Accepted as a poster at TOBI 2018, June 7-9, 2018, Las Vegas, Nevada.

Preclinical Evidence for SM04690, a Small Molecule Wnt Pathway Inhibitor, as a Potential Treatment for Degenerative Disc Disease

Vishal Deshmukh, Charlene Barroga, and Yusuf Yazici

Samumed, LLC, San Diego, CA

Objective: To evaluate the potential of SM04690, a novel, small-molecule, Wnt pathway inhibitor, to promote disc healing in a rodent model of degenerative disc disease (DDD).

Design: 1. *In vitro*: Wnt pathway inhibition was measured using a cell-based reporter assay, proliferation of rat NP cells by cell doubling index (CDI= cell number/initial cell number/days), and differentiation of NP cells into 'chondrocyte-like' cells by Alcian blue absorbance based quantification. Myofibroblast differentiation was assessed in TGF-β1 stimulated human dermal fibroblasts by measuring smooth muscle actin (αSMA) using immunocytochemistry. 2. *In vivo*: Histological and anatomical effects of intervertebral disc (IVD) injection of SM04690 in a rat coccygeal needle puncture model of DDD was evaluated.

Setting: *In vitro* and *in vivo* experiments.

Interventions: Single, intradiscal injection of SM04690 (0.33 µg/disc).

Main Outcome Measures: 1. *In vitro*: CDI and Alcian blue absorbance. 2. *In vivo*: Disc height index (DHI = disc height/vertebral height) and histological scoring of Safranin O-stained sections for annulus fibrosis (AF) integrity, AF and NP border, cellularity, and NP matrix.

Results: 1. *In vitro*: SM04690 induced dose-dependent proliferation of NP cells with CDI ~2-fold greater than vehicle ($p<0.05$). SM04690-treated cells showed increased Alcian blue absorbance vs. vehicle ($P<0.01$), indicating differentiation to "chondrocyte-like" cells and production of proteoglycan components of extracellular matrix (ECM). SM04690 inhibited TGF-β1-induced expression of α-SMA (EC_{50}=16.7 nM). 2. *In vivo*: In the rat model, SM04690 increased Safranin-O/Fast Green stained cartilage matrix (Figure A) vs. vehicle. SM04690 decreased histology scores at 8 weeks ($P<0.05$; Figure B), indicating reduced AF lamellar disorganization and fragmentation, larger NP area, increased cellularity of NP and increased ECM vs. vehicle. Radiographic measurement of disc height demonstrated increased DHI at 6 weeks ($P<0.05$; Figure C) in SM04690-treated rats vs. vehicle.

Conclusions: Findings support SM04690 as a potential DDD treatment. Further clinical trials are needed to advance understanding of the SM04690 molecule.

Figure: SM04690 stimulated differentiation of NP cells and improved disc height and health in a rat model of DDD.