

Angiogenic switch in Barrett's adenocarcinoma: the role of vascular endothelial growth factor

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1. ABSTRACT

The development of cancerous cells from the normal cells is the consequence of multiple genetic and epigenetic abuses. Activation of "Angiogenic switch" or formation of new blood vessels is one of the upshots of these abuses. Multiple factors are associated with the activation of angiogenic switch. Vascular endothelial growth factor (VEGF) and its down stream signaling molecules is important troupe of this event. In this article, we reviewed the role this troupe in the development of Barrett's adenocarcinoma and also discussed the possible remedies, which have the impact on blocking the function of this troupe.

2. INTRODUCTION

Barrett's esophagus is an acquired histopathological condition in which the innermost squamous epithelial lining of the esophagus is replaced by columnar epithelium with characteristic goblet cells (1). Indigestion and acidity leading to gastro esophageal reflux disease (GERD) is the crucial factor behind such gradual cellular replacement (2-4). The exact cause and mechanism of such cellular replacement is yet to be accurately deciphered. It is conjectured that during reflux, HCl of gastric juice and bile of duodenum induce slow but

steady alteration in the basal cellular layer of the lining of the esophagus (5,6). GERD induces biochemical as well as mechanical stress to the cells, which gradually induce this slow replacement (7, 8). In addition to GERD, adenocarcinoma of the esophagus is also attributed to obesity, higher socioeconomic status and the Caucasian race (9). The cellular replacement event is a slow and gradual process and it exhibits several stages of progression which is often characterized as low and high grade dysplasia (LGD and HGD) which ultimately give rise to Adenocarcinoma. Thus Barrett's esophageal condition is the initiation of this iceberg – "**Esophageal adenocarcinoma**" which has been gaining the status of the most dreadful cancerous lesion in the Western world and at present 5 years survival rate is still below 10% (10, 11).

The basic cause of every type of malignancy is undoubtedly the chromosomal or point mutations which lead to the deviation from normal pattern of cell cycle progression that results in altered growth and differentiation profiles that ultimately lead to malignancy (12, 13). Whatever the nature and site of malignancy, the prime requisite of malignant growth is constant supply of nutrients which is aided by neovascularization/angiogenesis. Thus angiogenesis is the

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basic event which helps to grow and differentiate the malignant tissues (14). Formation of new blood capillaries from the old one is a multistep process (15). At first the diseased tissue produce angiogenic growth factors which diffuse into the surrounding tissues and bind to specific receptors located on the endothelial cells of the surrounding preexisting blood vessels (16). Once the endothelial cells are activated they send specific signals to the nucleus producing specific enzymes leading to enhanced proliferation of the endothelial cells that escape through the pores of the basement membrane and eventually converge to form vessels that are supported and strengthened by the pericytes and smooth muscles (17). This protein for vascular endothelial cells is known as vascular endothelial growth factor (VEGF) which is a homodimeric glycoprotein of relative molecular mass 45,000, and is the only mitogen that specifically acts on endothelial cells (18). A number of different proteins (factors) are known to be structurally related to VEGF such as Placental growth factor (PlGF), VEGF-B and VEGF-C (19). Thus VEGF (now is known as VEGF-A) is the founder member of this family of pro-angiogenic proteins. VEGF was originally recognized in a tumor-conditioned medium but recent studies have demonstrated that it is expressed by various types of cells including tumor epithelial cells, vascular smooth muscle cells, monocytes, and megakaryocytes, in some of which the expression is constitutive (20).

The families of vascular endothelial growth factor (VEGF) proteins include potent and highly specific mitogens for vascular endothelial cells that function in the initiation of tumor stromal angiogenesis, vascular hyperpermeability, and vasodilatation (21-23). It basically renders the blood vessels highly permeable. For this reason it is also referred to as vascular permeability factor (VPF) (24, 25). In addition to this, it exerts a number of other important actions on vascular endothelium (26). VEGF's capacity to increase microvascular permeability is the most potent activity among the known cytokines (27). However other than VEGF A, only VEGF-C possess this micro vascular permeability activity (28). The role of VEGF-C in the induction of esophageal squamous cell carcinoma and esophageal lymphangiogenesis has been confirmed by several workers (29-31). VEGF is the most potent angiogenic molecule and participates in the tumor angiogenesis of various cancers like hepatocellular, esophageal, and pancreatic cancer. It is also revealed that VEGF is one of the molecules which is responsible for metastasis and prognosis of different cancers. However, this is not always the case, because it is reported that there is no prognostic relation with VEGF and the stage of esophageal tumor. (32). VEGF gene induces neovascularization in and around tumor that augment metastasis potential by accelerating proliferative activity after reaching the target organ (33).

