Review

Predictive Value of Serum VEGF Levels in Non-Small Cell Lung Cancer: A Review

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Predictive Value of Serum VEGF Levels in Non-Small Cell Lung Cancer: A Review

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Abstract: Vascular endothelial growth factor (VEGF) and its receptors (VEGFRs) serve an essential role in tumor angiogenesis and have emerged as potential therapeutic targets in lung cancer. This review explores the significance of serum VEGF levels as a predictive biomarker in non-small cell lung cancer (NSCLC). The VEGF family, consisting of VEGFA, VEGFB, VEGFC, VEGFD, and placenta growth factor (PlGF), engages with specific receptors, including tyrosine kinase receptors (VEGFR-1, VEGFR-2, and VEGFR-3) and neuropilin receptors (NRP-1 and NRP-2), to promote angiogenesis and lymphangiogenesis. VEGF-A, the primary component of the VEGF family, binds to VEGFR-2 to stimulate endothelial cell proliferation and migration, while VEGF-B, VEGF-C, and VEGF-D interact with VEGFR-1 and VEGFR-3 to regulate tumor angiogenesis, lymphangiogenesis, and metastasis. The VEGF/VEGFR signaling pathway activates various downstream effectors, including phospholipase Cγ1, MAPK, and PI3K/Akt, which are essential for maintaining vascular homeostasis and promoting angiogenesis. In NSCLC, elevated serum VEGF levels have been observed, and the VEGF/VEGFR axis is frequently impaired, leading to irregular blood vessel formation and metastatic spread. Despite the development of anti-VEGF therapies, their impact on lung cancer outcomes has been limited. Further research is needed to optimize the effectiveness of these treatments and elucidate the potential of serum VEGF as a predictive biomarker in NSCLC.

Keywords: non-small cell lung cancer (NSCLC); vascular endothelial growth factor (VEGF); VEGF receptors (VEGFRs); angiogenesis; biomarker; tumor angiogenesis

1. Introduction

Lung cancer is a significant contributor to cancer-related mortality in humanity. Due to its hostile nature, it is more often identified at a late stage and has an unfavorable prognosis. There are two main categories of lung cancer, non-small cell lung cancer (NSCLC) and small-cell lung cancer (SCLC). NSCLC corresponds around 80-85% of cases, while SCLC, a more aggressive tumor, accounts for approximately 15% [1]. Survival rates has been increased due to systematic therapy, with the combination of programmed cell death protein 1 (PD-1)/programmed death-ligand 1 (PD-L1) antibodies with platinum-doublet chemotherapy as the current standard of care for first-line treatment of advanced non-small-cell lung cancer (NSCLC) without a known targetable mutation regardless of PD-L1 score. Driver mutations like EGFR, KRAS, BRAF, and ALK were also detected, resulting in the development of targeted drugs. However, these medicines only provide advantages to certain groups of patients with specific molecular changes, and resistance mechanisms often restrict their effectiveness. This emphasizes the necessity for a more profound comprehension of the development of lung cancer and the discovery of novel targets for treatment [2].

2. Prospective Targeting of VEGF in Lung Cancer

Tumor angiogenesis, which involves the factors and signaling pathways, has emerged as a promising focus for therapeutic interventions in different malignancies, including lung cancer. VEGF and its receptors (VEGFRs) stimulate endothelial cell growth, movement, and infiltration via

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angiogenesis. VEGF promotes vascular permeability, aids in creating a temporary structure for the movement of endothelial cells and boosts the attraction of vascular precursor cells from the bone marrow. Recent research indicates that VEGF targets explicitly tumor cells, hence promoting the growth and spread of cancer. Both non-small cell lung cancer (NSCLC) and small cell lung cancer (SCLC) patients have been found to have increased expression of vascular endothelial growth factor (VEGF) in the bloodstream. Despite multiple anti-VEGF medications that are now in clinical development or have already been approved, their impact on lung cancer outcomes has been limited, with only minor improvements observed compared to the encouraging preclinical results [2,3].

