



# Overuse Following a Large Rotator Cuff Tear Alters Trabecular Bone Architecture but Not Glenoid Curvature in a Rat Model

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## Introduction

Rotator cuff tears are common shoulder injuries that cause significant pain and often require surgical intervention and physical therapy. Reestablishment of normal shoulder kinematics is often difficult and relies on balancing the active muscle forces and passive contact forces between the glenoid and the humeral head.<sup>1</sup> Previous studies have determined that damage to the rotator cuff tendons leads to alterations of subchondral mineralization patterns at the glenoid joint surface<sup>2,3</sup> with significant regional changes in articular cartilage thickness.<sup>4</sup> However, the influence of repetitive loading following a large rotator cuff tear on the glenoid surface geometry or underlying bony architecture has not been determined. Therefore, the purpose of this study was to utilize an established rat model<sup>5</sup> to analyze changes in the geometry of the glenoid articular cartilage and trabecular bone structure when a rat undergoes a supraspinatus and infraspinatus tear and is subsequently required to overuse the injured shoulder. We hypothesized that the presence of a rotator cuff tear will not affect the geometry of cartilage or underlying bony architecture in rats that are allowed to return to cage activity; however, repetitive overuse following the rotator cuff tear will result in significant changes of the geometry of articular cartilage surface and alterations in bony architecture.

## Methods

### *Experimental Design and Sample Preparation*

Twenty-three Sprague-Dawley rats were used in this study (IACUC approved). Four rats were used as controls (CTL) and the remaining animals were subjected to a 2 week training period followed by 4 weeks of overuse treadmill activity to create a tendinopathic condition as described.<sup>5</sup> A unilateral detachment of the supraspinatus and infraspinatus tendons was then surgically created to model an acute on chronic injury. Following surgery, animals were returned to cage activity (CA; n=10) or progressively returned to treadmill overuse (OV; n=9). All rats were sacrificed 8 weeks after surgery and frozen at  $-20^{\circ}\text{C}$  until testing.

