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FAT-Free p300 Is Good for Scar-Free Tissue Repair

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ABSTRACT

Fibrosis, the deadly pathological manifestation of an abnormal tissue remodeling in any organ due to excessive collagen deposition, is associated with a wide variety of organ failure-related human diseases. Chronic stress or repeated injury in a particular organ induces abnormal molecular signals that lead to super-activation of matrix protein producing fibroblasts, excessive matrix proteins accumulation, loss of physiological tissue architecture or elasticity, and ultimately leading to organ failure. There is no effective therapy for fibrosis. Factor acetyltransferase p300 (FATp300), a major epigenetic regulator that acetylates specific lysines in histones and transcription factors, is essential for elevated collagen synthesis and the levels of FATp300 are significantly elevated in different fibrotic tissues. Pharmacological inhibition of FAT activity of p300 is associated with decreased collagen synthesis by fibroblasts in tissues and amelioration of organ fibrosis. Therefore, FAT-free p300 is superior for physiological tissue repair and must be exploited as a viable therapeutic target against multi-organ fibrosis. J. Cell. Biochem. 115: 1486–1489, 2014.

KEY WORDS: ORGAN FIBROSIS; EPIGENETICS; p300; FIBROBLASTS; COLLAGENS; TISSUE REPAIR

Healthy tissue architecture and function in every organ is largely controlled by extracellular matrix composed of different matrix proteins like collagens. However, unnecessary synthesis and deposition of matrix proteins by hyperactive fibroblasts in injured or harassed tissues lead to the development of fibrosis—the major cause of organ failure-related mortality in humans [Hayden, 2011]. Although there have been great strides in advancing fibrosis research in the past several decades, there has yet to be an effective and safe therapy, underlining the urgency to illuminate novel targets and subsequently develop anti-fibrotic therapeutics.

Numerous studies over decades explicitly establish the pivotal role of transforming growth factor-beta (TGF-β) during physiological wound healing as well as pathological fibrogenesis in heart, lung, kidney, liver, skin, and bone marrow [Border and Noble, 1994]. However, systemic blocking of TGF-β signaling as a therapeutic approach for fibrosis is uncertain because TGF-β also controls cellular growth, tumorigenesis as well as immunity [Wahl, 1994]. Therefore, targeting extreme downstream factor(s) essential for amplification of profibrotic signaling in hyperactive fibroblasts to normalize matrix protein synthesis will be a viable approach to control fibrogenesis. In the late 1990s, several studies recognized the significance of epigenetics, a method of alteration of gene expression without affecting DNA sequences, in initiation and progression of different human diseases [Arrowsmith et al., 2012], and became a hot topic of biomedical research. In order to understand the epigenetic regulation of fibrogenesis, a novel project on the role of factor acetyltransferase p300 (FATp300), an epigenetic regulator, in extracellular matrix protein synthesis was initiated 15 years ago and demonstrated that an elevated level of FATp300 in fibroblasts leads to an increased synthesis of collagen, the major extracellular matrix protein in most fibrotic tissues [Ghosh et al., 2000]. Importantly, FATp300 is essential for sensitizing the fibroblasts to profibrotic cytokine TGF-β and stimulation of collagen synthesis via physical and functional interaction with Smad signaling molecules along with chromatin histone and transcription factor modification by acetylation in the transcription initiation complex formed on collagen gene promoter [Ghosh et al., 2000, 2001, 2004, 2009, 2013; Bhattacharyya et al., 2005] (Fig. 1). The stimulatory effect of FATp300 on collagen synthesis is specific because first, FATp300 interacting adenovirus E1A protein, but not E1A mutant which cannot interact with FATp300, completely blocks elevated collagen synthesis and E1A fails to blunt TGF-β-induced collagen synthesis in the presence of excess FATp300; second, FAT-free p300 fails to augment collagen synthesis in response to profibrotic signal [Ghosh et al., 2000]; and third, profibrotic signal fails to induce matrix protein synthesis in FATp300-deficient fibroblasts or FAT inhibitor-treated fibroblasts even in the presence of FATp300-related CREB-binding protein (CBP) [Ghosh et al., 2000, 2013; Bhattacharyya et al., 2005]. Interestingly, profibrotic signal

