Mechanisms of Tumor Angiogenesis and Resulting Therapy Resistance

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Abstract

In 2021, over 10 million people died of cancer worldwide. Although recent technology and innovations have saved the lives of many, a large portion of cancer cells continue to avoid these treatments; a key part of this is due to the formation of new blood capillaries, known as angiogenesis. Typically, angiogenesis occurs in tissue repair, embryonic development, the menstrual cycle, muscle growth, and organ-lining development.[1] However, tumor angiogenesis is uncontrolled and rapid, giving tumor cells the nutrient and oxygen supply they need to sustain themselves and spread through the bloodstream. Several treatments have been attempted to disrupt this process, including angiogenesis inhibitors, but more widespread therapies, such as chemotherapy, are often affected by both angiogenesis and its inhibition. To effectively administer treatments, it is essential that the mechanisms, including pro-angiogenic growth factors, signalling pathways, and oxygen levels, of this self-sustaining aspect of cancer cells is evaluated and taken into consideration. In this study, I aim to analyze the extent to which tumor angiogenesis allows these malignant cells to resist cancer treatments.

Introduction

Killing malignant tumor cells is a necessary goal in cancer treatment. A tumor is a mass of abnormal cells that forms in the body,[2] but it is only considered malignant, or cancerous, when it spreads uncontrollably through metastasis. In 1971, Folkman[3] observed that vascularization was more present in rapidly-growing tumors than in their dormant counterparts. A reduced efficacy of drug delivery and continued tumor growth are two of many complications involved in the treatment of tumors, both of which are a direct result of tumor angiogenesis.

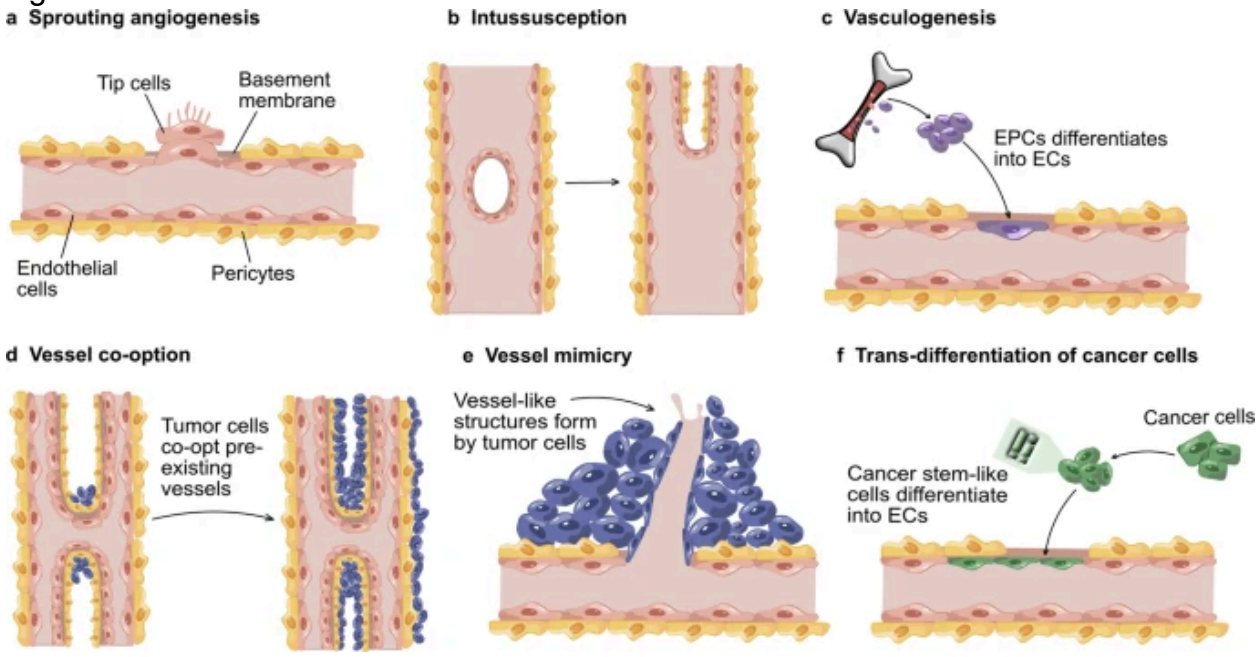
In its early stages and before tumor angiogenesis, a tumor is restricted between 2-3 mm^3 and relies on passive diffusion to receive oxygen and nutrients.[1] However, due to its rapid proliferation and aggressive vitality, the tumor requires much more nutrients and oxygen than a normal body cell. As it grows, the tumor utilizes angiogenesis in order to sustain itself sufficiently. Without these required blood vessels, tumors would experience a state of low oxygen levels in the microenvironment—known as hypoxia—which can lead to necrosis (passive cell death) or apoptosis (programmed cell death). To counter this, tumors trigger angiogenesis by secreting signalling molecules to surrounding host tissue. These molecules can activate a variety of genes—including vascular endothelial growth factor-A (VEGF-A), hypoxia-inducible factor 1-alpha (HIF-1α), and fibroblast growth factor 2 (FGF-2), among others—that encourage the growth of new blood vessels.

