Introduction
Subacromial impingement syndrome is a common clinical problem causing shoulder pain and disability. Inflammatory cytokines are identified in the subacromial space and the glenohumeral joint and are implicated in causing cytokine-induced inflammation (1). In bursal tissue of patients with impingement, a preponderance of inflammatory cells and increased vasularity is found, implying chronic inflammation (2, 3). These results suggest that the subacromial bursa is involved in the pathogenesis of impingement syndrome. Stromal cell-derived factor (CXCL12) is a potent chemotactic and angiogenic factor that has been proposed to play a role in the recruitment of inflammatory cells (4). The hypothesis of this study is that subacromial bursa becomes an inflammatory microenvironment with elevated expression of SDF-1. Furthermore, this chemokine can be regulated by NSAIDs and steroids.

Materials and Methods
1. 22 patients were included in this study; 18 patients with rotator cuff disease (bursitis) and 4 patients with anterior shoulder instability and proximal humeral fracture (control)  
2. cDNA array using GEAarray Series (Bioscience, MD) for human inflammatory cytokines and receptors were used for gene expression after RNA extraction. Statistical analysis was performed by Student t-test and Mann-Whitney nonparametric U test.  
3. Real-time RT PCR using SMART Cycler (Cepheid, Sunnyvale, CA) for SDF-1 with RNA obtained from 2 normal and 3 bursitis tissues.  
4. Bursal cells from 4 normal and 3 bursitis patients were isolated and cultured and stromal cells from 2 normal trabecular bone were cultured for ELISA study according to Institutional Review Board protocol.  
5. SDF-1a level in culture supernat was detected using ELISA kit (R&D Systems, MN) in 4 normal bursal, 3 bursitis and normal stromal bone cell lines (P2).  
6. The bursitis cell line was harvested in 1 day and 4 day culture each after treatment with COX-2 inhibitor (10µM) and Dexamethazone(10nM). Real-time RT PCR for SDF-1 with RNA obtained from cultured cells.

Results
Stromal Cell-Derived Factor 1 (CXCL12) is significantly elevated in bursitis.
1) cDNA array Fig.1 shows typical patterns of the cDNA array in the control and bursitis groups. A statistically significant difference (p=0.00401) was demonstrated for the stromal cell-derived factor 1 gene (SDF-1), as marked in Fig.1. The relative expression (in reference to the cDNA for GAPDH) was 0.421 in the bursitis vs. 0.0802 in the control bursa.

Figure 1. Average relative intensity of expression of SDF-1.(a). X-ray films of chemiluminescent detection membrane for Human Inflammatory Cytokine/receptor Gene Array (b).

2) Real-Time RT PCT The cDNA array results were confirmed by real-time RT PCR. The average expression level of SDF-1 (0.4033) in the bursitis group is higher than that of control (0.0389) as shown in Fig 2.

Figure 2. The average SDF-1 expression level of bursitis tissue is higher than that of control. Each sample was tested in duplicate and adjusted with GAPDH expression.

Conclusions & Discussion
The cDNA array analysis and real-time RT PCT of RNA from the bursa tissues showed a significantly higher expression of SDF-1 gene in bursitis lesions compared to the control tissue. Bursal cell cultures established from patients with subacromial bursitis over-expressed SDF-1 protein while SDF-1 expression was negligible in those from control bursa tissue. We believe that SDF-1 plays an important role in the pathogenesis of subacromial bursitis in the patient with impingement syndrome. Rheumatoid synovocytes are shown to overexpress SDF-1 which stimulates recruitment of monocytes (5,6). Therefore, SDF-1 overexpression by bursal cells may contribute to recruitment of monocytes and progression of inflammation in patients with subacromial bursitis. We demonstrated that SDF-1 m-RNA expression was decreased by the addition of COX-2 and dexamethasone to bursitis cultures. These results may explain one mechanism by which NSAIDs and steroids act to reduce inflammation in subacromial bursitis.

Reference