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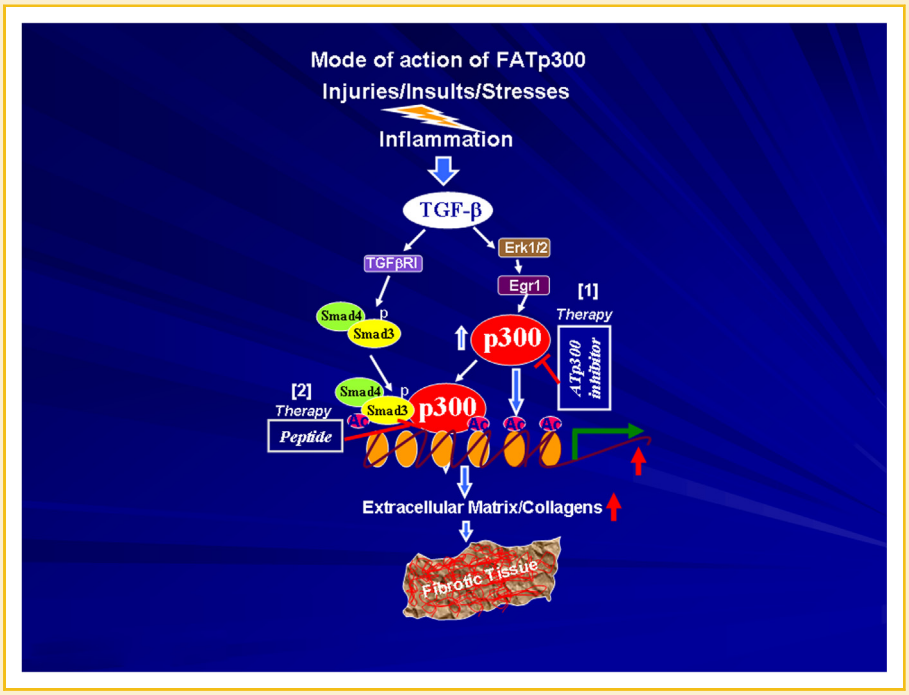

Fig. 1. Mode of action of FATp300 in profibrogenic pathway: FATp300 as a potential target for fibrosis therapy. Vascular injury (reparative) or stress (reactive)-induced sustained/repetitive inflammation causes imbalance of pro- and anti-fibrotic cytokines that leads to enhanced profibrotic signaling. Profibrotic TGF-β signaling significantly augments the levels of FATp300 via activation of ERK-MAPK-EGR1 pathway and activates Smad pathway via phosphorylation of Smad2/3 by TGFβRI kinase. Acetylation of histones and transcription factors in collagen gene regulatory elements by FAT activity of p300 as well as physical interaction of FATp300 with activated Smads contribute to excess synthesis of major matrix protein collagen that leads to fibrosis. Inhibition of FAT activity of p300 with small molecule inhibitors [1] or disruption of physical interaction of FATp300 with Smads using small peptide [2] will be an ideal approach to control fibrogenesis.

stimulates the expression of FATp300, and increased FAT activity of p300 acetylates histones on collagen gene promoter to create a relaxed chromatin suitable for transcription, and thus amplify profibrotic responses and fibrogenesis [Ghosh et al., 2000, 2009, 2013; Bhattacharyya et al., 2005]. An increased acetylation of a specific lysine residue in profibrotic transcription factor by FATp300 and increased activity of acetylated transcription factor may also contribute to increased expression of matrix protein collagen.

FATp300 not only augments profibrotic matrix protein synthesis [Ghosh et al., 2000]; several studies recognized the involvement of FATp300 in different antifibrotic pathways including antifibrotic action of IFN-γ-activated Stat1α, anti-diabetic drug-activated PPAR-γ, growth regulator p53, dibutyryl-cyclic AMP (Bt_2cAMP), and AMP-activated protein kinase (AMPK) [Ghosh et al., 2001, 2004, 2009; Schiller et al., 2010; Lim et al., 2012]. Interestingly, each of these antifibrotic agents suppresses profibrotic signal-induced matrix protein collagen synthesis by a common molecular mechanism that is via interaction and sequestration of FATp300 from active transcriptional complex formed on collagen gene promoter and thus decreases lysine acetylation in chromatin histones and collagen synthesis. However, these cellular repressors or its agonists fail to prevent induced collagen synthesis in the presence of excess FATp300 strongly signifying the essential contribution of FATp300 in elevated matrix protein synthesis [Ghosh et al., 2000, 2001, 2004, 2009]. In addition, p53 or PPAR-γ deficient fibroblasts produce excessive matrix proteins due to increased interaction of FATp300 with Smad-containing transcriptional complex [Ghosh et al., 2004, 2008, 2009]. Consistent with these original observations at the cellular level, recent animal studies demonstrate that pharmacological inhibition of p53 activity [Dagher et al., 2012] or p53 gene deficiency [Sutton et al., 2013] is associated with severe fibrosis in animal models of kidney injury. Similarly, while PPAR-γ-deficiency in fibroblasts is associated with increased susceptibility to bleomycin-induced murine skin fibrosis [Kapoor et al., 2009], antidiabetic drug rosiglitazone-activated PPAR-γ prevents bleomycin-induced murine skin fibrosis [Wu et al., 2009] and angiotensin II-mediated hypertension-induced rat cardiac fibrosis [Hao et al., 2008]. Collectively, these studies unequivocally support the original notion [Ghosh et al., 2000, 2001] that disruption of active Smad-FATp300 complex formation on collagen gene promoter using a druggable compound or small peptide will be a viable approach to halt initiation and progression of fibrogenesis in different injured or chronically stressed organs (Fig. 1).

