Mechanisms of Facet Degeneration Adjacent to Engineered Discs Implanted with Plate Fixation

Sarah E. Gullbrand[1,2], Ali Kiapour[3], Michael Hast[2], Brianna S. Orozco[1,2], Matthew Fainor[1,2], Stanton Godshall[2], Rachel Hilliard[2], Thomas P. Schaer[2], Robert L. Mauck[1,2], Harvey E. Smith[1,2]

[1]Corporal Michael J. Crescenz VA Medical Center, Philadelphia, PA, [2]University of Pennsylvania, Philadelphia, PA [3]Massachusets General Hospital, Boston, MA

INTRODUCTION: Intervertebral disc degeneration is commonly associated with back and neck pain, and standard surgical treatments do not restore spine function. Replacement of the degenerative disc with a living, tissue-engineered construct has the potential to restore normal structure and function to the spine [1]. Toward this goal, our group has developed endplate-modified disc-like angle ply structures (eDAPS) that recapitulate the native structure and function of the disc. These implants combine a cell-seeded hydrogel nucleus pulposus (NP) and an electrospun poly(ε-caprolactone) (PCL) annulus fibrosus (AF) with acellular PCL foam endplates [2,3]. Given literature demonstrating the potential for extrusion of engineered discs implanted without fixation in dogs [4], our initial large animal studies utilized rigid internal fixation of the eDAPS implanted level [5]. While this method ensured retention of the eDAPS, prior work has shown that rigid fixation of the motion segment can contribute to osteoarthritis (OA) of the adjacent facet joints [6]. The purpose of this *in vivo* and *in silico* study was to characterize facet health adjacent to eDAPS implants *in vivo* and understand alterations to the biomechanics of cervical spine with plate fixation. We hypothesized that plate fixation would increase stresses in the facet joints, contributing to the initiation and progression of facet pathology.

METHODS: *In vivo* studies: eDAPS sized for the goat and human cervical disc space (9 mm height, 16 mm diameter) were fabricated as previously described [4] and seeded with allogeneic caprine bone-marrow derived mesenchymal stem cells. eDAPS were cultured for a total of 12-15 weeks in a chemically defined medium with TGF-β3 prior to implantation. With IACUC approval, 7 male castrated large frame goats underwent a ventral midline discectomy to implant the eDAPS at the C2-C3 level. Implanted motion segments were immobilized with a titanium anterior cervical locking plate. All animals were euthanized 10 weeks following implantation. In one group (n=3), the cervical plate was left in place for the duration of the study (Fixed group). In the second group (n=4), a second surgery was performed at 5 weeks post-implantation to remove the plate and restore normal physiologic loading to the eDAPS implant (Remobilized group). The C3-C4 levels were utilized as internal healthy controls in all animals. Following euthanasia, the posterior facet joints at each level were disarticulated, stained with India Ink and imaged to quantify cartilage degeneration. Facets were then potted for creep indentation testing of the articular cartilage as previously described [7]. Following mechanical testing, the facets were fixed, decalcified and processed through paraffin for OARSI histopathology scoring. Statistical comparisons between groups were assessed via ANOVA using GraphPad Prism.

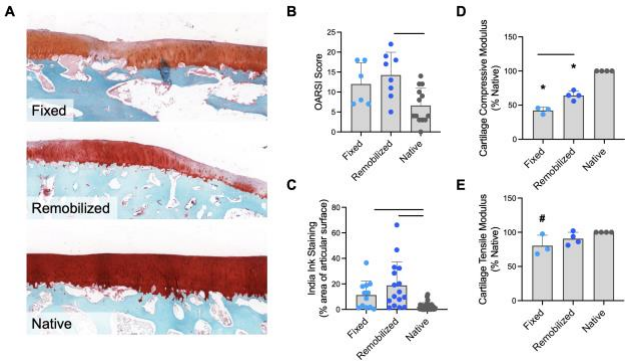

Figure 1. A). Safranin-O/Fast Green stained histology of facet cartilage in each experimental group. B). Facet OARSI scoring and C). India ink staining; quantification of facet cartilage D). compressive and E). tensile moduli. Bars denote significance $p<0.05$, * = $p<0.05$, # = $p<0.1$ compared to native.