VEGF is expressed as five splice variants encoded by a single gene - (VEGF121, VEGF145, VEGF165, VEGF189 and VEGF206) (34). Among the cell-associated isoforms, VEGF₁₈₉ plays the most crucial role in establishment of human colon and esophageal cancer. (35). However, Nagata (36) observed VEGF 121, 165 and 189 in the angiogenesis of esophageal carcinoma.

In some cases VEGF level maintains a close relation with the levels of other growth factors in the development of esophageal carcinoma like basic Fibroblast Growth Factor (37-39), Angiopoietin-1 (40) and Interleukin-10 (41). In patients with renal cell carcinoma, VEGF and PlGF levels were significantly higher. Their levels were noted to be significantly associated with histological grade and total tumor vascularity (TTV) (42). The action of VEGF-A is largely regulated by multiple factors including hypoxic, growth factors and steroid hormones. The expression of VEGF-A gene is reported to be regulated by 17- β estradiol and its action is mediated by ER- α dependent pathway (43). Its action is also known to be enhanced by 2ME₂, as well as 17 α -estradiol, and is modulated by ER- α dependent and independent pathways. (44). VEGF's action on cells is mediated by interactions with two high affinity receptors - VEGFR-1 (also known as Flt-1) and VEGFR-2 (also known as KDR in man and Flk-1 in rodents). Both receptors express predominantly on vascular endothelium (21). However VEGFR are substantially over expressed in many human and animal tumors (45) and although VEGF expression correlates significantly with co expression of its receptors, VEGFR-1 and VEGFR-2 do not appear to contribute directly to the development of esophageal squamous cell carcinoma (46). Our personal observation, however, confirms that the expression of VEGF receptor KDR/VEGFR-2 strongly correlates with the expression of VEGF in the development of Barrett's adenocarcinoma (Unpublished data).

Due to high potentiality of VEGF in augmenting tumor angiogenesis, most of the recent research workers have been paying attention to investigate the role of VEGF in tumor angiogenesis (47-56). Many workers have investigated the role of VEGF in squamous cell carcinoma of esophagus (57-61) but comparatively little attention has been paid to the evaluation of its role in the angiogenesis of Barrett's adenocarcinoma (62-65) leading to esophageal adenocarcinoma. With a view to this fact, the goal of this commentary is to review the up to date views of the recent investigators exploring the role of VEGF in the angiogenesis of esophageal adenocarcinoma and the possible role of VEGF inhibitors in the control of angiogenesis.

3. ANGIOGENESIS IN ESOPHAGEAL MALIGNANCY AND VEGF

In addition to the genetic alterations the most effective patho-physiological factor for tumor propagation is the induction of neovascularization/tumorogenesis for adequate supply of oxygen, and metabolites and for the effective removal of waste products (66). Induction of the antigenic switch depends on the balance in favor of proangiogenesis which mainly occurs in response to physiological stimuli like hypoxia, oncogenic activation, tumor suppression mutation (67,68) or even 17 β -estradiol (Sengupta et al 2004). It has been noted that tumor size remains static in the absence of neovascularization while it grows exponentially in the presence of new blood vessels. Tumor angiogenesis largely depends upon the metabolic need of the tissue particularly in response to increased tissue mass.