### *Micro-CT Analysis*

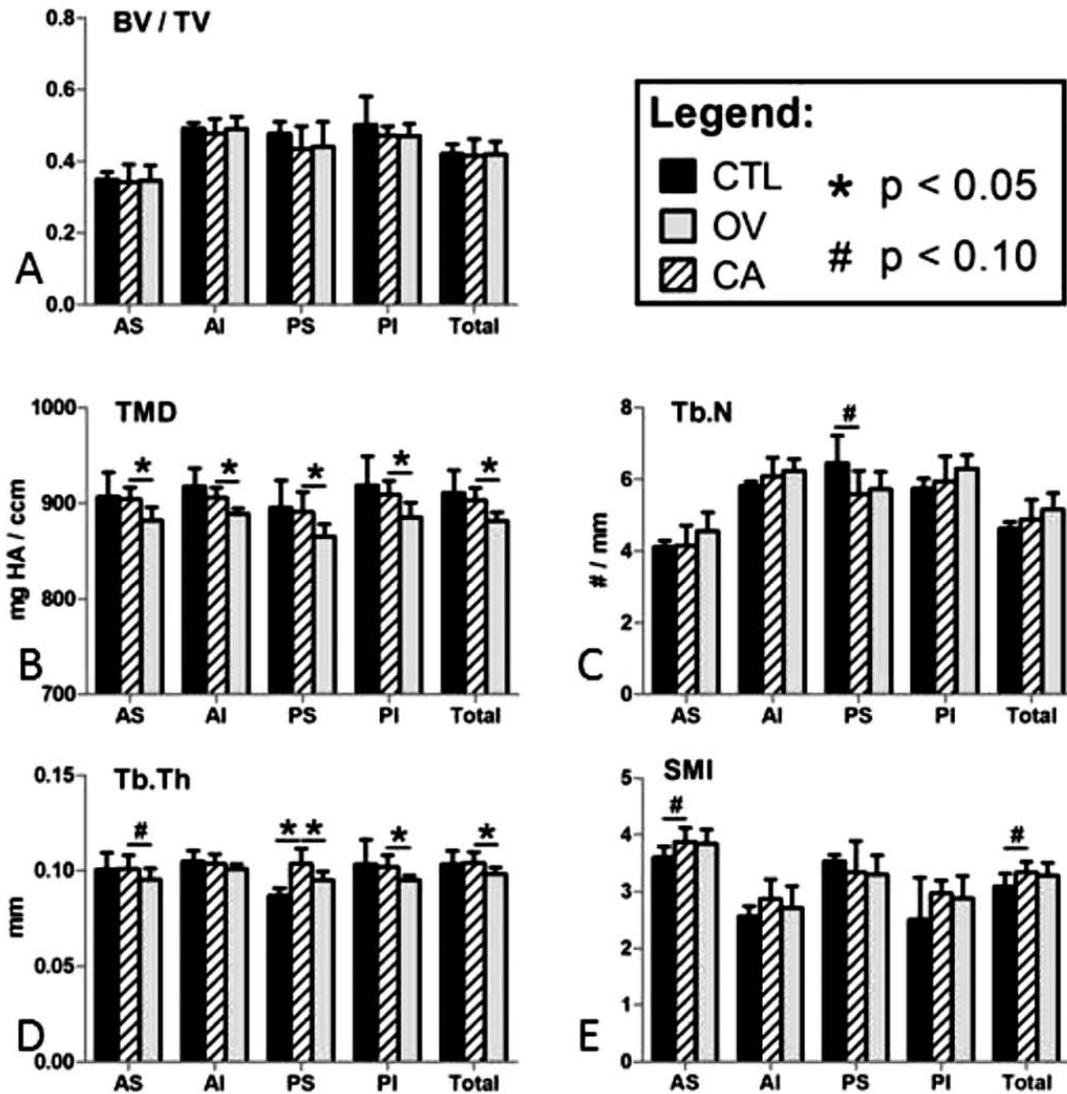
Following sacrifice, the left glenoid of each rat was carefully dissected free of all soft-tissue attachments and potted in a custom acrylic cylinder secured with polymethyl-methacrylate, leaving the glenoid fossa exposed. The glenoid cartilage was stained with Lugol's solution and each specimen was aligned such that the superior glenoid was parallel to the floor within a  $\mu\text{CT}$  system (VivaCT 40, Scanco Medical,  $10.5\ \mu\text{m}$  voxel size).  $\mu\text{CT}$  scan volumes were determined by choosing a point at the nadir of glenoid concavity and analyzing 100 slices above and below (for a total of 200 slices) the level of the subchondral bone. A rectangle was drawn to fit the glenoid at the nadir of concavity and sectioned into four equal quadrants for analysis of the anterosuperior (AS), anteroinferior (AI), posterosuperior (PS), and posteroinferior (PI) glenoid regions. Trabecular analysis was then completed for each of the 4 regions (AS, AI, PS, PI) individually, and as a whole (Total). Variables of interest include bone volume fraction (BV/TV), tissue mineral density (TMD), trabecular number (Tb.N), trabecular thickness (Tb.Th), and structural model index (SMI).

### *Glenoid Surface Geometry*

ITK-SNAP<sup>6</sup> and MeshLab<sup>7</sup> were used to create three-dimensional (3D) surface representations of the glenoid cartilage from  $\mu\text{CT}$  data. The 3D models were split into two separate surfaces to represent the bony and articular cartilage surfaces of the glenoid. A boolean function was employed in Meshlab to remove faces with normals that exceeded predetermined convexity inflection limits. The resulting bony and articular geometries were transformed into interpolated surfaces, and spheres were fit to the concave surfaces representing the four quadrants (AS, AI, PS, PI) and the entire glenoid (Total).

### **Statistics**

Statistical differences were evaluated with an ANOVA followed by planned comparisons between groups (CTL vs CA and CA vs OV to test study hypotheses) using two-tailed t-tests with significance set at  $p < 0.05$  and trends set at  $p < 0.10$ .



**Figure 1.** Trabecular analysis for bone volume fraction (A), tissue mineral density (B), trabecular number (C), trabecular thickness (D) and structural model index (E) in four quadrants of the glenoid (AS, AI, PS, PI) and as a whole (Total). Decreases in Tb.Th and TMD may indicate the degeneration of bone due to overuse after injury.