In this review article, I aim to explore how angiogenesis contributes to treatment resistance through an overview of its mechanisms in tumor growth, its role in reducing the efficacy of treatment, and anti-angiogenic therapies.

Mechanisms of Angiogenesis

Angiogenesis is characterized by the process of neovascularization, which etymologically refers to the creation of new blood vessels. Neovascularization occurs when a cell is stimulated by an up-regulation of pro-angiogenic factors (and a down-regulation of anti-angiogenic factors),[4] referred to as the "angiogenic switch". This disrupts vascular homeostasis, which is maintained by several pro- and anti-angiogenic factors. When this occurs, cells' basement membranes, layers of extracellular matrix that line tissues,[5] are injured by local proteases (destructive enzymes).[6] This causes immediate destruction and hypoxia. The endothelial cells (ECs) that were stimulated then migrate to the interstitial space between blood vessels. They continue to proliferate and set, allowing for formation of the lumen of blood vessels. This initiates the recruitment of pericytes (specialized cells located on the walls of vessels) to generate a new basement membrane.[7] Eventually, with this combination of angiogenic factors, anastomoses (connections between blood vessels) are created and blood flow occurs. This process is characterized as sprouting angiogenesis, one of six mechanisms of tumor blood vessel formation—sprouting angiogenesis, intussusceptive angiogenesis, vasculogenesis, vessel co-option, vessel mimicry, and transdifferentiation of cancer cells.[Fig 1]

Fig. 1



Mechanisms of tumor blood vessel formation. **a** Sprouting angiogenesis: most prevalent mode in which endothelial tip cells proliferate and migrate to fuse with an existing vessel or sprout. **b** Intussusception: formation of a double lumen which splits existing vessels into two. **c** Vasculogenesis: bone marrow-originating endothelial progenitor cells (EPCs) form the lumen of new blood vessels. **d** Vessel co-option: tumor cells hijack existing blood vessels. **e** Vessel mimicry: tumor cells mimic the structure of blood vessels in the same area as existing ones to direct the transport of oxygen and nutrients into tumor tissue. **f** Trans-differentiation of cancer cells: cancer stem-like cells differentiate into endothelial cells to form new vessels.[8]

There are several biomolecules that constitute pro-angiogenic factors. A main category of these are growth factors and their receptors. These include vascular endothelial growth factor (VEGF), platelet-derived growth factor (PDGF), epidermal growth factor (EGF), and fibroblast growth factor (FGF). These factors are expressed, often simultaneously, throughout the process of tumor angiogenesis.

