

Angiogenesis in Rheumatoid Arthritis: VEGF expression in Synovial Fluid.

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Introduction. The expansion of synovial epithelium in Rheumatoid Arthritis (RA), and the subsequent pannus invasion of underlying cartilage and bone, needs an increase in the vascular supply to the synovium. Angiogenesis is recognised as a key event in the formation and maintenance of the pannus in RA. The pro-angiogenic cytokine, vascular endothelial growth factor (VEGF), has been demonstrated to have a central involvement in the angiogenic process in RA. Therefore we evaluated gene expression levels of a set of genes associated with angiogenesis in Synovial Fluid (SF) from RA patients compared with osteoarthritis patients.

Patients and Methods: Knee SF samples were classified in two groups, Group I: RA with DAS 28 score > 5.1, high disease activity (n=10, 7F/3M, age: 56,3 ± 20,9, range: 17-84) and Group II: Osteoarthritis (OA, n=18, 13F/5M, age: 70,5 ± 6,6, range: 58-86). Levels of gene expression of three genes previously associated with angiogenesis, including pro-angiogenic factors: VEGF and Angiopoietin 1 (ANGPT-1) and anti-angiogenic factor, Thrombospondin I (TSP1) were evaluated using Quantitative Real Time PCR (QPCR). All amplifications were carried out in duplicate and threshold cycle (C_t) scores were averaged for calculations of relative expression values. The C_t scores were normalized against C_t scores by subtracting the corresponding β 2Microglobuline (β 2M) control, or $\Delta C_t = C_{t, gene} - C_{t, \beta 2M}$. To test for differential gene expression between groups, a two sample t-test was performed to compare the ΔC_t in the two groups.

Results: Gene expression level is showed in table 1.

Gene	Group I (RA)	Group II (OA)	p value
VEGF (mean ΔC_t)	4,95	9,09	p < 0,01
TSP-1 (mean ΔC_t)	8,68	5,18	p = 0,05
ANGPT1(mean ΔC_t)	18,89	14,93	p = NS

ΔC_t is inversely proportional to the gene expression.

After t-test, we observed a significant difference for VEGF (p < 0,01) and TSP-1 (p=0,05) gene expression level between groups. There is no significant difference in ANGPT1 expression. We evaluated data from ΔC_t analysis observing that the levels of mRNA of VEGF in Group I were higher than those from Group II and TSP-1 levels were lower than those from Group II.

Conclusions. In the present cross-sectional study, increased levels of VEGF and decreased levels of TSP-1 were observed in RA patients. These gene expression results might be associated with increased angiogenesis in RA patients with high disease activity.

Reference.

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- 2- Afuwape AO et al. The role of the angiogenic molecule VEGF in the pathogenesis of rheumatoid arthritis. Histol Histopathol 2002, 17: 961-972.