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Role of endplates in contributing to compression behaviors of motion segments and intervertebral discs

Jeffrey J. MacLean, Julia P. Owen, James C. Iatridis*

Spine Bioengineering Lab, Department of Mechanical Engineering, 231B Votey Building, 33 Colchester Avenue, University of Vermont, Burlington, VT 05405, USA

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Abstract

The purpose of this study was to gain an improved understanding of the mechanical behavior of the intervertebral disc in the presence and absence of the vertebral endplates. Mechanical behaviors of rat caudal motion segments, vertebrae and isolated disc explants under two different permeability conditions were investigated and viscoelastic behaviors were evaluated using a stretched-exponential function to describe creep and recovery behaviors. The results demonstrated that both vertebrae and discs underwent significant deformations in the motion segment even under relatively low-loading conditions. Secondly, disruption of the collagenous network had minimal impact on equilibrium deformations of disc explants as compared to disc deformations occurring in the motion segments provided that vertebral deformations were accounted for; however, differences in endplate permeability conditions had a significant effect on viscoelastic behaviors. Creep occurred more quickly than recovery for motion segment and explant specimens. In addition, disc explants and motion segments both exhibited non-recoverable deformations under axial compression under low- and high-loading conditions. Results have important implications for interpreting the role of vertebral endplates in contributing to disc mechanical behaviors and direct application to mechanobiology studies involving external loading to rodent tail intervertebral discs.

Keywords: Spine biomechanics; Functional spinal unit; Explant; Vertebral endplate; Permanent deformation; Permeability

1. Introduction

Intervertebral discs play essential biomechanical roles allowing load support and mobility of the spine. Overloading and immobilization both contribute to accelerated disc degeneration which compromises this function and involves mechanical damage, diminished nutrient transport, loss of cell viability and altered biosynthesis (Stokes and Iatridis, 2004). Mechanical behaviors of motion segments are related to combined and interactive responses of discs and vertebrae. Discs and vertebrae both exhibit time-dependent behaviors associated with flow-dependent and flow-independent viscoelasticity mechanisms. Some biomechanical studies of the disc assumed vertebral endplates are rigid and impermeable (Spilker et al., 1984; Zimmerman et al., 1992; Shirazi-Adl and Parnianpour, 1993), an assumption often considered reasonable because the material properties of the disc tissues are substantially lower than that of bone. However, vertebral endplates bulge under axial deformations (Brinckmann et al., 1983) and undergo fracture at higher loads (Brinckmann and Horst, 1985; Adams et al., 2000). This apparent contrast demonstrates a lack of complete understanding of the role of endplates in contributing to disc mechanical behaviors under compression. Furthermore, improved knowledge of interactions between vertebrae and discs in biomechanical testing will clarify the appropriate in

*Corresponding author. Tel.: +1 802 656 2774; fax: +1 802 656 1929.
E-mail address: james.iatridis@uvm.edu (J.C. Iatridis).

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vitro test configuration for studying disc degeneration, damage, remodeling, gene therapy and tissue engineering.

Evaluating discs in the motion segment complex most closely mimics in vivo conditions but creates difficulty separating disc and vertebral mechanical behaviors, and leaves minimal control over post-mortem boundary condition changes such as decreased permeability due to clotting in the endplate (Lee et al., 2006). Isolating discs as explants allow for direct assessments of disc mechanical properties and improved control over boundary conditions; however the consequences of cutting the annulus fibers that anchor the disc to the vertebrae must be accounted for. In preparing explants in this study, we cut through the annular fibers anchoring the disc to the vertebrae yet used porous platens as endplates to insure frictional, rigid and highly permeable boundary conditions. Investigating how endplate deformation and permeability conditions affect intervertebral disc biomechanics is also a priority since endplate permeability plays a role in the degeneration process (Roberts et al., 1996; Bibby et al., 2001; Benneker et al., 2005).

