Increased Synovial Inflammation and Tissue Degeneration Correlates with Tear Size of Diseased Rotator Cuff

INTRODUCTION:
The clinical outcomes of rotator cuff repair vary among patients and are affected by multiple factors including chronic inflammation. Although inflamed synovium is a common clinical observation at the time of surgery, the role of this inflammation plays on clinical outcomes is not well elucidated. Several recent studies have shown that there is elevation of pro-inflammatory cytokines (IL-1, IL-6, TNF-α, COX-2) and matrix metalloproteinases (MMP-1, MMP-13) in the synovium and torn tendon [1-6]. However, it is not clear from these studies whether such pro-inflammatory changes are affected by tear size (partial-thickness versus full-thickness). The objective of this study was to test the hypothesis that tear size would affect pro-inflammatory cytokines, angiogenesis factors, and tissue remodeling genes in the synovium, bursa, and torn supraspinatus tendon present at the time of surgery.

METHODS:
Twenty-four patients presented for arthroscopic rotator cuff repair (15 full, 9 partial) were prospectively enrolled. A small sample (~20 mm³) of synovium, bursa, torn supraspinatus tendon at the tear margin and the subcapsularis tendon as a control in the operated shoulder were collected according to IRB-approved protocols. Samples were taken under sterile conditions from the operating room, frozen, and later isolated for mRNA isolation using a RNeasy mini kit (Qiagen) before reverse transcribed into cDNA (iScript, BioRad). Real-time PCR was performed (Biorad) for selected pro-inflammatory factors (IL-1, IL-6, TNF-α, COX2), angiogenesis factors (VEGF) and tissue-remodeling genes (MMP-1, MMP-9, MMP-13, TIMP-1, COL1A1, type III collagen, SMA and Biglycan). GAPDH was used as a housekeeping gene for reference. All the primers were designed and tested for real-time PCR based on a human cell-line stimulated by IL-1. Statistical analysis was performed using Excel (Microsoft) or Systat (10.2, SPSS) to determine the relationship between rotator cuff tear size and the level of pro-inflammatory cytokines, remodeling genes, and angiogenesis factors by the linear correlation and Student’s t-test. The significance level was set at 0.05.

RESULTS:
H&E staining shows a decreased fibroblast population and less organized collagen fibrils in torn cuff of the FTh vs. PTh group (Figure 1). Increased pro-inflammatory cells, vascularity, surface irregularity and synovial thickening were found in the FTh group (Figure 1). The synovium in patients with full thickness rotator cuff tears had an increase of the IL-6 gene (p<0.05) when compared to the partial thickness group. A similar trend was also found in other pro-inflammatory cytokines including increased levels of IL-1 and COX2 (Figure 2). Full thickness tears also had increased levels of tissue remodeling genes including TIMP-1 (p<0.05) and COL1A1 (p<0.05) (Figure 3). Increased expression of torn supraspinatus tendon (Sup) were found in the Full-thickness group (Sup) as compared to that of Partial-thickness group using QPCR (*indicates p<0.05; ^ indicates 0.05<p<0.1). A decrease of iNOS and an increase of COX-2 expression of torn supraspinatus tendon (Sup) were found in the Full-thickness group.

DISCUSSION:
Our findings support our hypothesis that tear size can significantly affect pro-inflammatory genes (IL-6) as well as tissue remodeling (TIMP-1, MMP-1, and COL1A1) genes in the synovium from patients with rotator cuff tears in spite of a wide diversity of samples (gender, age, chronicity of tear, tear size of partial thickness.) Our findings also suggest that pro-inflammatory and neovascularization factors are closely associated with tendon remodeling and likely play a role in the pathogenesis of rotator cuff tears. This finding is consistent with previous studies [1, 2] that pro-inflammatory cytokines are closely related to angiogenesis and tissue remodeling in rotator cuff tendon. Further studies to determine the relationship between joint inflammation/tissue remodeling and clinical outcomes are warranted in order to elucidate the role inflammation has on the pathogenesis and repair of rotator cuff injury.

REFERENCES:

ACKNOWLEDGEMENTS:
This study was supported in part by Institutes of Sports Medicine and a NIH grant (AR50549). The authors thank Drs. Edward Craig, Frank Cordasco and Struan Coleman for their contribution.

Figure 1. H&E staining of supraspinatus, subscapularis, synovium and bursa with PTh and FTh tears.

Figure 2. Increased expression of pro-inflammatory genes (IL-6, TNF-α) and VEGF of synovium (Syn) in the Full-thickness group as compared to that of Partial-thickness group using QPCR (*indicates p<0.05; ^ indicates 0.05<p<0.1). A decrease of iNOS and an increase of COX-2 expression of torn supraspinatus tendon (Sup) were found in the Full-thickness group.

Figure 3. The expression of tissue remodeling genes in torn supraspinatus (Sup) and synovium (Syn). Increased tear size was positively correlated with significant overexpression of MMP-1 (Syn), MMP-13 (Sup) and Coll1 (Syn). (* for p<0.05; ^ for 0.05<p<0.1).

Figure 4 (A) A good correlation (R=0.90) between COL3A1 and Biglycan was found. (B) There was a good correlation between IL-18 versus MMP-9 in both FTh and PTh groups, though the correlation of pooled data was not significant (higher MMP-9 in FTh than PTh group).

Poster No. 539 • ORS 2011 Annual Meeting