[Frontiers in Bioscience 10, 666-674, January 1, 2005]

ANGIOPOIETIN/TIE2 SIGNALING, TUMOR ANGIOGENESIS AND INFLAMMATORY DISEASES

Hanako Kobayashi [1] and P. Charles Lin [1,2 3]

[1] Department of Radiation Oncology,[2] Department of Cancer Biology, Department of Cell and [3] Developmental Biology, Vanderbilt-Ingram Cancer Center, Vanderbilt University Medical Center, Nashville, TN 37232

TABLE OF CONTENTS

1. ABSTRACT

Mounting evidence demonstrates that the formation of new blood vessels, termed angiogenesis, plays critical roles in human disease development and progression. Based on these findings, there has been a tremendous effort to investigate the molecular mechanisms that drive blood vessel growth in adult tissues. Compared to physiological angiogenesis, inflammation is often accompanied with pathological angiogenesis and often is the underlying causes of many diseases such as cancer, arthritis, atherosclerosis, and others. Inflammation induces angiogenesis and reciprocally, angiogenesis facilitate inflammation. A study of the interaction between angiogenesis and inflammation will enhance our understanding of the mechanisms of diseases. It may generate novel approaches for therapy. Tie2 was recently identified as a receptor tyrosine kinase expressed principally on vascular endothelium, making it an attractive molecular target for angiogenic therapy. This review discusses the regulation of Tie2 and its angiopoietin ligand family in inflammation-associated angiogenesis focusing on cancer, arthritis, and atherosclerosis. The complexity of angiogenesis and context–dependent regulation of angiopoietin/Tie2 signaling in angiogenesis requires further studies.

2. INTRODUCTION

Angiogenesis is the process by which blood vessels are assembled and remodeled to form functional vascular networks. A large body of evidence has shown that angiogenesis is a crucial event for embryonic development. In adulthood, the vasculature is notably quiescent, but angiogenesis is required for normal female reproductive function and required for wound healing. Importantly, however, angiogenesis is reactivated in pathological conditions and contributes to the pathogenesis of a number of diseases including cancer, arthritis, obesity, atherosclerosis and two common causes of blindness, diabetic retinopathy and macular degeneration (28). Conversely, other diseases such as coronary artery disease and peripheral vascular disease are characterized by failure of the compensatory angiogenic response. Involvement of angiogenesis, or failure of angiogenesis, in these important diseases has led to a tremendous effort to define the molecular mechanisms that drive blood vessel assembly and remodeling.

One of the major differences between physiological and pathological angiogenesis is the presence of inflammation, which commonly accompanies pathological angiogenesis. Tissue injury induces inflammation, a fundamental host defense mechanism. There are growing evidence indicating that inflammation triggers angiogenesis (15, 16). Conversely, angiogenesis facilitates inflammation by supplying inflammatory cells, cytokines and nutrients. An increase in angiogenesis in chronic inflammatory disorders such as arthritis, chronic airway inflammation, gastrointestinal ulceration, and arteriosclerosis often leads to undesired outcomes. A study of the molecular mechanism of pathological angiogenesis offers great potential in understanding the disease mechanisms as well as the development of therapeutic interventions. This review discusses current knowledge of inflammation-associated angiogenesis, and focuses on the regulation of the Tie 2 receptor and its ligands, angiopoietins, in cancer, arthritis and atherosclerosis development. Much of the angiogenesis work regarding the Tie2/angiopoietin signal transduction in adult has been in cancer studies, with limited research in other inflammatory related diseases such as arthritis and atherosclerosis. It is uncertain whether the process in tumor angiogenesis and that in other inflammation-

associated angiogenesis is the same or not; thus, extrapolation of data from cancer research into other inflammatory diseases requires caution.

3. TIE2 RECEPTOR AND ANGIOPOIETINS

Tie receptors, including Tie1 and Tie2, were originally described as members of an orphan receptor tyrosine kinase (RTK) subfamily expressed predominantly in the embryonic endothelium (22, 38, 52, 67, 68, 70, 105). Tie2 was found to be highly conserved across vertebrate species predicting the importance of its biological function. In fact, the domain structure of Tie2 is highly conserved from zebrafish to human, with the greatest amino acid homology occurring in the kinase domain (50). Consistent with its expression pattern, disruption of the function of Tie2 in transgenic mice resulted in early embryonic lethality secondary to vascular abnormalities (21, 69). Tie2 null embryos showed a decreased number of endothelial cells and decreased contact between endothelial cells and the underlying perivascular cells (pericytes or smooth muscle cells) suggesting a role in the maturation and stabilization of the embryonic vasculature.

Tie2 was deorphanized with the discovery of the ligands, angiopoietins (Ang), which include Ang1 and Ang2. As is the case with other RTKs, Ang1 binding stimulated autophosphorylation of the kinase domain of Tie2. However, unlike activation of most growth factor RTKs, Ang1 activation of Tie2 did not stimulate mitogenesis suggesting a novel role in endothelial biology. In contrast to Ang1, Ang2 did not stimulate Tie2 autophosphorylation but instead blocked Ang1-mediated Tie2 activation and endothelial migration suggesting that Ang2 was a naturally occurring inhibitor of Tie2 activation (53, 88, 97). Consistent with this finding, mice lacking functional Ang1 expression and mice overexpressing Ang2 both displayed a phenotype similar to Tie2 null mice (53, 82). However, there are reports showing Ang2 as a Tie2 agonist, suggesting that the action of Ang2 as a Tie2 agonist or antagonist is context-dependent (12, 29, 40, 88). More recently, two other Tie2 ligands have been identified: mouse Ang3 and human Ang4 (95). Taken together, these findings suggest the fine regulation of angiopoietin/Tie2 function is crucial in the remodeling and maturation/stabilization of blood vessels.

4. ANGIOPOIETIN/TIE2 SIGNALING AND INFLAMMATION

An important step of the inflammation process is the passage of the plasma protein and leukocytes to the targeted tissues, a process regulated by tight junctions and the adherence junctions in blood vessels. Inflammatory cytokines induces vessel dilation and increases vessel permeability. Interestingly, Ang1 protects vessel leakage caused by the inflammatory cytokine treatment, suggesting Ang1 may function as an anti-inflammatory agent (30, 89, 90). This inhibition of leakage or permeability is speculated to be due to the regulation of platelet endothelial cell adhesion molecule-1 (PECAM-1), E-selectin, and vascular endothelial Cadherin (30) by Ang1. Furthermore, Ang1 was reported to inhibit VEGF-induced proinflammatory adhesion molecule expression: including intercellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1), and E-selectin (41). Not surprisingly, Ang1 also blocked vascular endothelial growth factor (VEGF)-induced vessel permeability and leukocytes adhesion to vascular endothelial cells (41).

