INTRODUCTION
Anterior cruciate ligament (ACL) injury is a common cause for post-traumatic degenerative changes of articular cartilage that is characteristic of osteoarthritis (OA), thereby promoting ACL transection (ACLT) as a reliable method for inducing experimental OA [1]. In the present study, early OA changes were investigated in an ACLT rabbit model. The specific aim was to investigate the sensitivity of selected MRI techniques (Adiabatic T_{1ρ} & T_{2ρ}) [2], continuous wave (CW) T_{1ρ}, CPMG T_{2}, Adiabatic double echo (DE) T_{2}, T_{1} during saturation, and relaxation along fictitious field (RAFF) [3]) to early degenerative changes in articular cartilage following ACLT. For reference, biomechanical testing of the load bearing cartilage was conducted.

MATERIALS AND METHODS
ACLT was unilaterally induced in the knees of skeletally mature New Zealand White Rabbits (n = 8). The contralateral (CTRL) joints were used as a non-transected control group. Femoral condyles of the joints were harvested for investigation 4 weeks after ACLT. The procedure was carried out according to the guidelines of the Canadian Council on Animal Care and was approved by the committee on Animal Ethics at the University of Calgary.

MRI was performed at 9.4 T (Oxford instruments Plc, Witney, UK) with a 19 mm quadrature RF volume transceiver (RAPID Biomedical GmbH, Rimpar, Germany) and Varian DirectDrive console (Varian Inc. Palo Alto, CA, USA). After biomechanical testing, the samples were immersed in Fomblin® inside a Teflon™ test tube. The selected MRI parameters were measured using a global magnetization preparation block followed by a multi-slice fast spin echo readout (TR = 5 s, ETL = 4, TE_{sat} = 5 ms, 256x128 matrix size, slice thickness 1 mm and FOV = 16x16 mm). Two slices were acquired during the TR period immediately after the preparation; the slices were positioned at the lateral and medial condyles, respectively, including the site of biomechanical testing. The parameters for the preparation block for different contrasts were: (1) adiabatic T_{1ρ}: a train of 0, 4, 8, 12 and 24 adiabatic full-passage (AFP) pulses with duration of 4.5 ms per pulse (T_{eff} = 2.5 kHz), (2) adiabatic T_{2ρ}: a train of 0, 4, 8, 12 and 24 AFP pulses between adiabatic half-passage (AHP) pulses with duration of 4.5 ms per pulse (T_{eff} = 2.5 kHz), (3) CW T_{1ρ}: CW spin-lock pulses (β_{1L} = 1 kHz) with durations of 0, 10, 20, 40, 80 and 160 ms embedded between AHP pulses, (4) RAFF: a train of 0, 2, 4 and 6 RAFF pulses of 9 ms length [4], (5) T_{1} saturation: off-resonance irradiation with duration of 0.1, 0.3, 0.8, 2 and 4 s at 10 kHz offset (β_{1L} = 250 Hz), (6) Double echo T_{2} TE = 4, 8, 16, 32, 64, 128 ms, TR = 5 s, (7) CPMG T_{2}: TE = 4, 8, 16, 32, 64, 128 ms, TR = 5 s. Relaxation time maps were calculated using MATLAB. 3-mm wide full-thickness ROI was drawn to the site of biomechanical testing in both slices and the mean ± standard deviation (SD) of each parameter was determined. The full-thickness ROI was further split into superficial and deep halves.

The biomechanical properties of the samples were determined using indentation tests with a plane-ended indenter (dia. 1 mm). A stepwise stress-relaxation test was performed to determine the equilibrium elastic modulus of cartilage. Dynamic elastic modulus was determined from sinusoidal dynamic tests with a frequency of 1 Hz. The Poisson’s ratios for cartilage were assumed to be 0.1 and 0.5 in determination of equilibrium and dynamic modulus, respectively [5].

Statistical analysis of the MRI and biomechanical data was performed with Kruskal-Wallis post hoc test using IBM SPSS Statistics 19.0 (New York, NY, USA).

RESULTS
Adiabatic T_{1ρ}, CW T_{1ρ} and DE T_{2} parameters showed a significant elongation in the superficial half of the cartilage in the ACL transected lateral femoral condyle (Figure 1). Furthermore, a significant elongation in superficial adiabatic T_{1ρ} was seen in the medial compartment. For full thickness ROIs, statistically significant changes were observed in adiabatic T_{1ρ}, T_{2ρ} and CW T_{1ρ} between lateral condyles. The equilibrium elastic modulus, which is mostly affected by the proteoglycan content [6], was significantly reduced on the medial and lateral sides of the ACLT experimental joints as compared to the contralateral reference joints (Table 1). The dynamic elastic modulus that is sensitive to collagen integrity was also reduced at the lateral and medial compartments of the ACLT samples. Pooling all the samples, significant correlations were observed between MRI and biomechanical parameters. Most notably, for equilibrium modulus, the highest correlation was found with DE T_{2} (r = -0.465, p < 0.05) and for dynamic modulus with CW T_{1ρ} (r = -0.462, p < 0.01).

DISCUSSION
According to the present results rotating frame of reference (RFR) techniques are sensitive in detecting early cartilage degeneration, as detected by the slight yet significant change in the biomechanical properties. Moreover, RFR techniques appeared more sensitive in detecting early cartilage degeneration as compared to conventional techniques, namely T_{2} and MT. Our findings are in concordance with a previous study reporting that T_{1ρ} was superior to T_{2} in differentiating OA patients from healthy subjects [7]. These findings may be due to the increased sensitivity of RFR techniques to very slow molecular motion, which is altered during the course of degeneration of the extracellular matrix. Unexpected finding in the present study is the low sensitivity of RAFF to the changes in cartilage after ACLT. While tissue degradation was detected by both MRI and biomechanical testing, the MRI parameters were not strongly associated with mechanical properties in this model. In conclusion, the high sensitivity of RFR techniques to early cartilage degeneration encourages to further investigate their implementation for the detection of early OA.

REFERENCES