Finite element modeling: CT images of three healthy goat cervical spines (C4-C5) were obtained. The images were segmented and converted to 3D anatomical geometries using Dicom-to-Print (D2P, Oqton Inc.). The 3D models of the vertebra were imported into Simplware (Synopsys Inc.) to create volumetric meshes for Finite Element (FE) analysis. The meshed geometries were then imported into Abaqus (Simulia Inc.) for the creation of three FE models of C4-C5 functional spine units (FSU) with connecting tissues and material details (**Figure 2A-D**). The FE models were then modified to simulate the two states including intact (healthy) and plated (instrumented with a 4-screw fixation plate system). Each model was then validated against cadaveric mechanical testing of the same specimens, which included 100 cycles of 300N compression loading. The validated models were then used to simulate the anatomical loadings of flexion/extension, left and right bending, and left and right axial rotation under 1.5 Nm bending load. The segmental ROM and load sharing at the facet joints of the three FSUs were compared in the healthy and plated conditions.

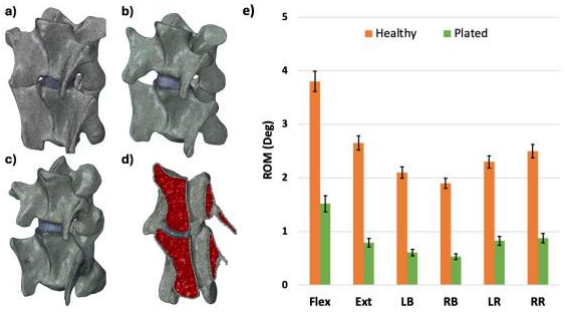

Figure 2. A) FE FSU model I, B) FE FSU model II, C) FE FSU model III and D) section view of FE FSU showing the cortical and cancellous bone segments. E). Comparison of anatomical ROM of FE Goat FSUs with three different conditions at 1.5 Nm bending load.

RESULTS: *In vivo* studies: Osteoarthritic changes to the facet joints were evident adjacent to eDAPS implants in both the fixed and remobilized groups, as evidenced by OARSI histopathology scoring and India ink staining for cartilage wear (**Figure 1A-C**). Facet cartilage compressive and tensile moduli were significantly reduced in facets adjacent to eDAPS in the fixed group, however remobilization mitigated these losses in function, with significant improvements in compressive modulus compared to the fixed group (**Figure 1D-E**). Finite element modeling: The average displacement motions of FE FSUs were within one STD of the cadaveric FSU data in healthy and plated states. When comparing the anatomic ROM of the FSU at 1.5 Nm load, the data were close to average data previously reported *in vitro* for the goat spine [8]. The FE analysis demonstrated a noticeable reduction in ROM following instrumentation with the plate system (**Figure 2E**), however the fixation construct resulted in slight shift in the load sharing, causing the peak stress on the facet joints to increase slightly in extension and axial rotation motions (**Table 1**).

	Healthy (Avg ± Std. Dev.)	Plated (Avg ± Std. Dev.)
Extension	2.60 ± 0.26	2.86 ± 0.05
Lateral Bending	3.30 ± 0.33	2.31 ± 0.07
Axial Rotation	2.50 ± 0.25	2.75 ± 0.05

Table 1. Peak stress (MPa) at the facet joints.

DISCUSSION: A challenge facing the translation of whole tissue engineered disc replacements is ensuring adequate stabilization of the construct following implantation while integration with the adjacent vertebral bone occurs. In prior studies, we stabilized the eDAPS via anterior cervical plate fixation, which is removed in a second surgery to restore physiologic loading to the implant. Our FE modeling results demonstrate, however, that such plate fixation shifts load sharing in the motion segment such that peak stresses on the facet joints increase during extension and axial rotation motions. This increase in facet loading is likely driving the osteoarthritic changes to the facet joints that we observe *in vivo* adjacent to the fixed eDAPS implants, as has been observed in other synovial joints [9]. Our *in vivo* studies also suggest that facet OA is mitigated, but not prevented, by early (5 week) removal of the plate fixation. Our ongoing work is focused on tuning the mechanics of a 3D printed bioresorbable fixation system to preserve spinal kinematics at implantation while still stabilizing the eDAPS implant, which may prevent the development of facet OA and eliminate the need for a second surgical procedure to remove fixation.

SIGNIFICANCE: This combination of *in vivo* and *in silico* work elucidates mechanisms of facet OA adjacent to engineered disc implants. This information will aid in the development of improved implants and fixation systems, advancing the clinical translation of this technology.

REFERENCES: [1] Bowles+*PNAS*, 2011 [2] Nerurkar+*Spine*, 2010 [3] Martin+*SciRep*, 2017 [4] Moriguchi+*PloS One*, 2017 [5] Gullbrand+*STM*, 2018 [6] Wang+*Spine J*, 2018. [7] Moore+*J Tribol*, 2016. [8] Dong+*JOSR*, 2023 [9] Felson+*OA&C*, 2013 [10] Smith+*2023 ORS Proceedings, Paper #1212*

ACKNOWLEDGEMENTS: This work was supported by the Department of Veterans' Affairs and the Penn Center for Musculoskeletal Disorders.