**Introduction:** Subacromial bursitis can be a source of pain in patients with rotator cuff disease. The ability of the subacromial bursa to mediate tissue remodeling and matrix degradation in the subacromial bursa and adjacent rotator cuff tendon has not been previously investigated. In our previous study, we demonstrated the significant elevation in gene expression of many inflammatory cytokines and their receptors, indicating the presence of inflammation in bursitis. The purpose of the present study is to investigate the gene expression profile of extracellular matrix and adhesion molecules of bursa samples derived from the patients with impingement syndrome and compare it with that of normal bursa.

**Materials and Methods:** Subacromial bursa tissues were obtained intraoperatively from patients during shoulder surgery and analyzed using cDNA Array technique. 22 patients (mean age 53.2, range 19-80) were candidates for this study. They were divided into two groups: Group I; bursitis from 18 patients with impingement syndrome, Group II; normal bursa from 4 patients with instability or fracture. Samples were placed in RNA Later® solution (Ambion, Austin, TX) immediately after surgery and frozen at -70ºC within 2 hours of suspension. RNA was extracted from the samples using the RNeasy® Fibrous Tissue Mini Kit (Qiagen, Valencia, CA) and stored at -70ºC. Complementary DNA-array hybridization was done using GEArray™ Q Series Kits (SuperArray, Frederick, MD) for human extracellular matrix and adhesion molecules. The arrays were hybridized with Biotin-dUTP (Roche Applied Science, IN) labeled cDNA probes, which were prepared from the extracted RNA according to the manufacturer’s protocol. Images of the bursal specimens were obtained by chemiluminescent detection on X-ray films. Data analysis and normalization was accomplished using ScanAlyze and GEArray™ Analyzer software. Mann-Whitney U test was used for statistical analysis. The P values less than 0.05 were considered significant. Human tissue were obtained with the approval of the Columbia University School of Medicine Institutional Review Board.

**Results:**

Expression of MMP-12 is significantly high and expression of TSP-1 is significantly low in subacromial bursitis tissue.

1) cDNA array The mRNA levels for the extracellular matrix and adhesion molecules were screened in the bursal tissue. Out of 96 genes screened, 2 genes, including MMP-12 and TSP-1 showed a statistical change. Fig 1. shows typical patterns of the cDNA array in the bursitis and control groups. As shown in Fig.2, the gene expression of MMP-12 was increased in Group I (bursitis) compared to Group II (normal bursa) (p<0.05). The gene expression of TSP-1 was increased in Group II compared to Group I (p<0.05).

**Fig. 1.** X-ray films of chemiluminescent detection membrane for Human Extracellular Matrix & Adhesion Molecule Gene Array. ; MMP 12 = Matrix metalloproteinase 12; TSP-1 = Thrombospondin 1; GAPD = Glyceraldehyde-3-phosphate dehydrogenase.

**Fig. 2.** Average relative intensity of expression of extracellular matrix and adhesion molecule genes in subacromial bursa specimen.

2) Real-Time RT PCR 6 bursitis and 3 normal samples were analyzed by Real-Time RT PCR for confirming the cDNA array results. The expression pattern of MMP-12 and TSP-1 in Real-Time RT PCR is consistent with cDNA array results. Each sample was tested in duplicate and adjusted with GAPDH expression.

**Fig. 3.** The average expression level of MMP-12 and TSP-1 analyzed with Real-Time RT PCR.

**Discussion and Conclusions:** While inflammation in the subacromial bursa has been implicated in the pathophysiology of patients with impingement syndrome, the exact roles of extracellular matrix and adhesion molecules during this inflammatory process are currently unknown. MMP-12 is an extracellular matrix-degrading metalloelastase expressed primarily in tissue macrophages. A broad spectrum of extracellular matrix protein are shown to be degraded by MMP-12. MMP-12 gene expression has been shown to be elevated by inflammatory factors such as IL-1β and TNFα. Among a variety of MMPs screened, MMP-12 was the only one which was elevated significantly in bursitis.

TSP-1 is a metricellular protein known to promote chemotaxis of leukocytes to inflammatory sites. On the other hand, the functional roles of adhesion molecules are suggested in mediating extracellular signals through integrins. The TSP-1 expression was decreased in bursitis tissues compared to the control bursal tissue. The decreased TSP-1 expression may be associated with the activated destructive stage of the disease.

Matrix proteins that are degraded by MMP-12 are being analyzed in bursitis. Furthermore, the membrane receptor for TSP-1 and its signaling mechanism are being investigated. These initial data demonstrate that adhesion molecules and metalloproteases may play a significant role in subacromial bursitis and may mediate tissue remodeling and matrix degradation in the pathogenesis of rotator cuff disease.