IN SITU MEASUREMENT OF THE DYNAMIC MODULUS OF BOVINE HUMERAL HEAD ARTICULAR CARTILAGE UNDER PHYSIOLOGICAL CONTACT LOADING CONDITIONS

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INTRODUCTION
One of the unresolved questions in articular cartilage biomechanics is the magnitude of the dynamic modulus and tissue compressive strains under physiological loading conditions. While load magnitudes in various diarthrodial joints have been estimated with reasonable confidence for a number of activities of daily living, and while articular contact stresses have been measured with good accuracy under such load magnitudes in a number of studies, there are only a few reports of measurements of cartilage deformation under physiological conditions. Armstrong et al. [1] reported compressive strains of up to 20% under physiological loads in human cadaveric hip joints, using a radiographic technique; Macirowski et al. [2] demonstrated changes in cartilage thickness on the order of 10% or less using ultrasound measurements; in their *in vivo* study using magnetic resonance imaging, Eckstein et al. [3,4] reported a decrease of approximately 6% in cartilage volume following 50 deep knee bends, while the cadaveric study by Herbehold et al. [5] showed a progressive decrease in cartilage thickness over four hours by 57% under a constant load. In a rabbit model, Kaab et al. [6] reported reductions in cartilage thickness of 46% under static loading and 22-34% under dynamic loading, when compared to unloaded controls.

Knowledge of the physiological level of strain magnitudes and dynamic modulus is essential to our understanding of articular cartilage metabolism (catabolic and anabolic activities) in response to physical loading, and chondrocyte mechanotransduction. In the current study, intact bovine humeral heads were loaded cyclically at physiological load magnitudes while simultaneously measuring the cartilage deformation and contact area. The resulting stress strain response was used to characterize the effective dynamic modulus of the tissue under physiological contact stress magnitudes.

MATERIALS AND METHODS
Four bovine calf shoulder joints (ages 2-4 months) were obtained from a local abattoir and immediately dissected free from surrounding soft tissues. The proximal humeral head was resected, covered with PBS-soaked gauze and stored at -25 ^{o}C until ready for use. On the day of testing the humeral head was thawed in PBS at room temperature and resected at the articular margin to leave the hemispherical articular layer with its underlying bone. The dissected humeral head was then rigidly fixed to a circular aluminum plate which was connected to the loading actuator of a materials testing system (Instron, Model 8800, Canton, MA). Applied load and displacement was measured by instruments built into the Instron system, and compliance between the testing chamber and loading device was further assessed using a digital dial gage (Model Series 543, Mitutoyo, Singapore). The bottom center of the testing chamber consisted of a ∅50 mm lens against the articular surface of the specimen was pressed (Fig. 1). A mirror assembly allowed the undersurface of the lens to be viewed by a high resolution digital video camera (Model DCR-VX 1000, Sony, 720×480 pixels, 30 frame/s), see Fig. 2. The specimen was immersed in 200 ml PBS bath to which ~ 1ml India ink was added to provide a dark background that contrasted with the whitish cartilage surface in contact with the lens. A tare load of 5N was initially applied onto the sample to maintain proper loading platen contact and the specimen was allowed to equilibrate for 20 minutes. A cyclical compressive load was then applied onto the specimen using sinusoidal profile nominally varying in amplitude from 0 to 800N, for 5 cycles at 1Hz, above the initial tare load. The contact area of the specimen visible from the lens undersurface (Fig. 2) was segmented from the digital video frames using a combination of software packages (Adobe Photoshop, Version 5.5; NIH Image, Version 1.62; PV-Wave, Version 7.50, Visual Numerics). At the completion of loading, the specimen was allowed to recover for at least one hour and a ∅8 mm osteochondral plug was cored out from the center of the contact region to measurethe local cartilage thickness.

RESULTS
The dynamic response from a typical specimen is shown in the time domain in Figure 3. The applied load was divided by the measured contact area to obtain the average true contact stress over the contact region. The measured displacement was divided by the thickness to obtain an average engineering strain measure. In this study, we report

representative results from the third cycle of loading. When plotting the stress versus strain over that cycle, it was found that the loading phase was very linear, $r^2 = 0.999 \pm 0.0$ (Figure 4). The slope of this line was used to determine the effective compressive dynamic modulus at 1 Hz, 23.1±4.8 MPa; the area of the hysteresis loop was equivalent to a phase angle of 8.3 ± 1.0^o. The amplitudes of the applied stress and measured strain in the third cycle were 5.8±0.5 MPa and 26.7±5.4%.