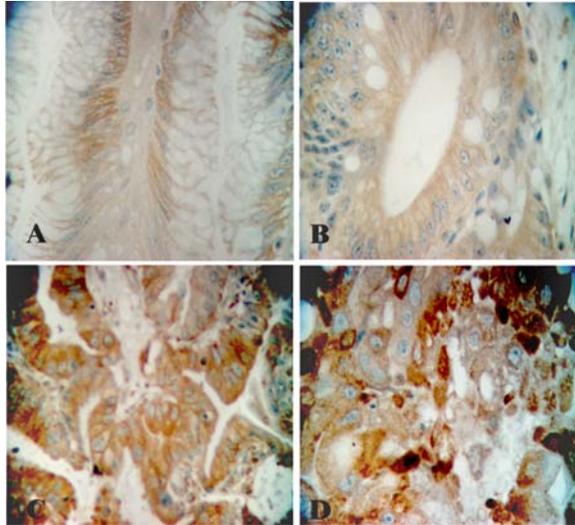


Figure 1. Immunohistochemical localization of VEGF-A (brown color) in NBE, LGD, HGD and cancer samples. (A) Normal, (B) LGD, (C) HGD and (D) cancer. VEGF expression was noted in the membrane of NBE but both in the membrane and cytoplasm of other grades particularly in HGD.

Tumor blood vessels are structurally different from normal blood vessels and appear to be leaky and hemorrhagic in response to the release of antigenic factors, particularly VEGF/VPF (69). VEGF mobilizes endothelial precursor cells from the bone marrow which are carried to the tumor via blood stream and carries out vasodilatation followed by leakage of plasma proteins in the perivascular matrix (70). The pericytes covering the vessels are then loosened followed by degradation of basement membrane and extra cellular matrix allowing migration of endothelial cells towards the chemotactic angiogenic stimuli (71). VEGF might be accumulated in the provisional matrix during initial steps of angiogenesis and contribute to the survival of migrating endothelial cells (72). The endothelial cells then multiply in the perivascular space and congregate around a lumen which is accompanied by basement membrane formation, pericyte involvement and fusion of adjacent vascular sprouts to form new vessels (73). Angiogenesis is thus a prime factor for tumor development but the phase of neovascularization varies according to the type of tumor. Usually tumor associated angiogenesis proceeds through two phases. The first phase is called avascular phase in which tumor proliferation and rate of apoptosis are so balanced that the tumor size remains static. However, in the second phase the tumor size grows exponentially with concomitant development of blood vessels.

Neovascularization is usually a prerequisite for tumor progression but all tumors do not require profuse blood supply. In fact blood vessels first regress as tumor size progresses and later on hypoxic conditions induce neovascularization which is aided by hypoxia induced antigenic factors - VEGF and VEGFR-2 in association with angiopoietin-2 (ANG2) (74).

VEGF also regulates vessel diameter. Low concentrations of both VEGF₁₂₁ and VEGF₁₆₅ promote growth of long, thin vessels, whereas higher concentrations of VEGF remarkably enhance vessel diameter. Placental growth factor, which binds to VEGFR-1 but not VEGFR-2, does not promote capillary sprouting. Moreover, specific inhibition of VEGFR-2 signaling results in a dramatic reduction of EC sprouting in response to VEGF, indicating the critical importance of this receptor. The increase in vessel diameter is the result of cell proliferation and migration, rather than cellular hypertrophy (75). The expression of VEGF has strongly been correlated with micro vessel diameter during the progression of esophageal squamous cell carcinoma (76,77)

VEGF isoforms show differential reactions to the extracellular matrix components which are mainly regulated by the qualitative and quantitative differences in the amino acid contents. An important biochemical property that distinguishes the larger VEGF isoforms from VEGF₁₂₁ is their ability to bind to heparin and heparin-sulfate of the extra cellular matrix (ECM) (78). Because of the binding capability, VEGF₁₄₅ and VEGF₁₆₅ can be localized to the cell surface and ECM, while VEGF₁₈₉ and VEGF₂₀₆ are almost completely sequestered in ECM, whereas VEGF₁₂₁ is a soluble form without binding to ECM. However, their potential role as true components of the extracellular matrix has not yet been investigated (79). It has been experimentally noted that extra cellular matrix can serve as a storage depot for VEGF₁₈₉ and VEGF₂₀₆ because they show greater affinity towards heparin sulphate (80). These isoforms also exhibit differential affinity towards different VEGF receptors on the endothelial cells. The VEGF family includes VEGF- B to VEGF- E with minor variations. VEGF-B occurs as two splice variants with 167 and 188 amino acids residues and can form heterodyme with VEGF₁₆₅. VEGF-C and D are prototypically processed molecules that are involved in the formation of lymphatic endothelium. VEGF-E is recently known as a viral form of VEGF (81). VEGF expression appeared to be related with matrix metalloproteinase-9 (MMP-9) showing close relationship with micro vessel density in relation with the development of esophageal carcinoma (82)