There is growing evidence indicating that Ang1 protects vessel leakage in different tissues that is essential to maintain a proper function of the organ. In testis, resistance to inflammation mediators in testicular microvasculature was suggested to be due to the constitutively local expression of Ang1 (32). In lungs, down-regulation of Ang1 and Ang4 by lipopolysaccharide (LPS) was suggested to be the cause of vascular leakage in the development of acute lung injury (ALI) (39). Conversely, decreasing Ang1 bioavailability by using soluble Tie2 receptors (ExTek) increased local inflammation and leukocytes infiltration in tumors, suggesting Ang1 is an anti-inflammatory cytokine (55). Collectively, this anti-leakage action of Ang1 may be advantageous to control chronic inflammation. In addition to the anti-leakage action, Ang1 was shown to regulate tissue factor (TF) expression. TF is an important cytokine that is responsible for blood coagulation and involved in thrombosis and inflammation associated with sepsis, atherosclerosis, and cancer (59). Tumor necrosis factor-alpha (TNF-alpha), a major proinflammatory cytokine, induces TF expression in endothelial cells, and very interestingly Ang1 was shown to inhibit the TF expression induced by TNF-alpha (42). This further demonstrates the critical role of angiopoietin/Tie2 signaling in inflammation and inflammation related disease development.

Studies begin to uncover the molecular mechanisms for the involvement of Ang1 in inflammatory processes. A recent study identified ETS transcription factor family member ESE-1 as capable of transactivating the Ang1 promoter and suggested ETS factor ESE-1 is responsible for the induction of Ang1 in the process of inflammation (5). Over-expression of ESE-1 in cells induced endogenous Ang1 gene expression, which supports a mechanism for the ETS factor ESE-1 as a transcriptional mediator of angiogenesis in the setting of inflammation (5). In another study, Tie2 was shown to directly interact with a potent regulator of inflammatory gene expression, A20 binding inhibitor of NF-? B activation-2 (ABIN-2), linking the function of Ang1 with NF-? B transcription factor and subsequent expression of many NF-? B-responsive genes (36).

In addition to Ang1, studies also showed that phorbol-myristate-acetate (PMA), an inflammatory agent, increased Ang2 expression in endometrial endothelial cells (44). Ang2 was found to be stored in Weibel-Palade bodies, suggesting a possible role of Ang2 in an inflammation process since molecules stored there control rapid vascular responses related to coagulation and inflammation (27). Recently we and others have demonstrated that TNF-alpha induces Tie2 expression via NF−κB transcription factor (19, 96). We have also

identified a dual functional role of the angiopoietin/Tie2 signaling in inflammation related angiogenesis: Tie2 activation enhanced low doses of TNF-alpha-induced angiogenesis, and attenuated high-doses of TNF-alpha-induced cell death (12). Taken together, these findings support a role of angiopoietin/Tie2 signaling in inflammation related angiogenesis and disease progression.

5.ANGIOPOIETIN/TIE2 SIGNALING AND CANCER

The chronic injury and irritation by the infectious or non-infectious agents initiates an inflammatory response. Besides tumor cells and endothelial cells, lymphocytes, macrophages, mast cells and neutrophils are often present in tumors. It is now becoming clear that the tumor microenvironment, which to a large extent is orchestrated by infiltrating cells, is an indispensable participant in the neoplastic process, fostering proliferation, survival and metastasis. These cells produce an environment and promote tumor angiogenesis and tumor development by supplying proangiogenic growth factors, cytokines and proteases (15) (16) (62) (92). Furthermore, a subsequent free radical release by recruited-leukocytes can damage healthy neighboring cells (37). The infiltrated inflammatory cells promote the formation and enlargement of peritumoral lymphatic vessels, and contribute to tumor to metastasize to distant organs (100). It has been estimated that chronic infection and associated inflammation contribute to about one in four of all cancer cases worldwide (37). In this section, we will discuss what is known about angiopoietins and Tie2 receptor in tumorigenesis.

5.1. Role of Ang1 in tumor angiogenesis

The role of Ang1 in tumor development is complex and studies have shown both pro- and anti-tumor effects in the presence of this growth factor. Ang1 is constitutively expressed in normal lung tissues; however, the expression was significantly decreased in non-small cell lung carcinoma (NSCLC) (98), indicating Ang1 may have adverse effects in tumor development. Indeed, studies show that over-expression of Ang1 inhibits tumor growth in several tumor models, including breast tumor (35) and skin tumor (34). Overexpressing Ang1 in colorectal cancer cell lines inhibited tumor vascular formation, tumor cell proliferation, tumor growth and metastasis (77, 78). The anti-tumor effects were suggested to be due to the vascular stabilization action of Ang1 (91). Yet Ang1 also regulates inflammation (30, 89, 90). It would be interesting to examine whether Ang1 affects inflammatory cell infiltration in these tumor models and whether the change of tumor microenvironment caused by the inflammatory cells contributes to tumor inhibition.

Besides the anti-tumor action of Ang1, opposite findings were also reported. Ang1 elevation has been reported in several cancers, including ovarian adenocarcinoma (54), breast cancer (7), non-small cell lung cancer (83), and high-grade glioblastoma (3). Higher expression of Ang1 was also detected in platelet lysate of breast cancer patients (8). Overexpression of Ang1 in cervical cancer cells actually promoted tumor growth by increasing tumor angiogenesis and decreasing apoptosis (74). These controversial findings paint a very complex role of Ang1 in tumor angiogenesis, which implies the role of Ang1 in tumor angiogenesis is context dependent. Ang1 is also known to affect inflammation. Whether inflammatory cells inhibit or promote tumorigenesis may depend on the tumor types and its microenvironment. It would be very interesting to reevaluate the infiltration of inflammatory cells in tumor tissues and their roles in tumorigenesis in these tumor models and determine whether these differences contribute to the opposite outcomes. Clearly, investigation of what factors are driving the differences in the outcomes in these diseases will enhance our ability to tailor different therapies for different cancer and different individuals.

5.2. Role of Ang2 in tumor angiogenesis

Compared to Ang1, the tumor-promoting role of Ang2 seems clear. Ang2 levels were increased in a variety of human cancer biopsies, which include hepatocellular carcinoma(45, 56, 57, 80, 85, 104), nuroblastoma (23), Kaposi's sarcoma (6), lung tumor (98), gastric cancer (24, 81), high-grade glioblastoma (3, 43), colon carcinoma (1, 99), metastatic colorectal cancer (61), hemangioma (102), skin carcinoma (34), and renal cell carcinoma (17). Elevated Ang2 expression is correlated with various observations in tumorigenesis such as microvascular density and tumor size in hepatocellular carcinoma (57, 80), the malignancy and poor survival in gastric cancer (24) and NSCLC (84), and lymph node invasion in breast cancer (72). The high ratio of Ang2/Ang1 was also associated with microvascular density, tumor size, tumor progression and tumor portal vein invasion in hepatocellular carcinoma (56) and the high ratio together with high expression of VEGF was correlated with high microvessel density in ovarian cancer (33). Furthermore, over-expression of Ang2 resulted in increased tumor vascular density, tumor cell proliferation and tumor growth in colon cancer cells and hepatocellular carcinoma (2, 64). It also led to highly metastatic tumors in gastric cancer cells (24). These data provide direct evidence supporting a tumor-promoting role of Ang2.

