibrosis including improved lung function, heart function, renal clearance, reduced risk of arrhythmia, reduction in biomarker of liver damage. From all this, it is strongly evident that PFD is a powerful anti-fibrotic drug as it reduces inflammatory, pro-fibrogenic and reduced oxidative markers.

Overall, the clinical and preclinical profile, clearly demonstrated systemic anti-fibrotic, anti-inflammatory and anti-oxidant effect that would eventually supports its clinical evaluation.

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