BASIC SCIENCE JOURNAL CLUB OF EUSTAR YIG

Jan-Apr 2018

January2018

The histone demethylase Jumonji domain-containing protein 3 (JMJD3) regulates fibroblast activation in systemic sclerosis

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Bergmann C, Brandt A, Merlevede B, Hallenberger L, Dees C, Wohlfahrt T, Pötter S, Zhang Y, Chen CW, Mallano T, Liang R, Kagwiria R, Kreuter A, Pantelaki I, Bozec A, Abraham D, Rieker R, Ramming A, Distler O, Schett G, Distler JHW. Ann Rheum Dis. 2018 Jan;77(1):150-158.

Fibroblast are constitutively activated in systemic sclerosis (SSc) and epigenetic alterations may play a determinant role in this endogenous activation. Trimethylation of histone H3 on lysine 27 (H3K27me3) is a common histone modification that represses the transcription of target genes. An imbalanced regulation of this methylation has been implicated in fibroblast functions, as the demethylation of H3K27 promotes fibroblast activation and induced fibrosis.

In this article, the authors explored the role of the Jumonji domain-containing protein 3 (JMJD3) and the ubiquitously transcribed tetratricopeptide repeat on chromosome X (UTX), two highly specific histone–demethylases which could participate to the constitutively activated state of SSc fibroblasts. The authors tested the hypothesis that targeted inhibition of H3K27me3 demethylases could be a novel approach to limit fibrosis in SSc.

Expression levels of UTX were comparable in fibroblasts from SSc patients and healthy donors. By contrast, JMJD3 was over-expressed in the skin fibroblasts of SSc patients even after several passages in vitro, and in various murine models of SSc such as bleomycin-induced skin fibrosis, scleroderma cGvHD or topol-induced dermal fibrosis. Using siRNA approa- ch targeting SMAD3/TGFb signaling pathway and a murine model of TGFb receptor overexpression, the author demon- strated that JMJD3 is induced by TGFβ in a SMAD3-dependent manner.

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The authors then demonstrated that inhibiting JMJD3, using the specific inhibitor GSKJ4, significantly reduces collagen release by fibroblasts. The authors secondly evaluated if these effects of JMJD3 regulation on collagen secretion were dependent on a regulation of FRA2, an AP1 transcription factor especially overexpressed in SSc dermal fibroblasts. H3K27me3 was reduced at the FRA2 promoter, and this reduced methylation was dependent on TGFβ signaling in SSc fibroblasts. GSKJ4 significantly prevented these effects of TGFβ on H3K27me3 at the FRA2 promoter. The inhibitory effect of GSKJ4 on collagen release was reduced when FRA2 was already down-regulated by siRNA, demonstrating that the reduction of collagen secretion by GSKJ4 was dependent on its effects on FRA2.

In vivo, GSKJ4 both significantly prevented and induced regression of skin fibrosis in the bleomycin-induced model of SSc and ameliorated topol-induced skin and lung fibrosis. Treatment with GSKJ4 increased H3K27me3 levels in both mice models in well-tolerated doses.

Altogether, this article offers an interesting view on the role of epigenetic in SSc, highlighting a deregulation of JMJD3 in SSc fibroblasts. JMJD3 modulates fibroblast activation by regulating the levels of H3K27me3 at the promoter of FRA2. Therefore, the targeted inhibition of JMJD3 by GSKJ4 ameliorates fibrosis in two mice models showing that epigenetic modulations may offer new therapeutic opportunities in the future.

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Scleroderma fibroblasts suppress angiogenesis via TGF-β/caveolin-1 dependent secretion of pigment epithelium-derived factor.

Liakouli V, Elies J, El-Sherbiny YM, Scarcia M, Grant G, Abignano G, Emma C Derrett-Smith EC, Esteves F, Paola Cipriani P, Emery P, Denton CP, Giacomelli R, Mavria G, Del Galdo F.
Ann Rheum Dis 2018; 77:431–440.

The authors have previously demonstrated that Transforming growth factor beta (TGF-β) induces caveolin 1 down-regulation and that Caveolin-1 (Cav-1) knock-down fibroblasts displayed increased TGF-β signaling and induced tissue fibrosis.