DISCUSSION

The methodology adopted in this study allows the measurement of the time-dependent contact area of an intact articular layer against an optically clear, rigid surface. Along with the applied load, this measure yields the average value of the true contact stress over the contact region, without any intervening pressure sensor. The peak stress magnitude applied in this study, ~6 MPa, falls in the physiological range of what would be considered moderate loading in activities of daily living for human joints. The corresponding peak deformation of the articular layer reached 27% of its thickness. This range is comparable to that observed by Kaab et al. [6] in a rabbit model under dynamic loading. However, it is somewhat greater than the 20% reduction observed by Armstrong et al. [1] in a cadaver hip joint under constant loading, and significantly greater than the ~5% reduction in thickness observed in vivo by Eckstein et al. [3,4] in human subjects *after* the completion of an exercising regimen. These findings are not necessarily contradictory because of the differences in loading protocols. The apparent dynamic modulus of bovine humeral head cartilage measured at 1 Hz was found to be ~23 MPa, which is consistent with the results obtained from cylindrical explants tested in dynamic unconfined compression (e.g., [7]). In summary, this study provides novel experimental findings on the physiological strain magnitudes and dynamic modulus achieved in intact articular layers under cyclical loading conditions.

ACKNOWLEDGMENTS

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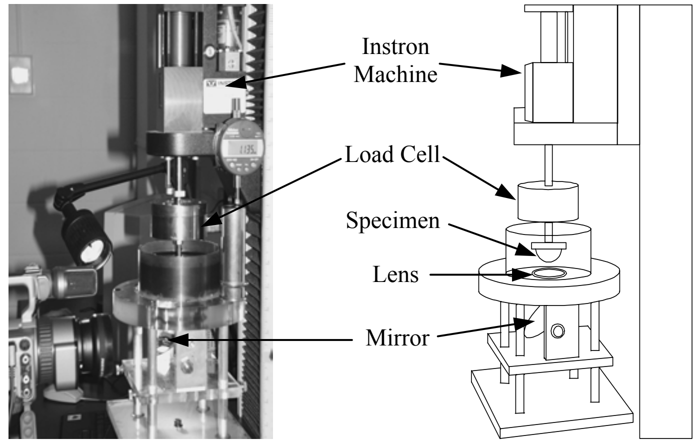

Figure 1 Testing apparatus

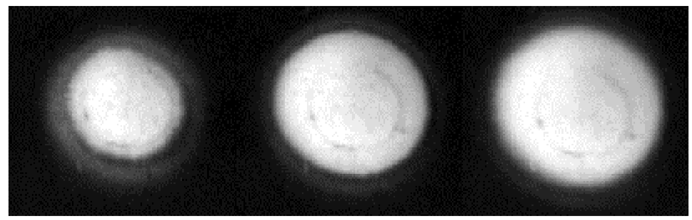

Figure 2 Contact area after 0s, 0.2s, and 0.5s, respectively

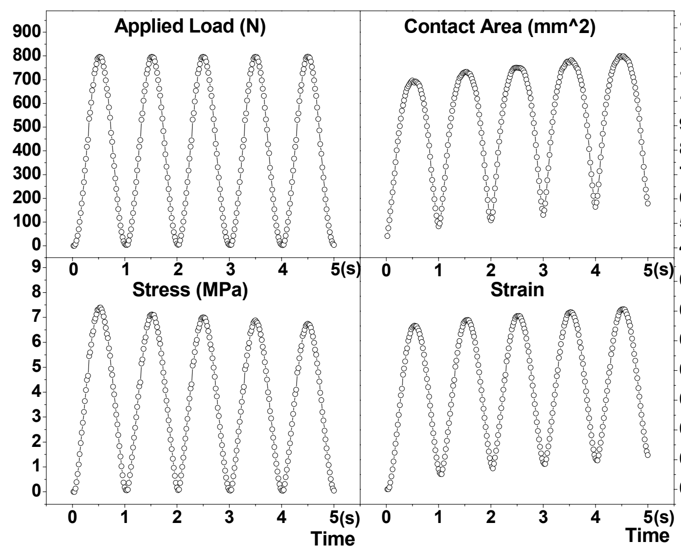

Figure 3 Dynamic response versus time for a typical specimen

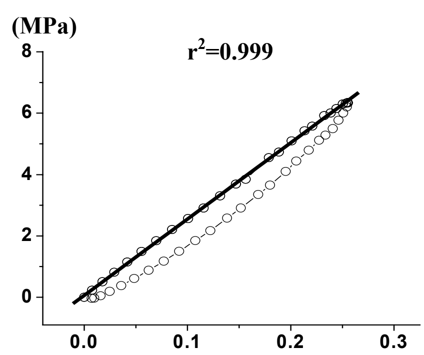

Figure 4 Stress-strain response for a typical specimen and linear curve fit of the loading data