In agreement with the correlation of Ang2 expression and tumor growth, anti cancer agent treatment decreases Ang2 levels in tumor. Endostatin treatment decreased Ang2 levels and inhibited tumor growth in a mammary cancer model (9). Cannabinoids treatment led to tumor apoptosis and that was accompanied by impairment of tumor vascularization and a decrease of Ang2 expression in a non-melanoma skin cancer model (11). It appears everything must have its exceptions. Yu et al reported that overexpression of Ang2 in Lewis lung carcinoma and TA3 mammary carcinoma cells inhibited their ability to form metastatic tumors and prolonged the survival; but Ang1 did not have any effects in these models (101).

5.3. Role of Tie2 in tumor angiogenesis

To begin to understand the role of Tie2 in pathological neovascularization, Tie2 expression was assessed in a large number of human breast cancer tumor

specimens (65). Consistent with results in adult vasculature, Tie2 was expressed in the vascular endothelium in both normal breast tissue and breast tumors. However, the proportion of Tie2 positive tumor microvessels was increased in tumors compared to normal breast tissue. Moreover, Tie2 expression was concentrated in "vascular hot spots" at the leading edge of invasive tumors (65). Subsequently, elevated Tie2 expression has been shown in a number of other human tumors including non small cell lung cancer (83), hepatocellular carcinoma (87), prostate cancer (7), hemangioma(102), and Kaposi's sarcoma (6). Tie2 expression and activation were correlated with increasing malignancy of highly vascularized astrocytoma (103).

The expression of Tie2 in tumor vasculature suggested a role for Tie2 in tumor angiogenesis, contributing to disease progression. To assess the role of Tie2 in tumor angiogenesis, we developed a soluble Tie2 receptor (ExTek) that functions as a Tie2 inhibitor (47). ExTek inhibited Ang1-mediated Tie2 phosphorylation and cell survival in vitro and inhibited tumor angiogenesis and tumor growth in vivo (46, 47). Moreover, the mammary tumor used in the study also express VEGF and tumor growth was significantly inhibited by either recombinant ExTek or a soluble VEGF receptor-2 (VEGFR2) (47, 48). These results suggest that Ang/Tie2 and VEGF/VEGFR pathways are both pivotal pathways that are either independent or interdependent for tumor angiogenesis. Other studies independently employing different versions of soluble Tie2 receptor have shown similar effects in different tumor models (75, 76, 79, 86, 103). Taken together these findings indicate that Tie2 signaling plays an important role in the development of the tumor vasculature and suggest that this pathway may be involved in the pathogenesis of other angiogenic diseases.

Tumor promoting and inhibiting factors such as oncogenes and tumor suppressor genes regulate tumor angiogenesis. Tumor suppressor gene p53 inhibited tumor growth by several known mechanisms, including suppression of cell proliferation and inhibition of tumor angiogenesis. A recent study identified a link between tumor suppressor gene, p53, and Tie2. Tse et al found that p53 downregulated Tie2 and VEGF and resulted in hemorrhage and abnormal tumor vascular architecture (93). These animals exhibited increased necrosis, reduction of tumor volume and prolonged survival. These findings are in agreement with the tumor suppressing role of p53 and indicating that p53 causes tumor regression by suppressing tumor proliferation and tumor vascular regression.

6. ANGIOPOIETIN/TIE2 SIGNALING AND ARTHRITIS

Rheumatoid arthritis (RA) is not only an autoimmune disease, but also a chronic inflammatory disease associated with intense angiogenesis and the formation of a tumor-like invasive pannus that is responsible for much of the cartilage and bone damage. TNF-alpha is a proinflammatory cytokine involved in the pathogenesis of RA (49), and TNF-alpha inhibitors were successfully used in the treatment of RA (26, 51, 58, 63).

Angiopoietin/Tie2 system has been implicated in the progression of arthritis. Studies including ours have shown that Ang1 is upregulated in RA synovium and TNF-alpha upregulates Ang1 expression in synovial fibroblast (19, 31, 71, 73, 94). TGF-beta also increases Ang1 expression but has no effect on Ang2 expression in RA synovial fibroblasts (71). Ang1 appears to be highly expressed in late stage RA synovial fibroblasts (RSF); however, in chronic inflamed synovial tissue, Ang2 was predominant. The difference in the regulation of Ang1 and Ang2 expression in arthritis tissue indicates distinctive roles of angiopoietins in arthritis development. In addition, we have shown that TNF-alpha also induces Tie2 expression in endothelial cells via the NF-? B transcription factor (19). Thus, it reveals a paracrine regulation mechanism of angiogenesis between endothelial cells and synoviocytes through angiopoietin and Tie2. Using a collagen-induced arthritis mice model (13), we observed that inhibition of Tie2 action by a soluble Tie2 inhibitor (ExTek)(46) significantly reduced inflammatory cell infiltration and tissue inflammation in arthritic joints, which resulted in a significant inhibition of arthritis development and bone protection (Lin unpublished data).

The mechanisms that underlie psoriatic arthritis (PsA) are thought to be similar or identical to RA. However, differences of angiogenic factor expression between PsA and RA have been noted. Ang2 expression was observed in early psoriatic arthritis and rheumatoid arthritis. Expression of Ang2 and VEGF was significantly greater in early PsA than RA. VEGF and TGF-beta 1 concentrations were also significantly higher in early PsA compared to RA. Distinct vascular morphology has been observed between PsA and RA, with tortuous pattern in PsA and a straight pattern in RA, which correlated with microscopic vascular scores and VEGF and angiopoietin levels (10, 25, 66). Ang1 expression was observed, but concentrations were markedly lower than Ang2 and VEGF(25). In addition, osteoarthritis (OA) is commonly classified as a non-inflammatory disease to distinguish it from inflammatory arthritis such as RA; however, angiogenesis and inflammation have been shown to play a role in progression of OA as well (4). In OA, subclinical inflammation is common and that often correlates to cartilage changes. Collectively, these studies indicate a close relationship between angiogenic factors, vascular morphology and arthritis disease progression, with important pathogenic and therapeutic implications.

7. ANGIOPOIETIN/TIE2 SIGNALING AND ATHEROSCLEROSIS

Atherosclerosis is a multi-factorial process and its outcome, coronary heart disease, is considered to be responsible for the greatest number of deaths worldwide. The pathophysiology of atherosclerosis has evolved in recent years from long believed hypercholesterolaemia to inflammation and endothelial dysfunction. It is known that local inflammation occurs in the formation of the plaques. Macrophages and monocytes are present in the inflamed endothelium in the early stage of the disease. Inflammation may also directly contribute to endothelial dysfunction. An inflammatory cascade, binding of monocytes to

endothelium at the lesion, developing into macrophages and form cells, and finally initiating the formation of fatty streaks, is a critical step in atherosclerotic development. Inflammatory cytokines induces vascular cell adhesion molecule 1 (VCAM-1) expression in endothelium, which plays a role in leukocyte binding in nascent atheroma. Interestingly, cholesterol feeding also increased VCAM-1 expression in the area that is prone to have lesions (18). Besides VCAM-1, P-and E-selectin have been shown to be involved in atherosclerosis development (20).

