EDITORIAL

The role of monocytes-macrophages in vasculogenesis in multiple myeloma

Inflammatory cells regulate endothelial cell functions related to physiological angiogenesis as well as inflammatory and tumor-associated angiogenesis. It was Rudolf Virchow in 1863, who critically recognized the presence of inflammatory cells infiltrating neoplastic tissues, and first established a causative connection between the 'lymphoreticular infiltrate' at sites of chronic inflammation and cancer. In neoplastic tissues, inflammatory cells act in concert with tumor cells, stromal cells and endothelial cells to create a microenvironment that is critical for the survival, development and diffusion of the neoplastic mass. These synergies may represent important mechanisms for tumor development and metastasis by providing an efficient vascular supply and an easy escape pathway.

Cells belonging to the monocyte-macrophage lineage are a major component of the leukocyte infiltration in tumors and there is growing evidence that they are part of inflammatory circuits that promote tumor progression, and favor invasion and metastasis.[1,2] The stimulating effect exerted by tumor-associated macrophages on the growth of the tumor mass is partly related to the angiogenic potential of these cells. Tumor-associated macrophages are a rich source of potent proangiogenic cytokines and growth factors, such as vascular endothelial growth factor, tumor necrosis factor alpha, interleukin-8 and fibroblast growth factor-2. They also express a broad array of angiogenesis-modulating enzymes, including matrix metalloproteinases (MMPs)-2, -7, -9, and -12 (MMP-2, MMP-7, MMP-9 and MMP-12, respectively), cycloxygenase-2 and colony-stimulating factor-1.[3–7] The many proangiogenic factors secreted by macrophages may promote tumor spread and help to explain the correlation between increased tumor-associated macrophage density and the augmented tumor vasculature recognized during experimental and human carcinogenesis. Moreover, macrophages take part in neovascularization by 'drilling' tunnels for new vasculature, producing tubular destruction of the matrix, distributing to form columns and capillary-like structures containing erythrocytes,[8] localizing in microvessels embedded in bundles of fibrillar collagen[9] and adhering to injured vessel walls, thus accelerating re-endothelization of the vascular barrier.[10]

In multiple myeloma (MM), bone marrow stromal cells increase the concentration of angiogenic factors and matrix-degrading enzymes in the bone marrow microenvironment by direct secretion, or by stimulation of myeloma cells or endothelial cells through paracrine interactions.[11–13] Bone marrow stromal cells, osteoclasts, osteoblasts and endothelial cells secrete several factors, including vascular endothelial growth factor, fibroblast growth factor-2, tumor necrosis factor alpha, interleukin-6, B-cell activating factor, stromal cell-derived factor 1α (also known as CXCL12) and various Notch family members, which are further upregulated by tumor cell adhesion to extracellular matrix proteins and/or bone marrow stromal cells.[14] Bone marrow stromal cells and other accessory cells supporting MM cell survival in the bone microenvironment constitute potential therapeutic targets.

We have recently shown that when bone marrow macrophages from MM patients are exposed to vascular endothelial growth factor and fibroblast growth factor-2, which are major angiogenic cytokines secreted by plasma cells,[15,16] and present in the bone marrow microenvironment at four- to five-fold higher levels than in peripheral blood,[17] they transform into cells functionally and phenotypically similar to paired MM endothelial cells, and generate capillary-like networks mimicking those of MM endothelial cells.[18] By contrast, macrophages from non-active MM, monoclonal gammopathies of undetermined significance and benign anemia patients display similar, albeit weaker features. Endothelial cell-like macrophages and apparently typical macrophages contribute sizeably to the formation of the neovessel wall in patients with active MM, whereas their vascular supply is minimal in non-active MM, and absent in MGUS patients and control patients.[18] In patients with active MM, fluorescence-activated cell sorting (FACS) analyses on freshly isolated bone marrow mononuclear cells showed higher percentages of CD14/CD68 double-positive cells than in those with non-active disease and MGUS. Furthermore, in active MM patients, bone marrow biopsies displayed macrophages with both endothelial cell-like (that is, CD68/FVIII-RA double positive) and apparently typical (that is, CD68 positive/FVIII-RA negative) features located in the microvessel wall and collaborating with MM endothelial cells to line the vessel lumen. Figures of this type were rare in non-active MM patients and absent in MGUS. Thus, macrophage involvement in the vasculogenic pathway proceeds in step with MM activity, and with progression of plasma cell tumors as well.[18] Overall, these data suggest that in active MM, macrophages contribute to neovascularization through a vasculogenic pathway.