**Results**

**Trabecular Bone Measurements**

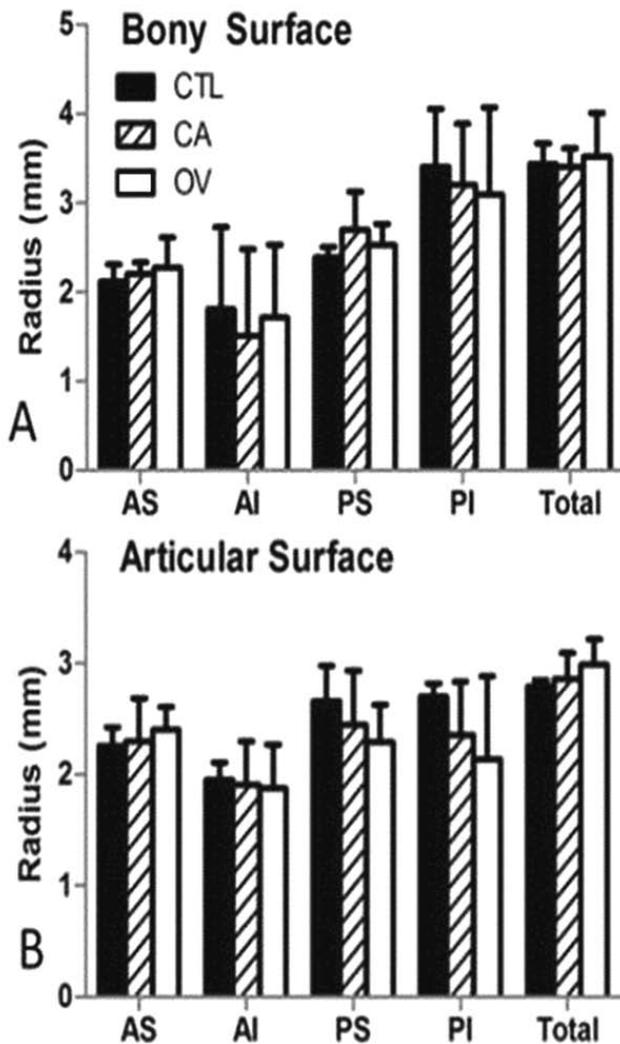
BV/TV (Figure 1A) and TMD (Figure 1B) of the trabecular bone stock in the glenoid were not affected by the presence of a rotator cuff tear. However, a decrease (trend) in Tb.N (Figure 1C) and an increase (significant) in Tb.Th (Figure 1D) in the PS region was seen. CA Rats also exhibited an increase (trend) in SMI in the AS region and in the overall bone volume (Figure 1E). The addition of overuse loading did not change BV/TV (Figure 1A), Tb.N. (Figure 1C), or SMI (Figure 1E). Significant decreases in TMD (Figure 1B) and Tb.Th (Figure 1D) were measured in rats that returned to overuse following injury.

**Geometric Measurements**

Surprisingly, there were no significant differences between groups for radii of curvature for the glenoid bone or cartilage surfaces (Figure 2).

**Discussion**

The results confirm our hypothesis that repetitive loading following rotator cuff rupture results in altered architectural properties of trabecular bone. Recent studies have related microstructural changes in trabecular bone to bone mineral density measurements as a surrogate for the “loading history” of the glenoid.<sup>2</sup> In the current study,  $\mu$ CT analysis has improved our ability to understand the adaptations of glenoid bone in response to rotator cuff tears and its response to subsequent rehabilitation protocols. Specifically, when comparing CTL to CA, it is notable that the PS region was the only area in which a decreasing trend in Tb.N was measured. Interestingly increases in Tb.Th were measured in the same region, which may be indicative of a compensatory mechanism taking place to account for losses in Tb.N. This change in bony architecture may be a result of the CA rats favoring the injured limb, and therefore decreasing the frequency and magnitude of ground reaction force vectors passing through this region. When



**Figure 2.** Sphere-fitting the concave regions of the bony (A) and articular cartilage (B) surfaces for four quadrants of the glenoid (AS, AI, PS, PI) and as a whole (Total). No changes were seen in bony or articular surface geometry as a result of injury or overuse.

comparing CA to OV, quantitative assessment of trabecular structures shows decreases in Tb.Th paired with decreases in TMD. This phenomenon is indicative of an undesirable loss

of mechanical strength of the bone structure in the OV rats, though this is yet to be confirmed by direct testing.

The results of this study did not support our hypothesis that overuse after injury would alter the geometry of the glenoid. We successfully fit spheres to surfaces representing the glenoid bone and articular cartilage, and no alterations in the radius of the bone or the articular cartilage were observed. While the overall shape of the bone and cartilage surfaces are unaffected by repetitive overuse, changes are clearly occurring in the underlying trabecular bone stock.

### Significance

Our study demonstrates that a return to activity after a large rotator cuff tear may be detrimental to the mechanical properties of the underlying trabecular microstructure, but the return to overuse did not have a significant effect on the glenoid surface curvature. This study has increased our knowledge of the effects of loading on the shoulder joint following a rotator cuff tear, but the longitudinal implications of such a scenario remains unclear. Follow-up *in vivo* studies that include mechanical testing at varied time points throughout a longer term will be necessary.

### Acknowledgments

This project was funded by the NIH/NIAMS.

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