Vascular endothelial growth factor (VEGF) is the most studied and potent regulator in tumor angiogenesis. It has a mitotic and anti-apoptotic effect on endothelial cells, and is associated with tumor progression, metastasis, and tumor recurrence.[9] It is also known to mediate vascular permeability and cell formation.[8] VEGF is a family of heparin-binded glycoproteins, which indicates that they are composed of two identical proteins containing a carbohydrate chain, or glycan. In humans, the VEGF family is comprised of several members with different functions: VEGF-A, VEGF-B, VEGF-C, VEGF-D, and placenta growth factor (PlGF). They each bind to cognate receptors (VEGFR) of tyrosine cell kinase, which are enzymes that catalyze phosphorylation from ATP to the amino acid tyrosine.[10] When binding to VEGFRs on the surface of endothelial cells, VEGF stimulates endothelial cells to grow and form blood vessels.[11] VEGFRs are composed of three subtypes: VEGFR-1, VEGFR-2, and VEGFR-3. VEGFR-2 is a transmembrane glycoprotein activated by VEGF-A, which is a crucial factor that maintains endothelial function and promotes cell mitosis and vascular permeability. Vascular permeability is a mechanism that allows exchange between blood vessels and surrounding tissues.[12] VEGF-A induces autophosphorylation (when a protein kinase adds a phosphate group to itself) and signal transduction (converting an extracellular stimulus into an intracellular response) of the VEGFR-2 receptor. VEGFR-2 is a more prominent orchestrator of angiogenesis compared to VEGFR-1, which is a co-receptor for all VEGF-A, VEGF-B, and PlGF, and it is expressed on vascular endothelial cells instead of lymphatic ones, unlike VEGFR-3. VEGFR-2 activates several pro-angiogenic signaling pathways, which could include PI3K/AKT/mTOR, p38 MAPK, and Ras/Raf/MEK/ERK that are related to the growth and survival of ECs and angiogenesis.[13] The VEGF-A/VEGFR-2 is the most crucial signalling pathway and is a popular target in angiogenesis inhibitor research. Over-expressed VEGFR-2 has been detected in melanoma,[14] ovarian cancer,[15] thyroid cancer,[16] and myeloid leukemia[17] specifically. Additionally, the role of PlGF of the VEGF family is highly debated due to its both pro- and anti-angiogenic effects, but is reported to initiate cross-talk between VEGFR-1 and VEGFR-2, to contribute towards stromal cell (fibroblasts, macrophages, smooth muscle cells, endothelial cells) activation and proliferation.[8]

Another signalling pathway is platelet-derived growth factor (PDGF), which is a protein secreted by blood platelets and some stromal cells. It is known to promote vascular maturation through the recruitment of parietal cells and is involved in cell growth and differentiation. It is the main mitogen of mesenchymal cells, which include fibroblasts, smooth muscle cells, and glial cells. It contains four soluble inactive polypeptide chains, which are PDGF-A, PDGF-B, PDGF-C, and PDGF-D. These chains are translated into combinations of proteins called homodimers (when proteins are similar) and heterodimers (when proteins are dissimilar), which include PDGF-AA, PDGF-AB, PDGF-BB, PDGF-CC, and PDGF-DD, among others. These homodimers and heterodimers each perform biological functions. PDGF-AA is a cancer promoter activated by PDGF-A that drives angiogenesis, especially when mediated by

PDGFR-α.[18] PDGF signals with two cell-surface tyrosine kinase receptors, PDGFR-α and PDGFR-β, which commonly regulate angiogenesis through promoting vessel maturation, recruitment of pericytes, and upregulation of VEGF. The PDGF-B/PDGFR-β pathway is the most potent in angiogenic activity as it commands pericyte recruitment to begin revascularization for wound healing. PDGFs/PDGFRs are commonly over-activated in several malignant tissues, such as neuroendocrine tumors,[19] ovarian cancer,[20] and hepatocellular carcinoma.[21]

