conferenceseries.com

J Biotechnol Biomater 2018, Volume 8
DOI: 10.4172/2155-952X-C1-089

JNK-targeting regenerative nanoparticles for augmented elastic tissue repair in proteolytic disorders

Anand Ramamurthi[1], **Andrew Camardo**[1] and **Dhruv Seshadri**[2]
[1]Cleveland Clinic, USA,
[2]Case Western Reserve University, USA

Proteolytic disorders involve chronic breakdown of elastic fibers by matrix metalloproteinases (MMPs). Adult cells are inherently deficient in effecting regenerative repair of elastic fibers. We previously showed that at low (<10 ug/ml) doses, doxycycline (DOX) inhibits MMPs as it does at much higher oral doses, but also stimulates elastic matrix neoassembly and crosslinking. In this work, we show that both these effects of low dose DOX are linked to its upregulation of transforming growth factor beta (TGF-β1) upon targeted inhibition of a regulatory protein c-Jun-N-terminal kinase 2 (JNK2). We also investigated if sustained and steady release of DOX from biodegradable polymer nanoparticles (NPs) we have developed that independently provide pro-elastogenic and anti-MMP effects, is able to synergistically improve quantity and quality (crosslinking, fiber formation and density, stability against proteolysis) in *in vitro* cultures of cytokine-activated rat smooth muscle cells from aortic aneurysms, a vascular proteolytic disease (EaRASMCs). Cytokine-activated EaRASMC cultures were treated with (1-20 ug/ml) or without DOX (treatment controls) and compared with cultures of healthy SMCs. Western Blots detected expression of JNK isoforms, pJNK, and TGF-β1 and outcomes were correlated with elastic matrix amounts, desmosine crosslinks, elastic fiber counts, MMP protein amounts and enzyme activities in the cell layers at 21 days of culture. Next, PLGA-PEG nanoparticles encapsulating DOX were formulated with pendant cationic amphiphile groups and shown to release DOX at the JNK inhibitory doses. Cytokine-activated EaRASMCs were cultured with the DOX-NPs for 30 min or 21 days. Healthy SMCs, and EaRASMCs cultured with blank NPs and no NPs served as controls and assessments were performed as in the earlier experiment. DOX inhibited expression and phosphorylation of JNK. Levels of JNK2 and pJNK, were lower in treated cultures and similar to healthy controls. JNK inhibition increased TGF-β1 expression and these outcomes were dose dependent & correlated positively to elastic matrix amounts, crosslinking and fiber counts and negatively to MMPs. DOX-delivery from the NPs more effectively in stimulated elastic fiber formation and crosslinking and inhibiting MMPs versus exogenous DOX. The results suggest that JNK inhibition is a useful metric to assess matrix-regenerative properties of DOX and emphasize synergy between DOX & our functionalized nanocarriers.

ramamua@ccf.org