The cell-associated isoform, VEGF₁₈₉, plays important roles in establishment of human esophageal cancer. The isoforms VEGF₁₂₁/VEGF₁₆₅ was expressed in 11.8% while the expression of VEGF₁₂₁/VEGF₁₆₅/VEGF₁₈₉ was noted in 88.2% cases (83). Nagata J, (84) noted the expressions of VEGF₁₂₁, VEGF₁₆₅ and VEGF₁₈₉ isoforms in 93.3%, 55.6% and 26.7% esophageal carcinoma cases, respectively.

Our studies (manuscript in preparation) confirm prominent expression of VEGF-A in normal esophagus. It expressed significantly in the membranes as well as in the cytoplasm. However, this growth factor was noted to be differentially expressed in low grade dysplasia (LGD), high grade dysplasia (HGD) and tumors. Expression of VEGF-A is significantly higher in HGD and cancer specimens (Figure 1).

4. ROLE OF THE ANGIOGENIC FACTORS

VEGF and basic fibroblast growth factor (bFGF) in relation to the development and progression of Barrett esophagus and adenocarcinomas of the esophagus is reported, that their expression levels were significantly increased in adenocarcinoma in comparison to normal squamous mucosa and dysplastic tissues (85). Esophageal adenocarcinoma presents a specific 'angiogenic switch' in the early periods of the development of metaplasia-dysphasia-carcinoma sequence and neovascularization phase often precede tumor growth (86). Micro vascular density (MVD) is reported to be associated with the expression of VEGF which were noted to increase significantly from metaplasia to HGD but not between HGD to adenocarcinoma (87). Angiogenesis of esophageal basaloid squamous carcinoma (BSC) and squamous cell carcinoma (SCC) were correlated with strong expression of VEGF and it has been experimentally noted that VEGF may participate in angiogenesis of esophageal BSC and may influence the rate of metastasis in esophageal BSC patients (88). The relationship between the expression of VEGF, MVD and esophageal carcinomas were noted to be closely correlated with the degree of differentiation and lymphatic metastasis, but not with the depth of cancer invasion (89). In early stages, the rate of expression of VEGF and MVD appear to be lower than in progressive stage (90) The expression of VEGF and MVD play an important role in tumor angiogenesis, growth and metastasis. MVD and the expression of VEGF may be two important indexes for patients' prognosis (91). But so far the literatures are concerned no such account related to VEGF and MVD in Barrett's adenocarcinoma is yet to be reported.

5. VEGF SPLICE VARIANTS

The VEGF gene has eight exons, seven introns, and a coding region of around 14 kilo bases (92). Alternative splicing of VEGF mRNA generates five different isoforms (i.e., VEGF₁₂₁, VEGF₁₄₅, VEGF₁₆₅, VEGF₁₈₉, and VEGF₂₀₆) encoded by a single gene (93). The three shorter isoforms (VEGF_{121,145} and ₁₆₅) are mainly diffusible while the two longer ones are sequestered in cell membranes after secretion. The existence of different isoforms indicates their differences in the mode of synthesis, organ specific distribution and biological roles (94). The isoforms share the same function, and the main difference among them lies in their ability to bind to heparin. Such differential heparin-binding properties are related to the bioavailability of the several isoforms (95). VEGF protein appears to become available to endothelial cells in at least two ways: as either freely diffusible proteins (VEGF₁₂₁ and ₁₆₅ in humans) or after protease activation and cleavage of the longer isoforms bound to the ECM (96). Among the factors that upregulate VEGF are the ovarian steroid hormones (97) and prostaglandins. VEGF is produced in the local tissues under various pathological conditions such as cancer, wound healing, ischemic myocardium, diabetic retinopathy and other abnormalities (98-100). In these tissues, VEGF₁₂₁ and VEGF₁₆₅ are commonly expressed, and the expression of VEGF₁₈₉ is