A few recent studies indicate Tie2 and angiopoietins in atherosclerosis. In coronary artery disease patients, the levels of endogenous soluble Tie2 in plasma were significantly higher than the healthy controls, indicating a role of the angiopoietin/Tie2 system in atherogenesis (14). Interestingly, over-expression of Ang1 in cardiac allograft arteriosclerosis model using an adenoviral vector protected the formation of arteriosclerosis, which is accompanied with a decrease of the level of plasma Ang2, the number of graft-infiltrating leukocytes, and the incidence and intensity of intimal lesions (60). Studies have shown that Ang1 down regulates those cell adhesion molecules on endothelial cells (30, 41). Thus, the protection seen by Ang1 may be due to its regulation of adhesion molecules. Studies in Tie2/angiopoietins system in atherosclerosis field are limited; however, this anti-inflammatory role of Ang1 could be beneficial for preventing or treating atherosclerosis in the future.

8. CONCLUSIONS

Angiogenesis plays essential roles in human disease development. Therefore, understanding the molecular mechanisms that drive blood vessel growth in adult tissues could provide novel approaches for therapy. Tie2 is a receptor tyrosine kinase expressed principally on vascular endothelium, making it an attractive target for angiogenic therapy. Multiple ligands for Tie2, named angiopoietins, have been identified. One of the unique properties of angiopoietins is the presence of agonist (e.g. Ang1) and antagonist ligands (e.g. Ang2) for Tie2, indicating a fine balance of Tie2 activation is critical for vascular development and remodeling. However, it has also been demonstrated that under some circumstances, Ang2 can stimulate Tie2 suggesting that the action of Ang2 as a Tie2 agonist or antagonist is context-dependent. The complexity of angiogenesis and context-dependent regulation of angiopoietin/Tie2 signaling require further studies from many disciplines in the biomedical field. One of the major differences between physiological and pathological angiogenesis is inflammation that commonly presents in disease conditions. Investigating the interaction of inflammation and Tie2/angiopoietin signaling mediated angiogenesis, as well as their role in inflammation will yield important information of disease mechanisms.

9. ACKNOWLEDGEMENTS

We thank Laura DeBusk and Candice Shaifer at Vanderbilt University Medical Center for critical reading and comments on the manuscript. This work was supported in part by NIH grants CA87756, CA 86283, CA095103, DK20593, CCSG P30-CA68485 and P50CA 86283. HK received training support from NIH (T32-CA093240).

10. REFERENCES

1. Ahmad, S. A., W. Liu, Y. D. Jung, F. Fan, N. Reinmuth, C. D. Bucana, and L. M. Ellis: Differential expression of angiopoietin-1 and angiopoietin-2 in colon carcinoma. A possible mechanism for the initiation of angiogenesis. *Cancer*. 92, 1138-43 (2001)

2. Ahmad, S. A., W. Liu, Y. D. Jung, F. Fan, M. Wilson, N. Reinmuth, R. M. Shaheen, C. D. Bucana, and L. M. Ellis: The effects of angiopoietin-1 and -2 on tumor growth and angiogenesis in human colon cancer. *Cancer Res*. 61, 1255-9 (2001)

3. Audero, E., I. Cascone, I. Zanon, S. C. Previtali, R. Piva, D. Schiffer, and F. Bussolino: Expression of angiopoietin-1 in human glioblastomas regulates tumor-induced angiogenesis: in vivo and in vitro studies. *Arterioscler Thromb Vasc Biol*. 21, 536-41 (2001)

4. Bonnet, C. S., and D. A. Walsh: Osteoarthritis, angiogenesis and inflammation. *Rheumatology (Oxford)* (2004)

5. Brown, C., J. Gaspar, A. Pettit, R. Lee, X. Gu, H. Wang, C. Manning, C. Voland, S. R. Goldring, M. B. Goldring, T. A. Libermann, E. M. Gravallese, and P. Oettgen: ESE-1 is a novel transcriptional mediator of angiopoietin-1 expression in the setting of inflammation. *J Biol Chem*. 279, 12794-803 (2004)

6. Brown, L. F., B. J. Dezube, K. Tognazzi, H. F. Dvorak, and G. D. Yancopoulos: Expression of Tie1, Tie2, and angiopoietins 1, 2, and 4 in Kaposi's sarcoma and cutaneous angiosarcoma. *Am J Pathol*. 156, 2179-83 (2000)

7. Caine, G. J., A. D. Blann, P. S. Stonelake, P. Ryan, and G. Y. Lip: Plasma angiopoietin-1, angiopoietin-2 and Tie-2 in breast and prostate cancer: a comparison with VEGF and Flt-1. *Eur J Clin Invest.* 33, 883-90 (2003)

8. Caine, G. J., G. Y. Lip, and A. D. Blann: Platelet-derived VEGF, Flt-1, angiopoietin-1 and P-selectin in breast and prostate cancer: further evidence for a role of platelets in tumour angiogenesis. *Ann Med*. 36, 273-7 (2004)

9. Calvo, A., Y. Yokoyama, L. E. Smith, I. Ali, S. C. Shih, A. L. Feldman, S. K. Libutti, R. Sundaram, and J. E. Green: Inhibition of the mammary carcinoma angiogenic switch in C3(1)/SV40 transgenic mice by a mutated form of human endostatin. *Int J Cancer*. 101, 224-34 (2002)

10. Canete, J. D., J. R. Rodriguez, G. Salvador, A. Gomez-Centeno, J. Munoz-Gomez, and R. Sanmarti: Diagnostic usefulness of synovial vascular morphology in chronic arthritis. A systematic survey of 100 cases. *Semin Arthritis Rheum*. 32, 378-87 (2003)

11. Casanova, M. L., C. Blazquez, J. Martinez-Palacio, C. Villanueva, M. J. Fernandez-Acenero, J. W. Huffman, J. L. Jorcano, and M. Guzman: Inhibition of skin tumor growth and angiogenesis in vivo by activation of cannabinoid receptors. *J Clin Invest.* 111, 43-50 (2003)