The use of animal models in spine research is commonplace and important in its ability to test specific hypothesis despite obvious differences in geometry of motion segments and vertebrae (e.g. Lotz, 2004; Ledet et al., 2005). Use of rodent models for in vitro mechanical testing is limited because the small size presents differences in geometry with the human and makes sample preparation difficult. Caudal discs were also used as a representative structural and compositional model for discs research (Ohshima et al., 1993; Demers et al., 2004), and remain an attractive model because of the geometric simplicity and lack of sagittal curvature. Rat caudal discs are used in this study because of the substantial and growing body of in vivo and ex vivo mechanobiology research using small animals (e.g. rabbits, rats, and mice) (Iatridis et al., 1999; Gruber et al., 2002; Ching et al., 2003; Hsieh and Lotz, 2003; Norcross et al., 2003; Risbud et al., 2003; Maclean et al., 2004; An et al., 2005; Kim et al., 2005; Sobajima et al., 2005). It was recently reported that mouse lumbar discs matched the previously reported nonlinear response of human lumbar discs while mouse caudal discs only matched the linear response provided differences in cross-sectional areas were accounted for (Sarver and Elliott, 2005).

This study addresses the role of endplate conditions on mechanical behaviors of intervertebral discs by investigating mechanical behaviors of rat caudal motion segments, caudal disc explants, and vertebrae. This study tested the hypotheses that: (1) deformations of both the vertebrae and intervertebral discs account for observed deformation behaviors of the intact motion segment; (2) mechanical behaviors of the intervertebral disc tested as explants are similar to those in the motion segments provided that endplate boundary conditions are similar; and (3) axial compression loading on motion segments will lead to non-recoverable deformations in both the vertebrae and intervertebral disc.

2. Methods

Four experimental groups consisting of motion segments (Group 1, vertebra–disc–vertebra), disc explants (Group 2—using permeable platens & Group 3—using impermeable platens), and single vertebrae (Group 4) were isolated from adjacent caudal levels of Sprague–Dawley rats ($n = 44$, 11 per group). Rats were obtained within 2 h of their death, skin and soft tissues were removed from the tail which was flash frozen in liquid nitrogen. A motion segment, disc explant and vertebra were harvested from c6–c8 levels of each tail, and 11 additional c7–8 discs were harvested for the second explant group (Fig. 1).

For 'motion segment' specimens, approximately $\frac{1}{3}$ of the proximal c6 and distal c7 vertebrae were removed in order to promote fluid flow while leaving enough bone to grip the specimens, which were potted in aluminum tubes using cyanoacrylate. Care was taken to insure axial alignment of the specimens within the pots, and that no adhesive covered either end of the vertebrae. Custom grips were used to load potted specimens in an environmental bath.

Disc 'explant' specimens (c7–8 with no endplates) were harvested using a scalpel by making transverse cuts through each endplate. A 5-mm biopsy punch and scalpel were then used to isolate the disc from the surrounding tissue. Care was taken to insure the disc remained completely frozen during dissection in order to preserve nucleus material. Disc height and diameter were measured using a dissecting microscope prior to mechanical testing. Explant specimens were placed in the test chamber between sintered steel platens with a permeability of $3.2 \times 10^{-10}\,\mathrm{m^4/N\,s}$ and pore size of $20\,\mu\mathrm{m}$ (Mott Metallurgical Corp., Farmington, CT) which is several orders of magnitude greater than that reported for native endplates (Setton et al., 1993; Ayotte et al., 2001). The second set of explants was tested using sintered steel porous platens that were modified to reduce the permeability without affecting surface roughness, by applying silicone sealant over the entire surface except for the area where the disc made contact. A permeability test demonstrated no fluid was transported through the modified platens indicating average platen permeability was reduced to zero.

'Single vertebra' specimens from c8 were harvested and, as with motion segments, approximately $\frac{1}{3}$ of the proximal end was removed and the remaining vertebrae was potted in an aluminum tube. The disc was dissected off of the distal end of the vertebrae with a scalpel,