also frequently observed. In contrast, VEGF₁₄₅ expression is restricted to the cells derived from reproductive organs and VEGF₂₀₆ is a rare form expressed in placental tissue (101). VEGF₁₂₁ and VEGF₁₆₅ cause vascular permeability and actively promote angiogenesis and induce proliferation of vascular endothelial cells (102). VEGF₁₂₁ and VEGF₁₆₅ were found to be more potent at inducing inflammation. In vitro, VEGF₁₆₅ has been noted to stimulate more potently intracellular adhesion molecule (ICAM)-1 expression on endothelial cells. VEGF₁₆₄ was also more potent in inducing the chemotaxis of monocytes (103). In an immortalized human leukocyte cell line, VEGF₁₆₅ was found to induce tyrosine phosphorylation of VEGF-R1 more efficiently. These data identify VEGF₁₆₅ as a proinflammatory isoform and identify multiple mechanisms underlying its pro inflammatory biology (104). VEGF₁₈₉ and VEGF₂₀₆ are also assumed to be capable of inducing endothelial cell proliferation (105).

6. VEGF RECEPTORS AND THEIR MULTIPHASIC ROLES IN INDUCING ANGIOGENESIS

Vascular endothelial growth factor (VEGF) being the key regulator of physiological and pathological angiogenesis associated with tumors is reported to exert its action through receptor tyrosine kinases called VEGFR-1 (fms-like tyrosine kinase - Flt-1), VEGFR-2 (Kinase insert domain containing receptor - KDR/Fetal liver kinase KDR/Flk-1) and VEGFR-3 (Flt-4). VEGFR-1 is a receptor for VEGF-A and VEGF-B. The ligands for VEGFR-2 are VEGF-A, C, D and E while VEGFR-3 binds VEGF-C and D only. Kinase insert domain containing receptor - KDR/Fetal liver kinase (Flk1) (VEGFR-2), and flt1(VEGFR-1) are expressed predominantly in vascular epithelium but differ considerably in signaling properties. VEGFR are substantially over expressed in many human and animal tumors (106) and although VEGF expression correlates significantly with co expression of its receptors, VEGFR-1 and VEGFR-2 do not appear to contribute directly to tumor progression (107). VEGF receptors are of three main types: VEGFR-1,2 and 3. These bind with 4 ligands VEGF A, B, C and D. VEGF-A and VEGF-B are significantly more abundant in adenomas compared to normal tissues, while VEGF-A and VEGF-C are significantly more expressed in carcinomas as compared to normal tissues. Significantly greater amount of VEGF-C expression has been noted in carcinomas compared to adenomas whereas there was a significant reduction of VEGF-B in carcinomas compared to adenomas. VEGF-D appears to be significantly more abundant in normal tissues than in adenomas and carcinomas. VEGF-A and VEGF-B play a critical role in early tumor development at the stage of adenoma formation while VEGF-C plays a role in advanced stages of cancer progression (108). Soker et al (109) reported that Neuropilin-1 (NRP-1) is an isoform-specific co-receptor of VEGFR-2, and enhances the bioactivity of VEGF₁₆₅ by increasing the binding affinity of the molecule to VEGFR-2. Among the receptors, NRP-1 is unique in that it acts as a co-receptor specific to VEGFR-2 and enhances the signal of VEGF₁₆₅. NRP-1 is a cell-surface receptor for both VEGF₁₆₅ and class 3 semaphorins that is expressed by neurons and endothelial cells. NRP-1