12. Chen, J. X., Y. Chen, L. DeBusk, W. Lin, and P. C. Lin: Dual functional roles of Tie-2/angiopoietin in TNF-

alpha-mediated angiogenesis. *Am J Physiol Heart Circ Physiol*. 287, H187-95 (2004)
13. Chen, Y., E. Rosloniec, J. Price, M. Boothby, and J. Chen: Constitutive expression of BCL-X(L) in the T lineage attenuates collagen-induced arthritis in Bcl-X(L) transgenic mice. *Arthritis Rheum*. 46, 514-21. (2002)
14. Chung, N. A., A. J. Makin, and G. Y. Lip: Measurement of the soluble angiopoietin receptor tie-2 in patients with coronary artery disease: development and application of an immunoassay. *Eur J Clin Invest* 33, 529-35 (2003)
15. Coussens, L. M., W. W. Raymond, G. Bergers, M. Laig-Webster, O. Behrendtsen, Z. Werb, G. H. Caughey, and D. Hanahan: Inflammatory mast cells up-regulate angiogenesis during squamous epithelial carcinogenesis. *Genes Dev*. 13, 1382-97 (1999)
16. Coussens, L. M., and Z. Werb: Inflammation and cancer. *Nature*. 420, 860-7 (2002)
17. Currie, M. J., S. P. Gunningham, K. Turner, C. Han, P. A. Scott, B. A. Robinson, W. Chong, A. L. Harris, and S. B. Fox: Expression of the angiopoietins and their receptor Tie2 in human renal clear cell carcinomas; regulation by the von Hippel-Lindau gene and hypoxia. *J Pathol*. 198, 502-10 (2002)
18. Cybulsky, M. I., and M. A. Gimbrone, Jr.: Endothelial expression of a mononuclear leukocyte adhesion molecule during atherogenesis. *Science*. 251, 788-91 (1991)
19. DeBusk, L. M., Y. Chen, T. Nishishita, J. Chen, J. W. Thomas, and P. C. Lin: Tie2 receptor tyrosine kinase, a major mediator of tumor necrosis factor alpha-induced angiogenesis in rheumatoid arthritis. *Arthritis Rheum*. 48, 2461-71 (2003)
20. Dong, Z. M., S. M. Chapman, A. A. Brown, P. S. Frenette, R. O. Hynes, and D. D. Wagner: The combined role of P- and E-selectins in atherosclerosis. *J Clin Invest*. 102, 145-52 (1998)
21. Dumont, D. J., G. Gradwohl, G. H. Fong, M. C. Puri, M. Gertsenstein, A. Auerbach, and M. L. Breitman: Dominant-negative and targeted null mutations in the endothelial receptor tyrosine kinase, tek, reveal a critical role in vasculogenesis of the embryo. *Genes Dev*. 8, 1897-909 (1994)
22. Dumont, D. J., T. P. Yamaguchi, R. A. Conlon, J. Rossant, and M. L. Breitman: tek, a novel tyrosine kinase gene located on mouse chromosome 4, is expressed in endothelial cells and their presumptive precursors. *Oncogene*. 7, 1471-80 (1992)
23. Eggert, A., N. Ikegaki, J. Kwiatkowski, H. Zhao, G. M. Brodeur, and B. P. Himelstein: High-level expression of angiogenic factors is associated with advanced tumor stage in human neuroblastomas. *Clin Cancer Res*. 6, 1900-8 (2000)
24. Etoh, T., H. Inoue, S. Tanaka, G. F. Barnard, S. Kitano, and M. Mori: Angiopoietin-2 is related to tumor angiogenesis in gastric carcinoma: possible in vivo regulation via induction of proteases. *Cancer Res*. 61, 2145-53 (2001)
25. Fearon, U., K. Griosios, A. Fraser, R. Reece, P. Emery, P. F. Jones, and D. J. Veale: Angiopoietins, growth factors, and vascular morphology in early arthritis. *J Rheumatol*. 30, 260-8 (2003)
26. Feldmann, M., and R. N. Maini: Anti-TNF alpha therapy of rheumatoid arthritis: what have we learned? *Annu Rev Immunol*. 19, 163-96 (2001)
27. Fiedler, U., M. Scharpfenecker, S. Koidl, A. Hegen, V. Grunow, J. M. Schmidt, W. Kriz, G. Thurston, and H. G. Augustin: The Tie-2 ligand angiopoietin-2 is stored in and rapidly released upon stimulation from endothelial cell Weibel-Palade bodies. *Blood*. 103, 4150-6 (2004)
28. Folkman, J.: Angiogenesis-dependent diseases. *Semin Oncol*. 28, 536-42. (2001)
29. Gale, N. W., G. Thurston, S. F. Hackett, R. Renard, Q. Wang, J. McClain, C. Martin, C. Witte, M. H. Witte, D. Jackson, C. Suri, P. A. Campochiaro, S. J. Wiegand, and G. D. Yancopoulos: Angiopoietin-2 is required for postnatal angiogenesis and lymphatic patterning, and only the latter role is rescued by Angiopoietin-1. *Dev Cell*. 3, 411-23 (2002)
30. Gamble, J. R., J. Drew, L. Trezise, A. Underwood, M. Parsons, L. Kasminkas, J. Rudge, G. Yancopoulos, and M. A. Vadas: Angiopoietin-1 is an antipermeability and anti-inflammatory agent in vitro and targets cell junctions. *Circ Res*. 87, 603-7 (2000)
31. Gravallese, E. M., A. R. Pettit, R. Lee, R. Madore, C. Manning, A. Tsay, J. Gaspar, M. B. Goldring, S. R. Goldring, and P. Oettgen: Angiopoietin-1 is expressed in the synovium of patients with rheumatoid arthritis and is induced by tumour necrosis factor alpha. *Ann Rheum Dis*. 62, 100-7 (2003)
32. Haggstrom Rudolfsson, S., A. Johansson, I. Franck Lissbrant, P. Wikstrom, and A. Bergh: Localized expression of angiopoietin 1 and 2 may explain unique characteristics of the rat testicular microvasculature. *Biol Reprod*. 69, 1231-7 (2003)
33. Hata, K., K. Nakayama, R. Fujiwaki, H. Katabuchi, H. Okamura, and K. Miyazaki: Expression of the angopoietin-1, angopoietin-2, Tie2, and vascular endothelial growth factor gene in epithelial ovarian cancer. *Gynecol Oncol*. 93, 215-22 (2004)
34. Hawighorst, T., M. Skobe, M. Streit, Y. K. Hong, P. Velasco, L. F. Brown, L. Riccardi, B. Lange-Asschenfeldt, and M. Detmar: Activation of the tie2 receptor by angiopoietin-1 enhances tumor vessel maturation and impairs squamous cell carcinoma growth. *Am J Pathol*. 160, 1381-92 (2002)
35. Hayes, A. J., W. Q. Huang, J. Yu, P. C. Maisonpierre, A. Liu, F. G. Kern, M. E. Lippman, S. W. McLeskey, and L. Y. Li: Expression and function of angiopoietin-1 in breast cancer. *Br J Cancer*. 83, 1154-60 (2000)
36. Hughes, D. P., M. B. Marron, and N. P. Brindle: The antiinflammatory endothelial tyrosine kinase Tie2 interacts with a novel nuclear factor-kappaB inhibitor ABIN-2. *Circ Res*. 92, 630-6 (2003)
37. Hussain, S. P., L. J. Hofseth, and C. C. Harris: Radical causes of cancer. *Nat Rev Cancer*. 3, 276-85 (2003)
38. Iwama, A., I. Hamaguchi, M. Hashiyama, Y. Murayama, K. Yasunaga, and T. Suda: Molecular cloning and characterization of mouse TIE and TEK receptor tyrosine kinase genes and their expression in hematopoietic stem cells. *Biochem Biophys Res Commun*. 195, 301-9 (1993)
39. Karmpaliotis, D., I. Kosmidou, E. P. Ingenito, K. Hong, A. Malhotra, M. E. Sunday, and K. J. Haley: Angiogenic growth factors in the pathophysiology of a murine model of acute lung injury. *Am J Physiol Lung Cell Mol Physiol*. 283, L585-95 (2002)