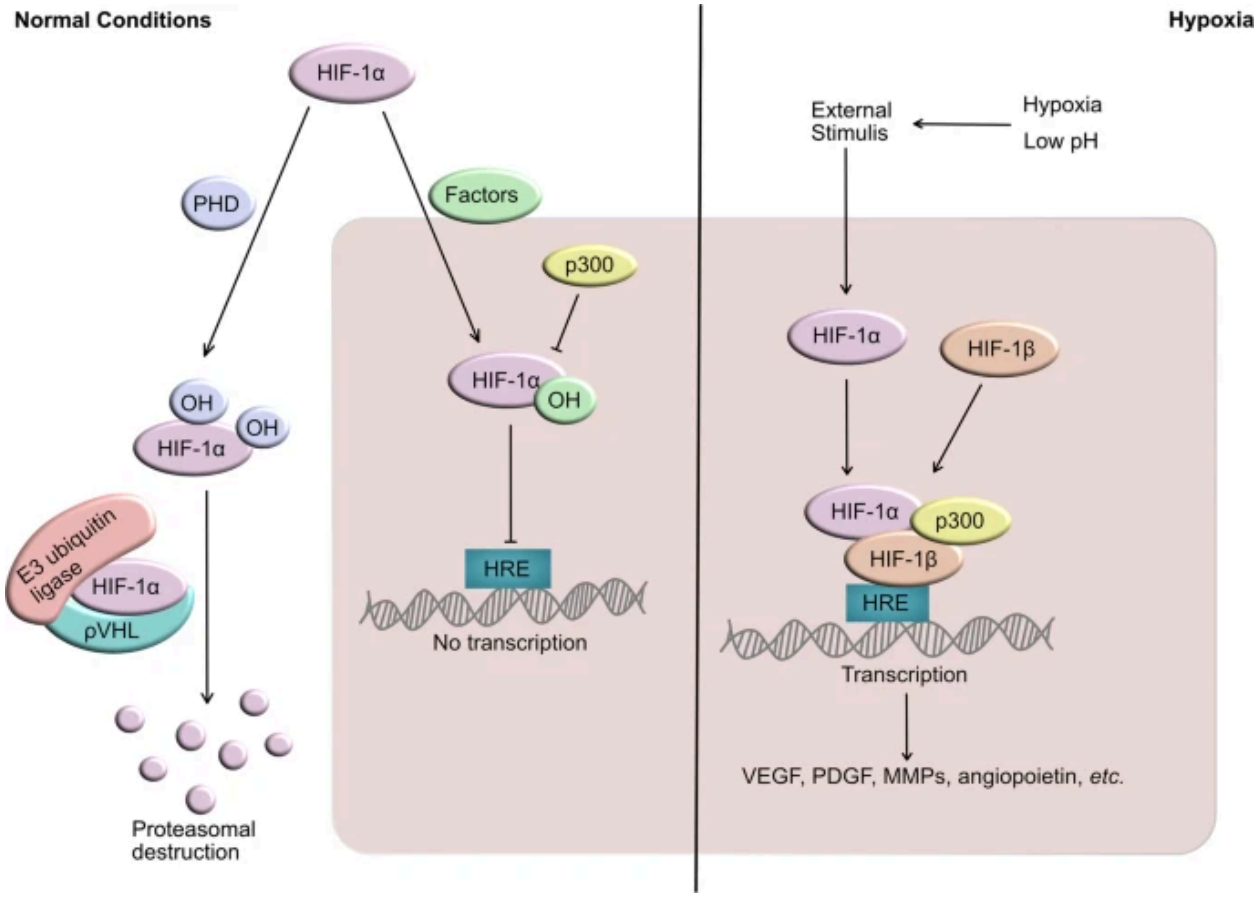
Epidermal growth factor (EGF) is a signalling pathway of a single-chain molecule polypeptide made up of 53 amino acid residues. EGF receptors consist of four proteins: EGFR, HER2, HER3, and HER4 (HER stands for human epidermal receptor). These receptors contain an extramembrane binding domain, a single-chain transmembrane domain that contains a single hydrophobic anchor sequence, and an intramembrane tyrosine kinases bonding domain that generates and moderates intracellular signals. EGFR specifically is used by EGF to widely promote tumor angiogenesis. It upregulates the synthesis, expression, and secretion of various angiogenic factors, such as VEGF, through the Angiopoietin-2 (Ang-2) ligand. HIF-1α, a transcription factor that responds to hypoxia,[22] may induce the expression of EGF and EGFR as EGFR up-regulates the expression of HIF-1α to enhance the oxygen tolerance of cells in a hypoxic environment. This encourages increased aggression in angiogenesis and tumor progression. Many various tumors contain an up-regulation of EGFR, such as in basal cell carcinoma, ovarian cancer, non-small cell lung cancer, glioblastoma multiforme, bladder cancer and pancreatic cancer.[23]

The signalling pathway of fibroblast growth factor (FGF), a potent mitogen and driver of endothelial cells due to its critical role in wound healing, is composed of the FGF family. It is the earliest discovered growth factor related to angiogenesis. It consists of 23 glycoproteins in the extracellular matrix, each with different structures, yet only 18 bind to the four fibroblast growth factor receptors (FGFRs). FGF is secreted by vascular endothelial cells, stem cells, and damaged cardiomyocytes. When signalled, heparinases, proteases, or specific FGF binding proteins release FGFs from the extracellular matrix (the space not including cells within all tissues and organs).[24] Upon liberation, FGFs bind to heparan sulphate proteoglycans (HPSGs) on the cell surface).[4] FGFs regulate angiogenesis through synergistic (cooperating) FGFRs, heparin sulfate polysaccharide, and αvβ integrins (transmembrane receptors that facilitate cell adhesion). The most influential pro-angiogenic factor in the FGF family is FGF-2, also referred to as basic FGF (bFGF). It uses paracrine systems to regulate the specialization of cell function of cardiac non-myocytes. It also stimulates angiogenic-related processes like migration and invasion of endothelial cells. The over-expression of bFGF is seen in basal cell carcinoma, lung cancer, bladder cancer, and leukemia, and it is also related to cancer metastasis and poor prognosis in patients. Therapies that block FGF are described to impair tumor progression when paired with VEGF inhibition.[25] FGFRs are a transmembrane receptor family with five members of FGFR1–5. They undergo autophosphorylation to activate several downstream pathways-activated by bFGF- similar to those activated by VEGFR-2. This family mediates several tumor processes even outside of angiogenesis, such as multiplication, survival, and drug resistance.

