Background

No therapy for osteoarthritis (OA) has yet to deliver both structural and symptomatic benefits. Symic is approaching this unmet clinical need with a novel matrix regulator, SB-061, inspired by aggrecan. Aggrecan through its binding to hyaluronic acid (HA) is critical for cartilage structure and plays a key role as part of the protective molecules shielding cartilage from degradation. Native aggrecan is lost early in the progression of OA, and restoring some aspects of its function is hypothesized to reduce OA progression. SB-061 was designed as a functional mimic of aggrecan, and was evaluated in a standard rodent model of OA.

Design of SB-061

SB-061 was synthesized by linking the glycosaminoglycan (GAG) chondroitin sulfate (CS) with peptides known to bind to hyaluronic acid (HA).

- CS, the predominant GAG on native aggrecan, has pleiotropic functionality as it provides a hydrophilic barrier, binds growth factors, and has demonstrated anti-inflammatory activity through the NF-kB pathway.
- HA-binding peptides mimic aggrecan by localizing SB-061 onto hyaluronic acid molecules within the joint. In binding to HA, SB-061 is a competitive inhibitor to the pro-inflammatory CD44-HA pathway.

Immobilization of HA and Binding of SB-061

- HA (10-20 kDa, LifeCore Biomedical) was covalently linked to an amine coated plate. Excess HA was rinsed, and the plate was then incubated with SB-061.
- SB-061 binding to the HA was detected using an antibody developed to the molecule.
- Specific binding of SB-061 to HA was verified by examining binding to plate surface without HA coating.

Method for SB-061 Binding to HA Gels

- HA gels (HySytem Hydrogel) were prepared per manufacturer’s protocol.
- Labeled SB-061 was added to the gels and allowed to incubate for 2 hours on an orbital shaker.
- Gels were then rinsed overnight with 1X PBS on an orbital shaker.
- Labeled SB-061 was detected using confocal microscopy.

SB-061 Binds and is Retained in HA Gels

- HA gels retained labeled SB-061 or the HA binding peptide, whereas the backbone GAG CS was not retained in the gel.
- The data supports binding and retention of SB-061 onto HA molecules present in the knee joint.

Study Design for Cell Based CD44 Inhibition

Competitive inhibition of SB-061 for activated inflammatory cells was evaluated as an anti-inflammatory mechanism.

- Calcein-AM stained peripheral blood mononuclear cells (PBMCs) were activated with TNFα to induce CD44 expression, and then incubated on an HA coated plate with or without SB-061 for 30 minutes prior to rinsing.
- PBMCs bound the plate were detected by fluorescence.

SB-061 Inhibits Inflammatory Cell Binding to HA

- SB-061 reduced PBMC binding to HA coated plates in a dose dependent manner.
- This data supports an anti-inflammatory mechanism of SB-061 presumably through inhibition of HA-CD44 complex.

Animal Model of Osteoarthritis

- Rats received a medial meniscal tear in their right hind-limbs.
- SB-061 was injected intra-articularly into the injured joint beginning 1-week after surgery and weekly throughout the study.
- SB-061 was delivered at doses between 0.04 and 5 mg/mL in 50 μL volume. Injections of vehicle were used as a negative control.
- Incapacitance testing was performed to detect differences in pain related responses (load bearing on the injured joint) throughout the duration of the study. Incapacitance testing took place 2 days post dosing.

SB-061 Results in Sustained Pain Reduction

- (Left) SB-061 produces approximately 30% maximal levels of pain relief (as measured by weight bearing) compared to vehicle treated control animals. The pain reduction was sustained throughout the 6-week duration of the study. The study was repeated in 4 independent experiments with similar effects each time. P<0.05 beginning at week 3. Arrowheads indicate when treatment dose was administered.
- (Right) SB-061 reduces pain in an dose dependent manner. A dose of 5 mg/mL, the highest concentration tested, provided the greatest pain reduction (as measured by weight bearing). Pain reduction with SB-061 reached approximately 30% as compared to vehicle treated control animals.

Conclusions

- SB-061 was designed as a functional mimic of native aggrecan. It binds to HA through peptides linked to a chondroitin sulfate (CS) backbone.
- SB-061 significantly reduces pain in an animal model of osteoarthritis. The pain reduction is sustained and dose dependent.
- SB-061 inhibits inflammatory cell binding to HA, presumably by masking CD44 interactions.
- SB-061 is expected to additionally have disease modifying effects.
- A clinical trial, called MODIFY2, is underway to evaluate SB-061 in osteoarthritis.

Declaration of Interest

- Symic Bio Funded all aspects of this work.
- All Symic Bio authors are employees and have stock options in the company.
- Authors from Lilly Research Laboratories have no financial relationships to disclose.
- Authors from Nordic have stock options in Symic Bio.

References