and VEGF activities were interdependent and NRP-1 regulates angiogenesis through a VEGF-dependent pathway (110). It is further reported that after VEGF stimulation, a soluble form of receptor - tie-1 is released from the endothelial cells and is mediated by a membrane associated metalloprotease. It may participate in regulating the bioavailability of a growth factor. Such receptors from membrane spanning receptor tyrosine kinases exhibit extra cellular ligand binding domain. These truncated forms contain all the structural features required to bind their ligands. VEGFR-1 and VEGFR-2 were originally thought to be expressed predominantly in endothelial cells, but recent studies have indicated that other types of cells also express one or both of the receptors; VEGFR-1 is expressed in trophoblasts, monocytes, and mesangial cells, and VEGFR-2 is expressed in hematopoietic stem cells, megakaryocytes, and retinal progenitor cells (111). VEGFR-1, 2 and 3 all share some common features, all show seven immunoglobulin-like-domains involved in ligand binding in their extra cellular part.

Neovasculogenesis from the precursor cells is mainly regulated by transmembrane VEGF receptors. It involves differentiation of haemangioblastic cells and their assembly into a primitive vascular plexus. Both VEGFR-1 and VEGFR-2 play a vital role in the development of angiogenesis. VEGFR-2 is found to play role in the early stages of vascular development and seems to mediate differentiation of precursor cells, migration and proliferation of endothelial cells while, VEGFR-1 is found to play role in later stages of vascular development. Even VEGFR-3 which mainly regulates lymphangiogenesis also plays crucial role in the development of blood vessels at the early stages of development. Binding of VEGF to its receptors on the endothelial cells stimulates not only their proliferation but also production of extra cellular matrix (ECM) degrading matrix metalloproteinases which plays a key role in the degradation of basement membrane of blood vessels and their surrounding extra cellular matrix (ECM) facilitating endothelial cell migration (112). In the genome of Orf virus, a VEGF homologue, - VEGF-E has been discovered, which binds with VEGFR-2 as well as neuropilin-1 (63). In addition to the occurrence of normal receptors, soluble VEGFR-1, 2, and 3 has been reported in mammals. The exact role of sVEGFR is still uncertain though it is speculated that sVEGFR act as a decoy receptor that reduce VEGF availability that inhibits overgrowth of endothelial cells in the lumen of the blood vessels. It binds with different isoforms of VEGF and does not require any accessory protein for ligand binding on the cell surface.

Our preliminary observation (manuscripts in preparation) depicts strong correlation in the expression of VEGF receptors Flk-1 and co-receptor NRP-1 during the progression of Barrett's adenocarcinoma through low and HGD. The expressions of VEGFR-2 were prominent both in the membrane as well as in the cytoplasm while NRP-1 expression was mainly noted in the membrane. The levels of expressions were higher in the adenocarcinoma samples than normal Barrett's esophagus (Figure 2).

7. MODE OF ACTION OF VEGF

Tumor growth and angiogenesis are complementary to each other because growth of tumor depends fully on steady blood circulation for the supply of nutrients as well as excretion of waste products. It is well established that angiogenesis is necessary for the growth and metastatic spread of solid tumors. Neovascularization is directly related with increased metabolic load or tissue mass. Increased tissue mass leads to local deficit of oxygen reflecting the need for vessels and inducing angiogenesis in a hypoxic fashion. Numerous convincing evidences have been generated to suggest that neovascularization is mainly mediated by VEGF and its mitogenic receptor VEGFR-2, in a hypoxia-driven paracrine manner (113).

The basic mode of action of VEGF has extensively been studied in different tumors and animal model but no such convincing report is available regarding the mode of action of VEGF in the development of esophageal carcinoma. However, the general pathways of the mode of action of VEGF may be applicable to esophageal tumorigenesis also. Tumor cells secrete various proangiogenic factors like fibroblast growth factor (FGF) and VEGF (VEGF-A, C, D and perhaps another VEGF family member P1GF) among which VEGF-A is the most ubiquitous because its induction is under ischemic condition in response to increased tissue mass and inability of the existing blood vessels to supply sufficient nutrition.