A common mechanism of angiogenesis, aside from signalling pathways, is hypoxia. Hypoxia is a term that refers to a decreased availability of oxygen, and is a typical feature in the tumor microenvironment. In normoxic conditions, the transcriptional regulators of

hypoxia-inducible factors (HIFs) maintain oxygen homeostasis and activate the genes that drive angiogenesis to increase oxygen delivery. However, in hypoxic conditions, hundreds to thousands of the genes that drive angiogenesis are either increased or decreased, creating chaotic and rapid vasculature. HIFs are heterodimeric proteins consisting of the oxygen-sensitive α-subunit (HIF-α) and oxygen-independent β-subunit (HIF-β). HIFs typically maintain oxygen homeostasis through a process in which an oxygen atom is inserted into a proline (an amino acid) residue of one of HIF-α's three isoforms: HIF-1α, HIF-2α, or HIF-3α. Another one of three HIF prolyl hydroxylases (PHD1, PHD2, PHD3) and a von Hippel-Lindau (pVHL) protein binds selectively to the HIF-α subunit to stabilize it. HIF-1α is the most responsible for activating pro-angiogenic responses under hypoxia due to its role as an oxygen regulator, increasing its delivery and reducing its consumption when necessary.[8] Under normal conditions, the small protein Ub binds to HIF-1α in a process called ubiquitination. Subsequently, the proteasome, a large protein complex composed of proteases,[26] breaks down HIF-1α and loses its transcription function in a process called proteasomal degradation. However, in an hypoxic environment, hydroxylation (the process of adding a -OH group to a molecule) is inhibited. Non-hydroxylated HIF-1α subunits accumulate, dimerize (combine) with HIF-1β, and bind the HREs (Hypoxia Response Elements) in certain target genes to activate transcription. Due to a lack of proteasomal degradation, HIF-1α is able to over-express several angiogenic growth factors, including VEGF and PDGF, that contribute to intratumoral vessels.[Fig. 2] The hydroxylation of HIF-α subunits negatively regulates their half-life and transcriptional activity.

Fig. 2



The processes of HIF-1α under normal and hypoxic conditions. Under normal conditions, the HIF-1α subunit undergoes hydroxylation by a PHD. This allows the E3 ubiquitin ligase working with pVHL to mark HIF-1α for degradation, leading to a loss of function. Alternatively, the co-activation factor p300 can also inhibit transcription. In hypoxia, non-hydroxylated HIF-1α subunits dimerize with HIF-1β and disrupt the interaction between HIF-1α and p300. HREs are then bound to activate transcription of several pro-angiogenic factors.[8]

Angiogenic Processes in Tumors Contributing to Therapy Resistance

There are many forms of categorized tumor resistance to antiangiogenic treatments. Congenital drug resistance is inherently in a patient's body, and can refer to the genes of the patient and tumor itself.[8] Acquired drug resistance is the second branch that researchers have focused on, and is composed of various mechanisms. Tumors are able to maintain angiogenesis even in the face of anti-VEGF drugs, as factors that are VEGF-independent, such as FGF-2, provide redundant angiogenic signalling as well.[27] The activation of the pro-angiogenic FGF signalling pathway induces mechanisms that tumor cells use to escape from VEGF-targeted therapies. Therefore, inhibition of FGF receptors lead to decreased vessel density and restored tumor sensitivity to anti-VEGF therapy.[25] This can apply to a variety of signaling pathways, such as PlGF.

