Chapter 6
Conclusions and Future perspectives
In this study, both transcriptomic and proteomic approaches were adopted to identify potential biomarkers for RA and OA that can assist in differential diagnosis as well as provide better insights into the disease pathogenesis.

Using single color whole genome oligonucleotide DNA microarrays (1x44), gene expression profiling of SFMCs obtained from 7 RA patients were carried out. SFMCs from OA patients did not yield enough RNA to carry out the microarray experiment. Hence this objective was not proceeded further.

Proteomic profiling of OA synovial fluid was carried out using high resolution mass spectrometry. 677 proteins from synovial fluid of patients with osteoarthritis were identified of which 545 proteins have not been previously reported in OA synovial fluid. These novel proteins included ADAM-like decysin 1 (ADAMDEC1), alanyl (membrane) aminopeptidase (ANPEP), CD84, fibulin 1 (FBLN1), matrix remodelling associated 5 (MXRA5), secreted phosphoprotein 2 (SPP2) and spondin 2 (SPON2). 300 proteins were identified using lectin affinity chromatography, including the glycoproteins afamin (AFM), attractin (ATRN), fibrillin 1 (FBN1), transferrin (TF), tissue inhibitor of metalloproteinase 1 (TIMP1) and vasorin (VSN). Gene ontology analyses confirmed that a majority of the identified proteins were extracellular and are mostly involved in cell communication and signaling. The expression of ANPEP, dickkopf WNT signaling pathway inhibitor 3 (DKK3) and osteoglycin (OGN) were also confirmed by MRM analysis of OA synovial fluid samples. This is the largest catalog of proteins reported in synovial fluid so far. Some of these identified proteins can be further evaluated for their potential as specific targets or useful biomarkers for OA. These proteins could further enhance our knowledge and provide better insights regarding the underlying mechanism of OA pathogenesis perhaps leading to better therapeutic strategies.
In the quantitative proteomics study, 575 proteins were identified, out of which 135 proteins were found to be differentially expressed by ≥3-fold in the synovial fluid of RA and OA patients. Proteins not previously reported to be associated with RA including, coronin-1A (CORO1A), fibrinogen like-2 (FGL2), and macrophage capping protein (CAPG) were found to be upregulated in RA synovial fluid. Proteins such as CD5 molecule-like protein (CD5L), soluble scavenger receptor cysteine-rich domain-containing protein (SSC5D), and TTK protein kinase (TTK) were found to be upregulated in the synovial fluid of OA patients. The upregulation of CAPG in RA synovial fluid samples were also confirmed by MRM assay as well as by Western blot. Pathway analysis of differentially expressed proteins revealed a significant enrichment of genes involved in glycolytic pathway in rheumatoid arthritis. Although the above-mentioned novel proteins have been observed to be differentially expressed in the synovial fluid of RA and OA patients, it is possible that they are not really associated with the disease but rather contributed by either individual variation or disease conditions other than arthritis in these patients. So, it becomes necessary to perform validation studies in a larger cohort of samples. In addition, it will be useful to determine if these proteins are detected in synovial or cartilage tissues to understand their association with RA and OA more completely.