The lining of the existing blood vessels comprise basement membrane, pericytes and vascular endothelial cells. During angiogenesis in response to VEGF the vascular basement membranes (BM) of the existing blood vessels degrade by several matrix-degrading enzymes like matrix metalloproteinase (MMP) (produced by endothelial cells or by tumor cells or by the immune cells that accumulate around the neoplastic cells (114,115). Degradation of BM causes liberation of endothelial cells from their cellular anchors (integrins) and their subsequent migration and proliferation along with detachment of pericytes from the vessel walls. From the basement membrane, tumor cells and immune cells, the growth factors are released which lead the endothelial cells to adhere to the wall of the capillary tubes (116). These bound endothelial cells are primarily quiescent and remain surrounded by an array of interstitial provisional matrix components like vitronectin, fibronectin, type I collagen and thrombin (117). These molecules provide proliferative cues while the assembled BM matrix molecules extracellular matrix like type IV, XV, XVIII collagen, laminin, heparin sulphate proteoglycans, perlecan, nidogen/entactin and SPARC/BM-40/osteopontin inhibit proliferation and maintain an environment for intercellular adhesion (118). The enzyme MMP-9 and, to a lesser extent, MMP-2 are required for mobilization of the adhered VEGF to initiate angiogenesis(119). MMP-9 effectively degrade type IV collagen effectively facilitating the disruption of basement membrane leading to the release of BM bound VEGF (120).