Here, the authors focused on Pigment Epithelial Derived Factor (PEDF), an antiangiogenic factor, member of the serpin superfamily, which expression was previously shown to be upregulated in SSc skin biopsies in proteomic studies. They investigated the effect of TGF-β and Cav-1 upon PEDF expression and the role of fibroblasts-secreted PEDF in modulating angiogenesis.

They performed in vitro studies and in vivo studies based on diffuse cutaneous SSc (dcSSc) patient's sera, skin biopsies from Healthy Controls (HC) and dcSSc as well as skin biopsies-derived fibroblasts. dcSSc and HC-derived fibroblasts were silenced either for Cav-1 or PEDF and their effect upon endothelial cell lines-based Matrigel angiogenesis assay was assessed. They also used a previously validated mouse model of ligand dependent-upregulation of TGF-β signaling in tissue fibroblasts that displayed endothelial injury-induced fibroproliferative vasculopathy.

They showed that PEDF expression is increased in SSc patients skin biopsies with a prominent expression in fibroblasts and myofibroblasts of the lower dermis.

They evidenced that TGF-β induced PEDF expression in SSc fibroblasts in vitro. In addition, SSc fibroblasts suppressed angiogenesis in a PEDF-dependent manner. As the expression levels of Cav-1 (low) and PEDF (high) were inversely correlated in the SSc patients skin biopsies, they further deciphered this link in vitro. To reproduce the low Cav-1 expression observed in SSc fibroblasts, the authors silenced Cav-1 expression in HC fibroblasts. Hence, the authors showed that Cav-1-knockdown HC fibroblasts exhibited increased secretion of PEDF. As decreased number of capillaries were observed in SSc skin biopsies,

the effect of Cav-1 silencing upon angiogenesis was assessed in vitro. Interestingly, Cav-1-knockdown fibroblasts inhibited the angiogenesis via the restriction of tubule formation without affecting endothelial cells viability or proliferation. They also showed that this anti-angiogenic effect was mediated by fibroblasts' secretion of PEDF.

Finally, using the TBRII delta K transgenic mice which exhibit TGF-β receptor hyperactivation in tissue fibroblasts, they confirmed that the phenotype of low Cav-1 and high PEDF expression associated with defective angiogenesis is driven by TGF-β signalling.

Altogether, the results of this study propose a model that goes further into the interplay between fibrosis and defective vasculopathy in SSc. They suggest an interesting mechanistic model to explain TGF-β-induced vasculopathy based on Cav-1 and PEDF where Cav-1 downregulation potentiates TGF-β-signalling which promotes PEDF secretion and its anti-angiogenic effect.

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Inhibitory regulation of skin fibrosis in systemic sclerosis by apelin/APJ signaling

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Ishikawa O, Motegi SI. Arthritis Rheumatol. 2018 Apr 20.

Apelin is a selective endogenous ligand of the G protein-coupled receptor APJ, and apelin/APJ signaling mainly regulates cardiovascular functions, fluid homeostasis, angiogenesis and adipose tissue functions. Apelin is secreted by endothelial cells and pericytes and binds to APJ, resulting in the dilation of blood vessels and increased blood flow.

The expression of Apelin in pulmonary arterial endothelial cells in PAH patients was significantly reduced. The authors wonder whether Apelin could have a role in skin fibrosis in SSc besides its role in the vasculopathy. They showed ex vivo in human that Apelin was less expressed by SSc fibroblasts and in SSc skin. In the peripheral blood, Apelin levels were inversely correlated to skin fibrosis. TGF-b decreased Apelin expression in fibroblasts. Silencing Apelin induced a profibrotic phenotype in fibroblasts.

They observed that Apelin has an inhibitory effect on TGF-b/Smad signaling in fibroblasts in vitro. Authors performed in vivo experiments using the bleomycine mouse model of SSc and showed that the administration of apelin inhibited the induced skin fibrosis. They finally tested an agonist of APJ that led to an inhibition of the TGF-b/Smad signaling in fibroblasts and of the induced skin fibrosis in the bleomycin mouse model. Collectively these results indicated a potential new target to reduce SSc fibrosis. The use of the synthetic agonist peptide proposed by the authors could present the advantage to have an even higher inhibition potential on the TGF-b/Smad signaling.

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