Background

- Degenerative Disc Disease (DDD), a major cause of low back pain, is characterized by degeneration of intervertebral discs (IVDs), which is composed of central nucleus pulposus (NP) surrounded by collagenous annulus fibrosus (AF) and cartilaginous endplate.

- The NP is comprised of progenitor cells that can differentiate into chondrocytes to form a proteoglycan and collagen-rich extracellular matrix (ECM), responsible for hydration and IVD function, loss of NP cellularity and hydration results in degraded disc height and function.

- Wnt signaling plays a key role in IVD development and growth. Excessive Wnt signaling results in inhibition of NP cell proliferation, upregulation of pro-inflammatory cytokines and ECM degrading enzymes, and apoptosis of NP cells, which lead to IVD degeneration and DDD.

- Treatment of DDD is limited to analgesics and/or surgery aimed at relieving symptoms and experimental biological treatments with as yet unknown efficacy. No current FDA approved therapy has been shown to reverse disc degeneration.

- SM04690, a potent small molecule Wnt pathway inhibitor, as a potential injectable therapeutic for the treatment of DDD.

Methods

- To identify Wnt signaling inhibitors, a small molecule chemical library was screened in a cellular Wnt pathway-based β-catenin/TCF-luciferase reporter assay in SM04690-untreated cancer cells.

- Anti-inflammatory activity was evaluated by measuring a panel of secreted pro-inflammatory cytokines from SM04690-treated with supernatant (SA) or ECM.

- Effects on IVDs were assessed in TGF-β1-treated human dermal fibroblasts (HDFs) by measuring smooth-muscle cells (αSMA).

- In vitro proliferation of NP cells isolated from rat disc tissue, treated with vehicle or SM04690 for 5 days, was measured by cell clumping index (ECM-off/number of cell numbers).

- Differentiation of NP progenitor cells into chondrocyte-like NP cells with 12 days of vehicle or SM04690 treatment was assessed by Alcian blue staining and absorbance-based quantification.

- Pharmacokinetics were evaluated by intravenous injection in rats and rabbits, followed by LC-MS analysis of compound concentrations in the disc and plasma.

- Rat cocultured IVD explant was evaluated in a 3D model.

- Injured discs were radiographed pre-surgery and 1 week (dosing point), 4 weeks, and 6 weeks post-surgery.

- Saturated OilFast Green or Masson’s Trichrome stained discs were histologically examined by blinded observers using a disc scoring system based on grading the integrity of AF, border between AF and NP, and cellularity and matrix of NP. Disc height index (DH) was calculated by averaging the anterior and posterior septum portions of the disc height and dividing by the average of the adjacent vertebral body.

Results

- SM04690 demonstrated specific and potent inhibition of Wnt signaling in vitro. SM04690 (300 nM) significantly reduced the expression of TCF4 and LFEl (Figure 3b).

- SM04690 stimulated NP cells to express Wnt target genes in vitro (Figure 3c).

- A single intradiscal injection of SM04690 improved disc height and shape in vivo model of degenerative intervertebral disc.

Discussion

- SM04690, a small molecule inhibitor of the Wnt signaling pathway, demonstrated potent anti-inflammatory and anti-fibrotic activity in vitro.

- SM04690 inhibited proliferation and differentiation of NP-derived progenitor cells in vitro.

- In vivo, the proteolytic effect of Wnt inhibition of SM04690 had sustained residence time in the disc and minimal systemic exposure in rats.

- In a radiologic model of DDD, a single intradiscal injection of SM04690 improved disc height, health, and shape after injury in vivo compared to vehicle controls.

- SM04690 inhibited the NP and IVD structure in this in vivo model of DDD.

- SM04690 has potential as a regenerative treatment for DDD.

- An Investigational New Drug application for SM04690 in ODD is open and human trials are planned for 2017.

References