Angiogenesis in Rheumatoid Arthritis


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Rheumatoid arthritis (RA) is a large social and economic burden in the United States with an estimated annual cost of 9 billion dollars. The average annual cost per patient is $19,000, including the indirect costs of loss of productivity. So it is a major health problem.

The histology of normal synovium shows that it is very loose tissue. Rheumatoid synovium is much denser, because it is packed with blood vessels often surrounded by leukocytes. Important stimulators of angiogenesis in RA include vascular endothelial growth factor (VEGF) and several proangiogenic cytokines such as interleukin-8, tumor necrosis factor-α, and members of the CXC chemokine family. VEGF-A, -C, and –D have been identified in joints from RA patients and high serum levels of VEGF or FGF-2 are correlated with poor prognosis. Induction of VEGF may be by transforming growth factor-β (TGF-β), IL-1 and hypoxia in RA joints. In a rodent model of RA, preventive administration of fumagillin (TNP-470) was found to reduce serum VEGF and vascularity in the synovium. In mouse collagen-induced arthritis, soluble VEGFR1 or anti-VEGFR1 reduce inflammation and new vessels in joints.

Cultured human synovial fibroblasts have been used to study how VEGF expression might be regulated in joints. IL-1, TNF-α, and TGF-β each increase VEGF expression by synovial fibroblasts.

Three independent studies have shown that antagonists of αvβ3 integrin attenuate joint inflammation and neovascularization in various models. A proapoptotic αvβ3 antagonist composed of an RGD peptide linked to a heptapeptide dimer selectively homes to mouse joints with inflammatory arthritis and not normal joints and provides therapeutic benefit.

It had been hypothesized that soluble adhesion molecules such as sE-selectin might bind leukocytes and reduce inflammation in joints; however it was found that soluble E-selectin and soluble vascular cell adhesion molecule-1 each stimulate angiogenesis in joints; sE-selectin acts through sialyl Lewisx (Lex) on endothelial cells and sVCAM-1 does so through VLA-4.

Cells were isolated from the joints of RA patients and only a subset of macrophages was found to promote angiogenesis. Monoclonal antibodies (mAbs) to the cells were generated and many were found that only recognized macrophages, but one recognized only endothelium and epithelium. Further characterization showed that it reacted with select endothelium in synovium, skin, and lymph nodes, but did not detect myeloid cells. The antigen, 4A11, is up-regulated on cultured endothelium by cytokines, such as TNFα, within minutes, but by 2 hours it is back to baseline. In vivo, there is some basal expression on endothelium in skin, and expression is increased 6-24 hours after administration of uroshiol, poison ivy extract. Compared to normal synovium, 4A11 is more highly expressed in RA synovium. It was found that mAb 4A11 recognizes the related glycoconjugates Lewisx (Lex) and H-2.
Lewis glycoconjugates, Le\(^x\), Le\(^y\), and H-2 constitute the chemical basis for several blood group systems, but they also have been implicated in other processes. They are expressed by some carcinomas and have prognostic significance. They are expressed in gastric mucin and by *Helicobacter pylori*, constituting molecular mimicry that can lead to autoimmune gastritis if antibodies generated against *H. pylori* also recognize gastric mucin.

Soluble forms of the antigens, H-2-glucose (H-2g) and Le\(^y\)-glucose (Le\(^y\)g), which are critical components of soluble 4A11, were tested for angiogenic activity. They stimulated chemotaxis of cultured endothelial cells and stimulated angiogenesis in corneal pocket assays, which was blocked by mAb 4A11. Synovial fluid from patients with RA were found to have substantially higher levels of soluble 4A11 than synovial fluid from patients with osteoarthritis. So, soluble 4A11 reacts with an unknown receptor on endothelial cells to stimulate angiogenesis and is found in high levels in the joints of patients with RA. Soluble 4A11 is also pro-inflammatory; it is highly potent in recruitment of leukocytes after intraperitoneal injection. It also mediates adhesion by upregulating ICAM-1 on endothelium. It signals in endothelium via a JAK-STAT pathway. It seems to be involved in joint-specific homing.

The action of the pro-angiogenic agents in joints is balanced by inhibitors. The inflamed synovium is adjacent to avascular cartilage. Marsha Moses has shown that a cartilage-derived factor Troponin-I has antiangiogenic activity. In collaboration with Olga Volpert and Noel Bouck, it was noted that IL-4 is strongly angiostatic in the cornea and this effect could be blocked by antibody to IL-4. Adenoviral vector (Ad)-mediated gene transfer of IL-4 to the ankle joint of rats was followed by *M. batrycium* injection, which causes arthritis. Compared to null vector-injected joints, those injected with AdIL-4 showed less swelling and inflammation. Gene transfer to joints of thrombospondin, angiostatin, endostatin, and troponin-I each cause significant benefit in arthritis models.