A major mechanism that allows tumor cells to avoid drugs and therapies is chaotic vasculature. An ordered network of hierarchical vessel division, with clear arteries, arterioles, capillaries, venules, and veins is what is necessary for efficient circulation. Tumor vessels, on the other hand, are characterized by reduced blood flow, endothelial cell sprouting, disruption of endothelial cell junctions, loss of pericytes coverage, increased vessel leakiness. Pro-angiogenic factors in a tumor's microenvironment regulate angiogenesis. Due to pro-angiogenic signalling, the new vasculature may fail to mature and prune, divide into sections, have vessels of different sizes, and chaotic blood flow. This could lead to uneven blood flow within tumor parenchyma, creating areas of persisting hypoxia. In tumor vessels, endothelial junctions are often disrupted, which enhances permeability and increases interstitial fluid pressure. The efficacy of cancer therapy, in turn, reduces as the compression of tumor vessels and poor vascular perfusion hinder drug delivery. This impaired therapeutic delivery may be an example of vascular hyper-permability directly impacting the tumor microenvironment. Specifically, treatment delivery can be affected through VEGF-induced angiogenesis, which, as mentioned, is also increased by VEGF. VEGF induces vascular permeability through several mechanisms such as junctional remodeling, induction of fenestrae, and vesiculo-vascular organelles (VVOs). In cancer, this mechanism is dysregulated and oftentimes leads to hyper-permeability. This can directly change the tumor microenvironment through increased interstitial pressure leading to impaired blood and therapeutic delivery.

Aggressively growing tumor cells can also form vessel-like structures in a process called vascular mimicry. These structures are formed without endothelial cells and are an alternate source of oxygen and nutrients. The cells secrete the collagens IV and VI, proteoglycans, heparan sulfate, laminin, and tissue transglutaminase antigen 2, which is what leads to tubular structure formation and stabilization. A 2012 study found that in uveal melanoma, tumor cells that utilize vascular mimicry have a cancer stem cell phenotype.[28] In gliomas specifically, increased vascular mimicry has been noticed as an unfortunate result of anti-angiogenic therapy.

Aside from tumor angiogenesis, tumors can be vascularized through vascular co-option, where tumor cells directly utilize pre-existing vasculature of non-malignant tissue as a supply of oxygen and nutrients. This suggests that anti-angiogenic treatments could be essentially ineffective. The first evidence of vascular co-option was documented in a 2016 study assessing the effects of the anti-angiogenic Sorafenib for advanced hepatocellular carcinoma. It used an orthotopic human HCC model, which injects animals (typically a mouse) with cancer cells. A biomarker was used to track how these cells responded to the treatment and found that resistant tumors co-opted liver-associated vessels rather than utilizing sprouting angiogenesis.[29]

In hypoxia-induced resistance, the hyperproliferation of tumor cells increases oxygen consumption. It is a feature of solid cancers which arises when there is a mismatch between oxygen supply and consumption. The tumor mass grows to be larger than the blood supply, making the blood hypoxic. Once cancer cells co-opt its system, hypoxia is a main promoter of angiogenesis. This hypoxic environment produces several pro-angiogenic factors leading to rapid and chaotic blood vessel formation to sustain tumors. Increased HIF activity is the result of intratumoral hypoxia. The hypoxia-induced stabilization of HIF-1α promotes the upregulation of pro-angiogenic genes like VEGF, FGF, and PDGF. Mutations in the von Hippel-Lindau (VHL)

protein, which plays a role in the oxygen-signaling pathway that promotes HIF-α proteasome-mediated degradation under normoxia, can impact the stabilization of HIF-1α and the activation of target pro-angiogenic genes. This is seen in many tumors and is associated with tumor progression and poor patient outcome, as well as drug resistance in anti-angiogenic treatment.[30]

Tumors can additionally use angiogenesis to avoid treatment by utilizing the immune system of the body. The immune cells of a tumor's microenvironment depend on the endothelial cells in order to enter the tumor and attack it. However, different kinds of immune cells are a source for soluble factors that can influence angiogenesis. The white blood cell macrophages are specialized phagocytes that clear invading microbes and cell debris, present antigens to the adaptive immune system, and release various immunomodulatory cytokines (signaling proteins). Tumor-associated macrophages (TAMs) are known to modulate and support angiogenesis. They can have different phenotypes depending on their microenvironment, but generally resemble M2 macrophages. M2 macrophages are associated with immune suppression, tissue remodeling, and angiogenesis. Under hypoxia, TAMs produce proangiogenic factors that facilitate the proliferation of endothelial cells, induction of sprouting, tube formation, and maturation of new blood vessels, all promoting angiogenesis. TAMs release a variety of angiogenesis-modulating molecules that act in synergy to trigger the degradation of the basement membrane and extracellular matrix components. This destabilizes the vasculature and promotes migration and proliferation of endothelial cells. TAMs can also inhibit the expression of angiogenesis inhibitors.