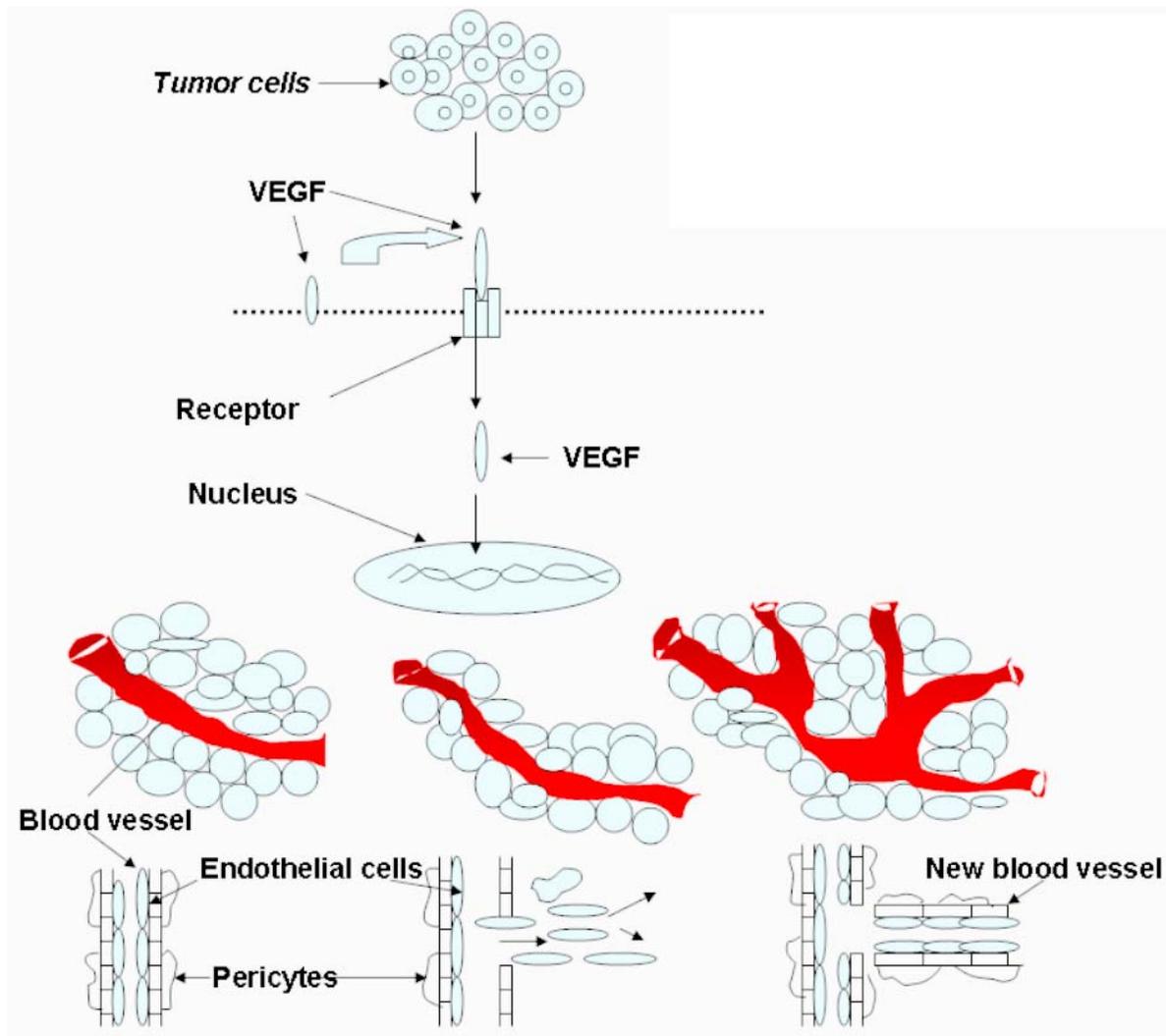


Figure 2. Diagrammatic representations of the autocrine-paracrine role of VEGF in angiogenic switch during the development of Barrett's adenocarcinoma.

VEGF after releasing from its source (dimeric form) come in contact with the receptors on the surface of the endothelial cells. The small domain of the VEGF extending from the core of the VEGF bind to heparin on the extracellular matrix modulating the activity of VEGF. The portion of the receptor inside the cell is a tyrosine kinase; VEGF actually brings together two receptors so that inside the cell two kinase portions are close enough to add phosphate groups to each other. These phosphate groups are recognized by the signaling apparatus inside the cell to initiate the process of angiogenesis.

VEGF mainly mediates its action of neovascularization by two pathways – paracrine and autocrine pathways. In the paracrine pathway, VEGF, after entering into the cytoplasm of the endothelial cell, directly activates several activating molecules and enters into the nucleus and transactivates some genes that regulate cell proliferation by MAPK or ERK pathways. In the autocrine pathway, VEGF, after releasing from the tumor

cells or tumor blood vessels is again received by the receptors of the same cell and initiate angiogenesis by the same process as in paracrine mechanism (121). The enzymes produced as a result of such activation mechanism escape through the pores of the basement membrane and eventually converge to form vessels that are supported and strengthened by the pericytes and smooth muscles. Tumor cells through mutation, gain the ability to create abnormally large amounts of VEGF or to block the action of angiogenic inhibitors. This is often termed the "angiogenic switch," marking a key transition as cancer cells gain the ability to direct their own blood supply. Given a ready blood supply, the tumor can grow much larger.

8. ANTIANGIOGENIC THERAPY AND FUTURE DIRECTION

On the basis of the importance of angiogenesis there is an attractive process for the design of cancer therapy by selectively inhibiting the growth of new blood

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vessels; starving tumor cells. Effective methods have been developed by using drugs or antibodies to block the formation of VEGF or the binding of VEGF to its receptors. Researchers have found, however, that these methods are effective for stopping the growth of a tumor, but generally not for reducing the size of an existing tumor. They are powerful tools, however, when used in combination with agents that attack other key points in the tumor cell.

The antiangiogenic factors inhibit the growth of the primary tumor as quick as before so that the spread of the disease becomes stabilized. They also hinder the accessibility of the cancer cells that have already spread to other organs from gaining blood supply so that they may remain dormant or harmless. Large tumors may shrink when antiangiogenic drugs are combined with chemotherapy regimens or with radiation therapy (122).

The antiangiogenic drugs commonly used are interferon alfa-2a, Thalomid, Celebrex, Avastin, Irinotecan, 5-FU/Leucovorin, PTK787/zk222584 (PTK787), BAY 43-9006, Neovastat, Revimid, SU 11248, Vioxx etc. These drugs commonly target both angiogenesis by blocking the actions of VEGF as well as other growth factor – platelet derived growth factor (PDGF) (123). Therefore, the targeting of VEGF in Barrett's esophageal cancer by employing above or new anti-angiogenic remedies might help in blocking or preventing the disease.

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