The clinical significance of FATp300 in fibrogenesis is evidenced by elevated levels of FATp300 in fibrotic skin and fibroblasts derived from systemic sclerosis patients [Bhattacharyya et al., 2005; Ghosh et al., 2013], and in the ventricular tissues derived from heart failure patients (akg, unpublished). Similarly, elevated FATp300 is associated with increased levels of acetylated histone positive nuclei and elevated collagen deposition in hypertension-induced murine

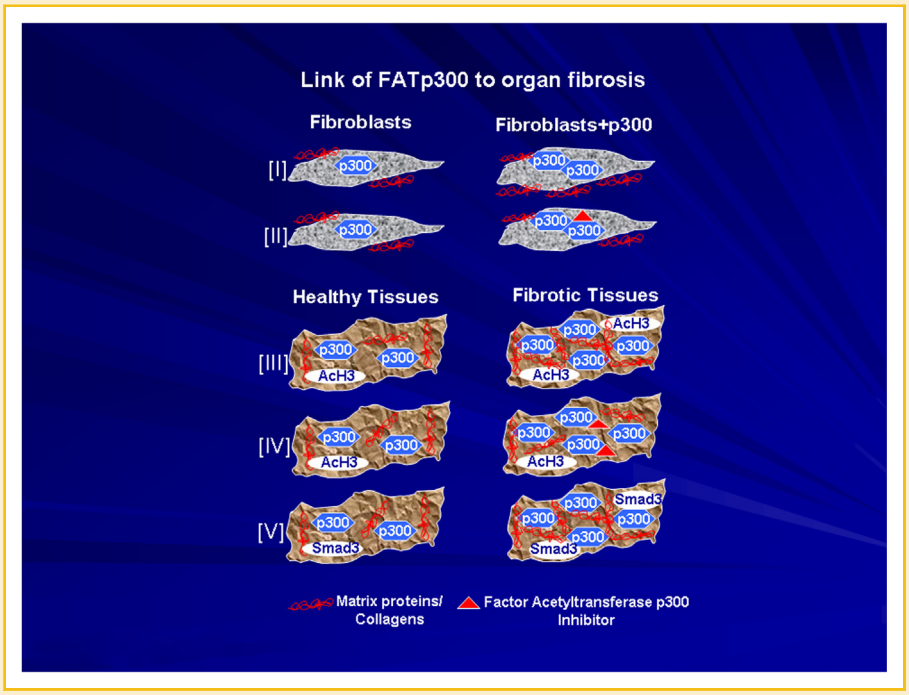

Fig. 2. Link of factor acetyltransferase p300 (FATp300) to organ fibrosis. I. Excess FATp300 stimulates matrix protein synthesis in hyperactive myofibroblasts originated from resident fibroblasts or vascular endothelial cells or bone-marrow-derived progenitor cells. II. FATp300 inhibitor blunts excessive synthesis of matrix protein collagen. III. FATp300 and acetylated histones in specific residues are significantly elevated in different fibrotic tissues like skin and heart. IV. Inhibition of FAT activity of p300 is associated with decreased fibrogenesis. FATp300 is an ideal therapeutic target for fibrosis treatment. V. Physical interaction of FATp300 with Smad3 is significantly higher in fibrotic tissues. Disruption of Smad3–FATp300 interaction using small peptide is a viable approach to control fibrogenesis.