Myeloid-derived suppressor cells (MDSCs) are a heterogeneous population of immature cells originating from the bone marrow that expand and accumulate under pathological conditions, such as cancer. Their capacity to regulate tumor angiogenesis is similar to M2. MDSCs promote tumor angiogenesis through secretion of matrix metalloproteinases (MMPs). MMPs are a family of zinc- and calcium-dependent enzymes that break peptide bonds within a chain. MMP-9 is the central protease for protein and polysaccharide-filled extracellular matrix, which allows for endothelial cell migration.[7] This increases the bioavailability of VEGF to further trigger MSDC recruitment,[31] which boosts angiogenesis and stimulates tumor neovasculature. MMP-2 controls angiogenesis and tumor growth,[32] and MMP-14 promotes the formation of blood vessel lumen.[31] MDSC accumulation in tumors correlates with intratumoral VEGF concentration during disease progression. In the presence of VEGFs, MDSCs can create a pro-angiogenic environment within the tumor by secreting angiogenic factors. This promotes MDSC accumulation in tumors as it induces resistance to anti-angiogenic therapy. MDSC ablation also has synergistic effects with anti-VEGF/VEGFR treatment.

Another form in which treatment is resisted is through the recruitment of stromal cells, immune cells, and progenitors. Specifically, bone-marrow derived cells (BMDCs) can cause resistance to the anti-angiogenic vatalanib, so the depletion of these cells can potentiate the drug's effects.[33] The release of proangiogenic factors in response to blockers can recruit endothelial progenitor cells (EPC) from the bone marrow, which contribute to tumor vascularization and resistance to anti-VEGF therapy. Similarly, the recruitment of pro-angiogenic myeloid cells allows tumors to bypass the inhibitory effects of anti-angiogenic therapy. These cells can be used as an alternative source of pro-angiogenic chemokines and cytokines.

Chemokines are a family of secreted proteins that contribute to angiogenesis either directly or indirectly. They can bind chemokine receptors expressed on endothelial cells or recruit inflammatory cells and progenitors. Cytokines are signaling proteins that control inflammation in the body.[34] The inhibition of the VEGF signalling pathway also creates alternative pro-angiogenic signalling pathways, like ANGPT-2, FGF-2, and IL-8.

Treatment Strategies Targeting Angiogenesis:

Anti-angiogenic drugs typically utilize one of three mechanisms capitalizing on angiogenesis. In vessel depletion, tumors are “starved” by blocking their vessels. However, this treatment has been unsuccessful as a monotherapy due to the previously-mentioned several mechanisms that tumors can utilize to counter the lack of oxygen and nutrients, including hypoxia-induced resistance, vascular mimicry, vessel co-option, among others. Vessel normalization is another strategy that aims to target the chaotic vasculature hindering drug delivery and treatment. A hypothesis introduced by Jain[35] in 2001 suggests that sub-maximal doses of anti-angiogenic therapy can restore the normal structure of vessels to improve therapeutic administration. This hypothesis could be a reason for why chemotherapy treatment is more effective when used in combination with anti-angiogenic therapy, as it creates a short “time window” for a blood vessel to exhibit normal structure,[36] rather than just as a monotherapy. In the mechanism of immune activation, adhesion molecules that allow immune cells to enter the tumor are up-regulated. Typically, tumors escape immune cell infiltration by down-regulating these adhesion molecules of endothelial cells, a process which immune activation counteracts.