40. Kim, I., J. H. Kim, S. O. Moon, H. J. Kwak, N. G. Kim, and G. Y. Koh: Angiopoietin-2 at high concentration can enhance endothelial cell survival through the phosphatidylinositol 3'-kinase/Akt signal transduction pathway. *Oncogene*. 19, 4549-52 (2000)
41. Kim, I., S. O. Moon, S. K. Park, S. W. Chae, and G. Y. Koh: Angiopoietin-1 reduces VEGF-stimulated leukocyte adhesion to endothelial cells by reducing ICAM-1, VCAM-1, and E-selectin expression. *Circulation Research (Online)* 89, 477-9 (2001)
42. Kim, I., J. L. Oh, Y. S. Ryu, J. N. So, W. C. Sessa, K. Walsh, and G. Y. Koh: Angiopoietin-1 negatively regulates expression and activity of tissue factor in endothelial cells. *The Faseb Journal : Official Publication of the Federation of American Societies For Experimental Biology.* 16, 126-8 (2002)
43. Koga, K., T. Todaka, M. Morioka, J. Hamada, Y. Kai, S. Yano, A. Okamura, N. Takakura, T. Suda, and Y. Ushio: Expression of angiopoietin-2 in human glioma cells and its role for angiogenesis. *Cancer Res*. 61, 6248-54 (2001)
44. Krikun, G., F. Schatz, T. Finlay, S. Kadner, A. Mesia, R. Gerrets, and C. J. Lockwood: Expression of angiopoietin-2 by human endometrial endothelial cells: regulation by hypoxia and inflammation. *Biochem Biophys Res Commun*. 275, 159-63 (2000)
45. Kuroda, H., A. Ohtsuru, M. Futakuchi, Y. Kawashita, Y. Nagayama, E. Fukuda, H. Namba, T. Shirai, T. Kanematsu, and S. Yamashita: Distinctive gene expression of receptor-type tyrosine kinase families during rat hepatocarcinogenesis. *Int J Mol Med*. 9, 473-80 (2002)
46. Lin, P., J. A. Buxton, A. Acheson, C. Radziejewski, P. C. Maisonpierre, G. D. Yancopoulos, K. M. Channon, L. P. Hale, M. W. Dewhirst, S. E. George, and K. G. Peters: Antiangiogenic gene therapy targeting the endothelium-specific receptor tyrosine kinase Tie2. *Proc Natl Acad Sci U S A*. 95, 8829-34. (1998)
47. Lin, P., P. Polverini, M. Dewhirst, S. Shan, P. S. Rao, and K. Peters: Inhibition of tumor angiogenesis using a soluble receptor establishes a role for Tie2 in pathologic vascular growth. *J Clin Invest.* 100, 2072-8. (1997)
48. Lin, P., S. Sankar, S. Shan, M. W. Dewhirst, P. J. Polverini, T. Q. Quinn, and K. G. Peters: Inhibition of tumor growth by targeting tumor endothelium using a soluble vascular endothelial growth factor receptor. *Cell Growth Differ*. 9, 49-58 (1998)
49. Lupia, E., G. Montrucchio, E. Battaglia, V. Modena, and G. Camussi: Role of tumor necrosis factor-alpha and platelet-activating factor in neoangiogenesis induced by synovial fluids of patients with rheumatoid arthritis. *Eur J Immunol*. 26, 1690-4 (1996)
50. Lyons, M. S., B. Bell, D. Stainier, and K. G. Peters: Isolation of the zebrafish homologues for the tie-1 and tie-2 endothelium-specific receptor tyrosine kinases. *Dev Dyn*. 212, 133-40 (1998)
51. Maini, R. N., P. C. Taylor, E. Paleolog, P. Charles, S. Ballara, F. M. Brennan, and M. Feldmann: Anti-tumour necrosis factor specific antibody (infliximab) treatment provides insights into the pathophysiology of rheumatoid arthritis. *Ann Rheum Dis*. 58 Suppl 1, I56-60 (1999)
52. Maisonpierre, P. C., M. Goldfarb, G. D. Yancopoulos, and G. Gao: Distinct rat genes with related profiles of expression define a TIE receptor tyrosine kinase family. *Oncogene*. 8, 1631-7 (1993)
53. Maisonpierre, P. C., C. Suri, P. F. Jones, S. Bartunkova, S. J. Wiegand, C. Radziejewski, D. Compton, J. McClain, T. H. Aldrich, N. Papadopoulos, T. J. Daly, S. Davis, T. N. Sato, and G. D. Yancopoulos: Angiopoietin-2, a natural antagonist for Tie2 that disrupts in vivo angiogenesis. *Science.* 277, 55-60 (1997)
54. Martoglio, A. M., B. D. Tom, M. Starkey, A. N. Corps, D. S. Charnock-Jones, and S. K. Smith: Changes in tumorigenesis- and angiogenesis-related gene transcript abundance profiles in ovarian cancer detected by tailored high density cDNA arrays. *Mol Med*. 6, 750-65 (2000)
55. Melani, C., A. Stoppacciaro, C. Foroni, F. Felicetti, A. Care, and M. P. Colombo: Angiopoietin decoy secreted at tumor site impairs tumor growth and metastases by inducing local inflammation and altering neoangiogenesis. *Cancer Immunol Immunother*. 53, 600-8 (2004)
56. Mitsuhashi, N., H. Shimizu, M. Ohtsuka, Y. Wakabayashi, H. Ito, F. Kimura, H. Yoshidome, A. Kato, Y. Nukui, and M. Miyazaki: Angiopoietins and Tie-2 expression in angiogenesis and proliferation of human hepatocellular carcinoma. *Hepatology (Baltimore, Md.)* 37, 1105-13 (2003)
57. Moon, W. S., K. H. Rhyu, M. J. Kang, D. G. Lee, H. C. Yu, J. H. Yeum, G. Y. Koh, and A. S. Tarnawski: Overexpression of VEGF and angiopoietin 2: a key to high vascularity of hepatocellular carcinoma? *Mod Pathol*. 16, 552-7 (2003)
58. Moreland, L. W., S. W. Baumgartner, M. H. Schiff, E. A. Tindall, R. M. Fleischmann, A. L. Weaver, R. E. Ettlinger, S. Cohen, W. J. Koopman, K. Mohler, M. B. Widmer, and C. M. Blosch: Treatment of rheumatoid arthritis with a recombinant human tumor necrosis factor receptor (p75)-Fc fusion protein. *N Engl J Med*. 337, 141-7 (1997)
59. Morrissey, J. H.: Tissue factor: a key molecule in hemostatic and nonhemostatic systems. *Int J Hematol*. 79, 103-8 (2004)
60. Nykanen, A. I., R. Krebs, A. Saaristo, P. Turunen, K. Alitalo, S. Yla-Herttuala, P. K. Koskinen, and K. B. Lemstrom: Angiopoietin-1 protects against the development of cardiac allograft arteriosclerosis. *Circulation*. 107, 1308-14 (2003)
61. Ogawa, M., H. Yamamoto, H. Nagano, Y. Miyake, Y. Sugita, T. Hata, B. N. Kim, C. Y. Ngan, B. Damdinsuren, M. Ikenaga, M. Ikeda, M. Ohue, S. Nakamori, M. Sekimoto, M. Sakon, N. Matsuura, and M. Monden: Hepatic expression of ANG2 RNA in metastatic colorectal cancer. *Hepatology*. 39, 528-39 (2004)
62. Ono, M., H. Torisu, J. Fukushi, A. Nishie, and M. Kuwano: Biological implications of macrophage infiltration in human tumor angiogenesis. *Cancer Chemother Pharmacol*. 43 Suppl, S69-71 (1999)
63. Paleolog, E. M., S. Young, A. C. Stark, R. V. McCloskey, M. Feldmann, and R. N. Maini: Modulation of angiogenic vascular endothelial growth factor by tumor necrosis factor alpha and interleukin-1 in rheumatoid arthritis. *Arthritis Rheum*. 41, 1258-65 (1998)
64. Peng, L., J. Sun, W. D. Wang, Z. X. Jian, and J. R. Ou: Biological effect of ectopic expression of angiopoietin-1