model of cardiac fibrosis (akg, unpublished). FATp300 is also significantly elevated during "endothelial-to-mesenchymal transition" a biological process that contributes to cardiac fibrogenesis in adults [Ghosh et al., 2010, 2012]. Moreover, FATp300 is elevated in human glomerulonephritis [Kassimatis et al., 2006] and left ventricle derived from myocardial infarction-induced heart failure animal model [Sunagawa et al., 2011]. Based on these seminal observations, it is reasonable to propose that fibrogenesis is an epigenetic event and FATp300 is a budding biomarker of fibrogenesis. The feasibility of FATp300 as a therapeutic target of fibrogenesis has been supported by the observations that hypertension reducing drug enalapril reduces the level of FATp300 and myocardial infarction-induced perivascular and interstitial fibrosis signifying the contribution of FATp300 in cardiac fibrogenesis [Sunagawa et al., 2011]. Similarly, turmeric derivative curcumin-mediated inhibition of p300-FAT activity is associated with reduced hypertension-induced cardiac hypertrophy, fibrosis, and improved heart function in an animal model [Morimoto et al., 2008]. Moreover, the red grape-derivative anti-inflammatory resveratrol-induced deacetylase sirtuin 1 reduces cardiac hypertrophy and cardiac fibrosis via downregulation of FATp300 [Kuno et al., 2013] further supporting the potential link between FATp300 and organ fibrogenesis. Therefore, neutralization of elevated p300-FAT activity using small molecule inhibitor is a viable approach to normalize excess matrix protein synthesis in harassed organs and amelioration of organ fibrosis (Figs. 1 and 2).

Collective experimental evidence of the past decade present a paramount role of elevated FATp300 in organ fibrosis and its potentiality as a therapeutic target for treatment of patients at risk of developing organ fibrosis. The biggest challenge in this passage to fibrosis therapy is the identification of a safe druggable small molecule inhibitor of p300-FAT activity or small peptide that diminish increased interaction of FATp300 to transcriptional complex on collagen gene promoter and to test its efficacy in preclinical studies using a large cohort of animal models of organ fibrosis. Based on the promising preclinical outcomes including its excellent bioavailability, cell permeability, and target specificity; safest and most potent small molecule compound or peptide will be eventually used in human clinical trials involving patients with a wide variety of chronic stress or repeated injury-associated fibrosis in multiple organs.

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REFERENCES

Arrowsmith CH, Bountra C, Fish PV, Lee K, Schapira M. 2012. Epigenetic protein families: a new frontier for drug discovery. Nat Rev Drug Discov 11:384–400.

Bhattacharyya S, Ghosh AK, Pannu J, Mori Y, Takagawa S, Chen G, Trojanowska M, Gilliam AC, Varga J. 2005. Fibroblast expression of the coactivator p300 governs the intensity of profibrotic response to transforming growth factor beta. Arthritis Rheum 52:1248–1258.

Border WA, Noble NA. 1994. Transforming growth factor beta in tissue fibrosis. N Engl J Med 331:1286–1292.

Dagher PC, Mai EM, Hato T, Lee SY, Anderson MD, Karozos SC, Mang HE, Knipe NL, Plotkin Z, Sutton TA. 2012. The p53 inhibitor pifithrin-α can stimulate fibrosis in a rat model of ischemic acute kidney injury. Am J Physiol Renal Physiol 302:F284–F291.

Ghosh AK, Yuan W, Mori Y, Varga J. 2000. Smad-dependent stimulation of type I collagen gene expression in human skin fibroblasts by TGF-beta involves functional cooperation with p300/CBP transcriptional coactivators. Oncogene 19:3546–3555.

Ghosh AK, Yuan W, Mori Y, Chen SJ, Varga J. 2001. Antagonistic regulation of type I collagen gene expression by interferon-gamma and transforming growth factor-beta. Integration at the level of p300/CBP transcriptional coactivators. J Biol Chem 276:11041–11048.

Ghosh AK, Bhattacharyya S, Varga J. 2004. The tumor suppressor p53 abrogates Smad-dependent collagen gene induction in mesenchymal cells. J Biol Chem 279:47455–47463.

Ghosh AK, Wei J, Wu M, Varga J. 2008. Constitutive Smad signaling and Smad-dependent collagen gene expression in mouse embryonic fibroblasts lacking peroxisome proliferator-activated receptor-gamma. Biochem Biophys Res Commun 374:231–236.

Ghosh AK, Bhattacharyya S, Wei J, Kim S, Barak Y, Mori Y, Varga J. 2009. Peroxisome proliferator-activated receptor-gamma abrogates Smad-dependent collagen stimulation by targeting the p300 transcriptional coactivator. FASEB J 23:2968–2977.