Chen *et al.*[19] showed that monocytes induces vascular endothelial cell gene expression and develop tube-like structures when they are cultured with bone marrow from patients with MM that express an angiogenic factor, namely, pleiotrophin, effect specifically blocked with antibodies anti-pleiotrophin. Moreover, when co-injected with human MM cells into SCID mice, green fluorescent protein-marked human monocytes were found incorporated into tumor blood vessels and expressed human vascular endothelial cell protein markers and genes that were blocked by anti-pleiotrophin antibodies. These results suggest that vasculogenesis in human MM may develop from tumoral production of pleiotrophin, which induces the transdifferentiation of monocytes into vascular endothelial cells.

During embryo life, blood vessels first seem as the result of vasculogenesis, that is, the formation of capillaries from endothelial cells differentiating *in situ* from groups of mesodermal cells. The primitive heart and primitive vascular plexus form in this way.[20] Remodeling of the primary vascular plexus into a more mature vascular system is thought to occur by angiogenesis, a term applied to the formation of capillaries from existing vessels, that is, capillaries and post-capillary venules,[21] on the basis of endothelial sprouting or intussusceptive (non-sprouting) microvascular growth.[21] In tumors, near angiogenesis, vasculogenesis contributes to the formation of the microvascular plexus that is important for local and systemic diffusion.[22]
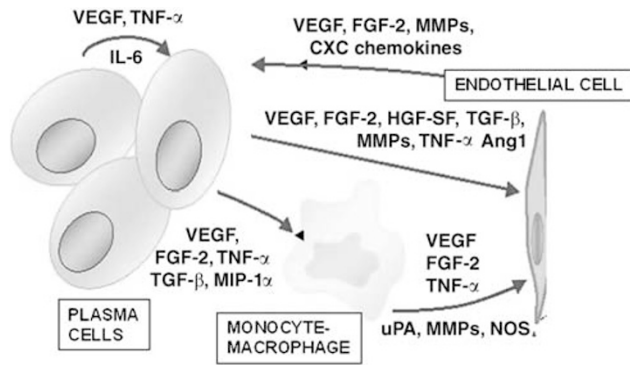
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Figure 1 Interplay between plasma cells, monocytes-macrophages and endothelial cells in multiple myeloma. VEGF, vascular endothelial growth factor; MMPs, matrix metalloproteinases; TNF-α, tumor necrosis factor alpha; TGF-β, transforming growth factor beta; IL-6, interleukin-6; FGF-2, fibroblast growth factor-2. Ang 1, angiopoietin 1; CXC, two N-terminal cysteines of chemokines separated by one amino acid (X); uPA, urokinase plasminogen activator; HCG-SF, hepatocyte growth factor-scatter factor; MIP-1α, macrophage inflammatory protein 1 alpha; NOS, nitric oxide synthase.

Experimental data reported here documented that in MM, in association to angiogenesis,[23] vasculogenesis also occurs and monocyte-macrophages participate actively in this process (Figure 1), together with hematopoietic stem and progenitor cells, as we have recently shown.[24]

The ability of tumor to support continued growth through non-angiogenesis-dependent pathways represents an important mechanism of tumor resistance to conventional anti-angiogenic treatment, and requires adjustment of these therapies to counter such new pathways.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgements

D Ribatti[1] and A Vacca[2]
[1]Department of Human Anatomy and Histology, University of Bari Medical School, Bari, Italy and
[2]Department of Internal Medicine and Clinical Oncology, University of Bari Medical School, Bari, Italy
E-mail: ribatti@anatomia.uniba.it

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