and -2 in hepatocellular carcinoma cell line. *Hepatobiliary Pancreat Dis Int*. 2, 94-7 (2003)
65. Peters, K. G., A. Coogan, D. Berry, J. Marks, J. D. Iglehart, C. D. Kontos, P. Rao, S. Sankar, and E. Trogan: Expression of Tie2/Tek in breast tumour vasculature provides a new marker for evaluation of tumour angiogenesis. *Br J Cancer*. 77, 51-6 (1998)
66. Reece, R. J., J. D. Canete, W. J. Parsons, P. Emery, and D. J. Veale: Distinct vascular patterns of early synovitis in psoriatic, reactive, and rheumatoid arthritis. *Arthritis Rheum*. 42, 1481-4 (1999)
67. Runting, A. S., S. A. Stacker, and A. F. Wilks: tie2, a putative protein tyrosine kinase from a new class of cell surface receptor. *Growth Factors*. 9, 99-105 (1993)
68. Sato, T. N., Y. Qin, C. A. Kozak, and K. L. Audus: Tie-1 and tie-2 define another class of putative receptor tyrosine kinase genes expressed in early embryonic vascular system. *Proc Natl Acad Sci U S A* 90, 9355-8 (1993)
69. Sato, T. N., Y. Tozawa, U. Deutsch, K. Wolburg-Buchholz, Y. Fujiwara, M. Gendron-Maguire, T. Gridley, H. Wolburg, W. Risau, and Y. Qin: Distinct roles of the receptor tyrosine kinases Tie-1 and Tie-2 in blood vessel formation. *Nature.* 376, 70-4 (1995)
70. Schnurch, H., and W. Risau: Expression of tie-2, a member of a novel family of receptor tyrosine kinases, in the endothelial cell lineage. *Development*. 119, 957-68 (1993)
71. Scott, B. B., P. F. Zaratin, A. Colombo, M. J. Hansbury, J. D. Winkler, and J. R. Jackson: Constitutive expression of angiopoietin-1 and -2 and modulation of their expression by inflammatory cytokines in rheumatoid arthritis synovial fibroblasts. *J Rheumatol*. 29, 230-9 (2002)
72. Sfiligoi, C., A. de Luca, I. Cascone, V. Sorbello, L. Fuso, R. Ponzone, N. Biglia, E. Audero, R. Arisio, F. Bussolino, P. Sismondi, and M. De Bortoli: Angiopoietin-2 expression in breast cancer correlates with lymph node invasion and short survival. *Int J Cancer*. 103, 466-74 (2003)
73. Shahrara, S., M. V. Volin, M. A. Connors, G. K. Haines, and A. E. Koch: Differential expression of the angiogenic Tie receptor family in arthritic and normal synovial tissue. *Arthritis Res*. 4, 201-8 (2002)
74. Shim, W. S., M. Teh, A. Bapna, I. Kim, G. Y. Koh, P. O. Mack, and R. Ge: Angiopoietin 1 promotes tumor angiogenesis and tumor vessel plasticity of human cervical cancer in mice. *Exp Cell Res*. 279, 299-309 (2002)
75. Shirakawa, K., M. Shibuya, Y. Heike, S. Takashima, I. Watanabe, F. Konishi, F. Kasumi, C. K. Goldman, K. A. Thomas, A. Bett, M. Terada, and H. Wakasugi: Tumor-infiltrating endothelial cells and endothelial precursor cells in inflammatory breast cancer. *Int J Cancer*. 99, 344-51 (2002)
76. Siemeister, G., M. Schirner, K. Weindel, P. Reusch, A. Menrad, D. Marme, and G. Martiny-Baron: Two independent mechanisms essential for tumor angiogenesis: inhibition of human melanoma xenograft growth by interfering with either the vascular endothelial growth factor receptor pathway or the Tie-2 pathway. *Cancer Res*. 59, 3185-91 (1999)
77. Stoeltzing, O., S. A. Ahmad, W. Liu, M. F. McCarty, A. A. Parikh, F. Fan, N. Reinmuth, C. D. Bucana, and L. M. Ellis: Angiopoietin-1 inhibits tumour growth and ascites formation in a murine model of peritoneal carcinomatosis. *Br J Cancer*. 87, 1182-7 (2002)
78. Stoeltzing, O., S. A. Ahmad, W. Liu, M. F. McCarty, J. S. Wey, A. A. Parikh, F. Fan, N. Reinmuth, M. Kawaguchi, C. D. Bucana, and L. M. Ellis: Angiopoietin-1 inhibits vascular permeability, angiogenesis, and growth of hepatic colon cancer tumors. *Cancer Res*. 63, 3370-7 (2003)
79. Stratmann, A., T. Acker, A. M. Burger, K. Amann, W. Risau, and K. H. Plate: Differential inhibition of tumor angiogenesis by tie2 and vascular endothelial growth factor receptor-2 dominant-negative receptor mutants. *Int J Cancer*. 91, 273-82. (2001)
80. Sugimachi, K., S. Tanaka, K. Taguchi, S. Aishima, M. Shimada, and M. Tsuneyoshi: Angiopoietin switching regulates angiogenesis and progression of human hepatocellular carcinoma. *J Clin Pathol*. 56, 854-60 (2003)
81. Sun, X. D., X. E. Liu, J. M. Wu, X. J. Cai, Y. P. Mou, and J. D. Li: Expression and significance of angiopoietin-2 in gastric cancer. *World J Gastroenterol*. 10, 1382-5 (2004)
82. Suri, C., P. F. Jones, S. Patan, S. Bartunkova, P. C. Maisonpierre, S. Davis, T. N. Sato, and G. D. Yancopoulos: Requisite role of angiopoietin-1, a ligand for the TIE2 receptor, during embryonic angiogenesis. *Cell*. 87, 1171-80 (1996)
83. Takahama, M., M. Tsutsumi, T. Tsujiuchi, K. Nezu, K. Kushibe, S. Taniguchi, Y. Kotake, and Y. Konishi: Enhanced expression of Tie2, its ligand angiopoietin-1, vascular endothelial growth factor, and CD31 in human non-small cell lung carcinomas. *Clin Cancer Res*. 5, 2506-10 (1999)
84. Tanaka, F., S. Ishikawa, K. Yanagihara, R. Miyahara, Y. Kawano, M. Li, Y. Otake, and H. Wada: Expression of angiopoietins and its clinical significance in non-small cell lung cancer. *Cancer Res*. 62, 7124-9 (2002)
85. Tanaka, S., M. Mori, Y. Sakamoto, M. Makuuchi, K. Sugimachi, and J. R. Wands: Biologic significance of angiopoietin-2 expression in human hepatocellular carcinoma. *J Clin Invest.* 103, 341-5 (1999)
86. Tanaka, S., K. Sugimachi, Y. Yamashita Yi, T. Ohga, K. Shirabe, M. Shimada, and J. R. Wands: Tie2 vascular endothelial receptor expression and function in hepatocellular carcinoma. *Hepatology*. 35, 861-7. (2002)
87. Tanaka, S., K. Sugimachi, Y. Yamashita Yi, T. Ohga, K. Shirabe, M. Shimada, and J. R. Wands: Tie2 vascular endothelial receptor expression and function in hepatocellular carcinoma. *Hepatology*. 35, 861-7 (2002)
88. Teichert-Kuliszewska, K., P. C. Maisonpierre, N. Jones, A. I. Campbell, Z. Master, M. P. Bendeck, K. Alitalo, D. J. Dumont, G. D. Yancopoulos, and D. J. Stewart: Biological action of angiopoietin-2 in a fibrin matrix model of angiogenesis is associated with activation of Tie2. *Cardiovasc Res*. 49, 659-70 (2001)
89. Thurston, G., J. S. Rudge, E. Ioffe, H. Zhou, L. Ross, S. D. Croll, N. Glazer, J. Holash, D. M. McDonald, and G. D. Yancopoulos: Angiopoietin-1 protects the adult vasculature against plasma leakage. *Nat Med*. 6, 460-3 (2000)
90. Thurston, G., C. Suri, K. Smith, J. McClain, T. N. Sato, G. D. Yancopoulos, and D. M. McDonald: Leakage-resistant blood vessels in mice transgenically overexpressing angiopoietin-1. *Science*. 286, 2511-4 (1999)
91. Tian, S., A. J. Hayes, L. J. Metheny-Barlow, and L. Y. Li: Stabilization of breast cancer xenograft tumour