Ghosh AK, Bradham WS, Gleaves LA, De Taeye B, Murphy SB, Covington JW, Vaughan DE. 2010. Genetic deficiency of plasminogen activator inhibitor-1 promotes cardiac fibrosis in aged mice: involvement of constitutive transforming growth factor-beta signaling and endothelial-to-mesenchymal transition. Circulation 122:1200–1209.

Ghosh AK, Nagpal V, Covington JW, Michaels MA, Vaughan DE. 2012. Molecular basis of cardiac endothelial-to-mesenchymal transition (EndMT): differential expression of microRNAs during EndMT. Cell Signal 24:1031–1036.

Ghosh AK, Bhattacharyya S, Lafyatis R, Farina G, Yu J, Thimmapaya B, Wei J, Varga J. 2013. P300 is elevated in systemic sclerosis and its expression is positively regulated by TGF-β: epigenetic feed-forward amplification of fibrosis. J Invest Dermatol 133:1302–1310.

Hao GH, Niu XL, Gao DF, Wei J, Wang NP. 2008. Agonists at PPAR-gamma suppress angiotensin II-induced production of plasminogen activator inhibitor-1 and extracellular matrix in rat cardiac fibroblasts. Br J Pharmacol 153:1409–1419.

Hayden T. 2011. Scarred by disease. Nat Med 17:18–20.

Kapoor M, McCann M, Liu S, Huh K, Denton CP, Abraham DJ, Leask A. 2009. Loss of peroxisome proliferator-activated receptor gamma in mouse fibroblasts results in increased susceptibility to bleomycin-induced skin fibrosis. Arthritis Rheum 60:2822–2829.

Kassimatis TI, Giannopoulou I, Koumoundourou D, Theodorakopoulou E, Varakis I, Nakopoulou L. 2006. Immunohistochemical evaluation of phosphorylated SMAD2/SMAD3 and the co-activator P300 in human glomerulonephritis: correlation with renal injury. J Cell Mol Med 10:908–921.

Kuno A, Hori YS, Hosoda R, Tanno M, Miura T, Shimamoto K, Horio Y. 2013. Resveratrol improves cardiomyopathy in dystrophin-deficient mice through SIRT1 protein-mediated modulation of p300 protein. J Biol Chem 288:5963–5972.

Lim JY, Oh MA, Kim WH, Sohn HY, Park SI. 2012. AMP-activated protein kinase inhibits TGF-β-induced fibrogenic responses of hepatic stellate cells by targeting transcriptional coactivator p300. J Cell Physiol 227:1081–1089.

Morimoto T, Sunagawa Y, Kawamura T, Takaya T, Wada H, Nagasawa A, Komeda M, Fujita M, Shimatsu A, Kita T, Hasegawa K. 2008. The dietary compound curcumin inhibits p300 histone acetyltransferase activity and prevents heart failure in rats. J Clin Invest 118:868–878.

Schiller M, Dennler S, Anderegg U, Kokot A, Simon JC, Luger TA, Mauviel A, Böhm M. 2010. Increased cAMP levels modulate transforming growth factor-beta/Smad-induced expression of extracellular matrix components and other key fibroblast effector functions. J Biol Chem 285:409–421.

Sunagawa Y, Morimoto T, Wada H, Takaya T, Katanasaka Y, Kawamura T, Yanagi S, Marui A, Sakata R, Shimatsu A, Kimura T, Kakeya H, Fujita M, Hasegawa K. 2011. A natural p300-specific histone acetyltransferase inhibitor, curcumin, in addition to angiotensin-converting enzyme inhibitor, exerts beneficial effects on left ventricular systolic function after myocardial infarction in rats. Circ J 75:2151–2159.

Sutton TA, Hato T, Mai E, Yoshimoto M, Kuehl S, Anderson M, Mang H, Plotkin Z, Chan RJ, Dagher PC. 2013. P53 is renoprotective after ischemic kidney injury by reducing inflammation. J Am Soc Nephrol 24:113–124.

Wahl SM. 1994. Transforming growth factor beta: the good, the bad, and the ugly. J Exp Med 180:1587–1590.

Wu M, Melichian DS, Chang E, Warner-Blankenship M, Ghosh AK, Varga J. 2009. Rosiglitazone abrogates bleomycin-induced scleroderma and blocks profibrotic responses through peroxisome proliferator-activated receptor-gamma. Am J Pathol 174:519–533.