Anti-VEGF therapy is a combination of all three of the aforementioned mechanisms. It depletes vessels and can promote T cell infiltration. It can also induce an early and transient phase of vessel normalization, a window of equal pro-angiogenic and anti-angiogenic factors, promoting tumor drug delivery and efficacy. Anti-angiogenesis as a monotherapy creates only limited benefits in certain tumor types like advanced-stage renal cell carcinoma, hepatocellular carcinoma, and colorectal carcinoma. This could be due to increased tumor aggressiveness after anti-angiogenic drug administration. In anti-VEGF monotherapy, agents bind to VEGF and interfere with its ability to activate its receptors. The first in which this therapy takes form is through the use of a monoclonal antibody, bevacizumab, and a soluble decoy receptor construct (VEGF trap) (aflibercept). Bevacuzimab is an FDA-approved humanized monoclonal IgG1 antibody against VEGF-A. It binds to VEGF-A and prevents interaction with the VEGF receptor. It is to be used in combination with chemotherapy, as it is only used as monotherapy for recurrent glioblastoma. However, in a 2012 clinical trial assessing the effectiveness of VEGF-inhibitor bevacizumab on colorectal cancer, patients who had been adjuvantly treated with it were more likely to relapse due to an increased tumor progression after anti-angiogenic therapy.[37] Aflibercept is a fusion protein—a complex formed through genetical engineering by binding a segment of immunoglobulin to a functional protein[8]—of VEGF receptors 1 and 2. It binds to VEGF-A and -B and placental growth factors with more affinity than bevacizumab. The second way that anti-VEGF monotherapy is completed is through the inhibition of VEGFR activation through a monoclonal antibody (ramucirumab) or receptor tyrosine kinase inhibitor (RTKI). A fully humanized IgG1 antibody with a high affinity to VEGFR-2 inhibits ligand binding by targeting its extracellular domain. Ramucirumab was approved by the FDA in 2014 to treat

gastric adenocarcinoma.[8] RTKI results in the inhibition of the downstream signal transduction pathway by blocking phosphorylation. The first anti-angiogenic small-molecule RTKI was sorafenib, approved by the FDA to treat renal cell carcinoma in 2005.[38] It blocks the tyrosine kinase receptor Raf of the downstream Ras/Raf/MEK/ERK pathway that is also potentially activated by VEGFR-2 and inhibits the phosphorylation of kinase receptors such as VEGFR and PDGFR.[39] Other growth factor receptors like FGF receptors also have a tyrosine kinase domain. There are eight anti-angiogenic FDA-approved RTKIs that target the VEGF receptor—sorafenib, sunitinib, pazopanib, regorafenib, cabozantinib, axitinib, vandetanib and lenvatinib.

The FDA approved that anti-angiogenic therapy can be successfully used to treat cancer after a 2003 clinical trial in which a combination of chemotherapy and anti-VEGF therapy was proven to prolong survival of patients with metastatic colorectal cancer. Targeting VEGF/VEGFR signalling can increase the expression of adhesion molecules, allowing T-cells to enhance the efficacy of cancer immunotherapy.[40] Aside from chemotherapy, anti-angiogenic therapy can be also used in combination with surgery, radiation therapy, immunotherapy, and other agents.[41]

Anti-VEGF drugs can also be combined with immunotherapy to reverse negative effects that excess VEGF would have had on the body's immune system. A potential pathway VEGF takes to weaken immune cells' anti-tumor effects is through the upregulation of immunosuppressive cells like MDSCs and M2-like tumor-associated macrophages activity.

Another treatment strategy used to block tumor blood supply is embolization, in which a minimally-invasive procedure is performed to stop blood flow to a tumor. It delivers tiny particles, known as embolic agents, through a catheter inserted towards the treatment area. Examples of embolic agents include balloons, liquid glue, liquid sclerosing agents, metallic coils, and particulate agents.[42] However, as embolization increases hypoxia in a tumor, it may still up-regulate the release of more angiogenic factors to further expand any existing vasculature.[43]

Conclusion

Although advancements have been made towards success in anti-angiogenic therapy, there still exist several mechanisms that tumor cells utilize that negate the effects of this therapy, as well as more widespread, effective ones such as chemotherapy. These mechanisms encompass a wide range, from exploiting the state of hypoxia, to tumor blood vessel formations to even simple congenital drug resistance. However, positive results are more likely to be attained when combination therapy is administered, allowing for the tumor-reducing impact of chemotherapy to work effectively with a minimal threat of tumor avoidance or regrowth from angiogenesis. Research into other combinations of therapies, aside from the already-explored anti-VEGF and chemotherapy treatments, could expand the scope of potential clinical outcomes. The strategy of vascular normalization provides a promising destination for anti-angiogenic therapy to be rerouted, as the resulting stabilized blood vessels ensure better therapeutic delivery for other treatments. Furthermore, more research into other lesser-explored angiogenic pathways, such as FGF, could provide significant findings that research into the prominent VEGF pathway may not, especially as they are alternatively utilized by the tumor to evade anti-VEGF treatment.

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