neovasculature by angiopoietin-1. *Br J Cancer*. 86, 645-51 (2002)
92. Torisu, H., M. Ono, H. Kiryu, M. Furue, Y. Ohmoto, J. Nakayama, Y. Nishioka, S. Sone, and M. Kuwano: Macrophage infiltration correlates with tumor stage and angiogenesis in human malignant melanoma: possible involvement of TNFalpha and IL-1alpha. *Int J Cancer*. 85, 182-8 (2000)
93. Tse, V., Y. Yung, J. G. Santarelli, D. Juan, M. Hsiao, M. Haas, G. t. Harsh, and G. Silverberg: Effects of tumor suppressor gene (p53) on brain tumor angiogenesis and expression of angiogenic modulators. *Anticancer Res*. 24, 1-10 (2004)
94. Uchida, T., M. Nakashima, Y. Hirota, Y. Miyazaki, T. Tsukazaki, and H. Shindo: Immunohistochemical localisation of protein tyrosine kinase receptors Tie-1 and Tie-2 in synovial tissue of rheumatoid arthritis: correlation with angiogenesis and synovial proliferation. *Ann Rheum Dis*. 59, 607-14 (2000)
95. Valenzuela, D. M., J. A. Griffiths, J. Rojas, T. H. Aldrich, P. F. Jones, H. Zhou, J. McClain, N. G. Copeland, D. J. Gilbert, N. A. Jenkins, T. Huang, N. Papadopoulos, P. C. Maisonpierre, S. Davis, and G. D. Yancopoulos: Angiopoietins 3 and 4: diverging gene counterparts in mice and humans. *Proc Natl Acad Sci U S A*. 96, 1904-9 (1999)
96. Willam, C., P. Koehne, J. S. Jurgensen, M. Grafe, K. D. Wagner, S. Bachmann, U. Frei, and K. U. Eckardt: Tie2 receptor expression is stimulated by hypoxia and proinflammatory cytokines in human endothelial cells. *Circ Res*. 87, 370-7 (2000)
97. Witzenbichler, B., P. C. Maisonpierre, P. Jones, G. D. Yancopoulos, and J. M. Isner: Chemotactic properties of angiopoietin-1 and -2, ligands for the endothelial-specific receptor tyrosine kinase Tie2. *J Biol Chem*. 273, 18514-21 (1998)
98. Wong, M. P., S. Y. Chan, K. H. Fu, S. Y. Leung, N. Cheung, S. T. Yuen, and L. P. Chung: The angiopoietins, tie2 and vascular endothelial growth factor are differentially expressed in the transformation of normal lung to non-small cell lung carcinomas. *Lung Cancer*. 29, 11-22 (2000)
99. Yoshida, Y., Y. Oshika, Y. Fukushima, T. Tokunaga, H. Hatanaka, H. Kijima, H. Yamazaki, Y. Ueyama, N. Tamaoki, S. Miura, and M. Nakamura: Expression of angiostatic factors in colorectal cancer. *Int J Oncol.* 15, 1221-5 (1999)
100. Yu, J. L., and J. W. Rak: Host microenvironment in breast cancer development: inflammatory and immune cells in tumour angiogenesis and arteriogenesis. *Breast Cancer Res*. 5, 83-8 (2003)
101. Yu, Q., and I. Stamenkovic: Angiopoietin-2 is implicated in the regulation of tumor angiogenesis. *Am J Pathol*. 158, 563-70 (2001)
102. Yu, Y., J. Varughese, L. F. Brown, J. B. Mulliken, and J. Bischoff: Increased Tie2 expression, enhanced response to angiopoietin-1, and dysregulated angiopoietin-2 expression in hemangioma-derived endothelial cells. *Am J Pathol*. 159, 2271-80 (2001)
103. Zadeh, G., B. Qian, A. Okhowat, N. Sabha, C. D. Kontos, and A. Guha: Targeting the Tie2/Tek receptor in astrocytomas. *Am J Pathol*. 164, 467-76 (2004)
104. Zhao, Z. C., S. S. Zheng, Y. L. Wan, C. K. Jia, and H. Y. Xie: The molecular mechanism underlying angiogenesis in hepatocellular carcinoma: the imbalance activation of signaling pathways. *Hepatobiliary Pancreat Dis Int*. 2, 529-36 (2003)
105. Ziegler, S. F., T. A. Bird, J. A. Schneringer, K. A. Schooley, and P. R. Baum: Molecular cloning and characterization of a novel receptor protein tyrosine kinase from human placenta. *Oncogene.* 8, 663-70 (1993)

Key Words: Angiogenesis, Inflammation, Angiopoietin, Tie2, Cancer, Arthritis And Atherosclerosis, Gene, Review

Send correspondence to: P. Charles Lin, Ph.D., 315 PRB, 2220 Pierce Avenue, Vanderbilt University Medical Center, Nashville, TN 37232, Tel: 615-936-1749, Fax: 615-936-1872, E-mail:charles.lin@vanderbilt.edu

http://www.bioscience.org/current/vol10.htm