1. Overview of VEGF/VEGFR Axis

The Vascular Endothelial Growth Factor (VEGF) family, consisting of VEGFA, VEGFB, VEGFC, VEGFD, and placenta growth factor (PlGF), plays an integral part in the mechanism of angiogenesis. The functions of the members of the VEGF family are processed by the process of connecting with their particular receptors. VEGF receptors are classified into two types: tyrosine kinase receptors (VEGF receptors, VEGFR), consisting of VEGFR-1, VEGFR-2, and VEGFR-3, and neuropilin receptors (NRPs), which include NRP-1 and NRP-2 [4,5]. NRPs function as co-receptors for VEGF, and the connection between VEGF and NRPs enhances the stability of the receptor complex [4]. The members of the VEGF family exhibit a specific affinity for VEGFR. VEGF-A is the primary constituent of the VEGF family that promotes the formation of new blood vessels. It is present in all vascular tissues, macrophages, tumor cells, and other types of cells [6,7]. Furthermore, it can attach to both VEGFR-1 and VEGFR-2. Still, it mainly attaches to the latter to form a pair, self-phosphorylate, and activate, thereby playing a vital role in subsequent signaling processes. This leads to the growth and movement of endothelial cells and carries out duties related to forming new blood vessels. [8,9]. VEGF-B mainly attaches to VEGFR-1 and NRP-1 and significantly impacts tumor angiogenesis and the enhancement of ischemic damage situations [10,11]. VEGF-C and VEGF-D mostly attach to VEGFR-3 and contribute to the process of lymphangiogenesis [11,12]. VEGF-D is linked to the spread of tumors to nearby lymph nodes [13,14]. Additionally, PIGF primarily attaches to VEGFR-1 and controls the development and maturity of blood arteries by preventing the multiplication of endothelial and parietal cells [15].

VEGFR-1 is expressed in numerous cell types other than endothelial cells and has an important function in regulating leukocyte migration. VEGFRs consist of seven immunoglobulin (Ig) homology regions which contain the area where the ligand binds. Additionally, they have an intracellular domain that exhibits tyrosine kinase activity, which is responsible for transmitting signals within the cell. VEGF contact induces the activation of phospholipase Cγ1, the MAPK pathway via Ras/Raf1 activation, and the PI3K/Akt pathway. Phospholipase Cγ1 holds a critical role in controlling the concentration of Ca+2 ions within cells and the production of endothelial nitric oxide synthase. The collective impact of these series of events is crucial for preserving the fundamental stability of blood vessels and facilitating processes such as the formation of new blood vessels, cell division, and migration of cells. The VEGF/VEGFR signaling system is frequently altered in several cancer types, which results in the formation of malformed blood vessels and the spreading of metastatic cancer cells [16,17].

2. Role of VEGF in Angiogenesis and Cancer

Several proteins and genes that play a crucial role in controlling the cell cycle, angiogenesis, and apoptosis have been identified as markers that greatly influence the response to treatment and clinical prognosis in patients with non-small cell lung cancer (NSCLC) [18,19]. Angiogenesis is a vital factor for the proliferation and metastasis of tumors and has been found to be a separate prognostic factor [20]. VEGF is a crucial and significant activator of tumor angiogenesis. Among the conflicting influences of proangiogenic and antiangiogenic factors, a signal appears that stimulates the generation of Vascular Endothelial Growth Factor (VEGF). This specific signaling system enables VEGF to perform many effects in the process of neoangiogenesis. VEGF expression mostly appears in endothelial cells, but it has also been detected at high levels in several types of tumors, including

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lung tumor cells. Hypoxia produces increased levels of VEGF in tumor tissue and stabilizes and enhances the expression of the transcription factor HIF-1α (Hypoxia-inducible factor-1α). HIF-1α, in turn, promotes the transcription of VEGF, which is then released, diffuses through the tissue, and binds to specific receptors on the surface of endothelial cells. Several studies indicate a correlation between the genetic variation of VEGF and the sensitivity, prognosis, and therapeutic responsiveness of individuals with NSCLC [21].

3. Role of VEGF in the Procedures of TME Cell Components in NSCLC

According to studies VEGF plays a role in malignancies by inducing the growth of new blood vessels (angiogenesis), but also by affecting tumor cells [22]. VEGF may stimulate the formation and spread of tumors by binding to receptors found on tumor cells through autocrine and paracrine processes [23]. NRPs, together with tyrosine kinases, have the ability to control the activity and motion of growth factor receptors and integrins. This makes them essential in aiding the effects of VEGF on malignant cells [23]. Malignant cells evade the immune response by inhibiting the function of T cells, such as by elevating the levels of T cell checkpoints [24,25]. VEGF-A promotes the production of PD-1 and other suppressive checkpoints, like CTLA-4, on the surface of T cells. Moreover, it impedes the operation of CD8+ T cells, resulting in a persistent malfunction that ultimately hinders the effector role of T cells [26,27]. Recent research indicates that tumor hypoxia, angiogenesis, and immunosuppression could mutually disrupt each other, fostering tumor progression and reducing the efficacy of cancer therapy [28]. VEGF not only directly modulates T cell activity but also potentially suppresses T cell function by regulating the levels of Fas ligand (FasL). VEGF-A amplifies the presence of FasL in the tumor microenvironment (TME) [29,30]. FasL is present in the outer layer of T cells and in cancer endothelial cells, while being absent in a healthy vascular system. The presence of FasL in endothelial cells in human carcinomas leads to the reduction of CD8+ T lymphocytes [31,32]. Regulatory T cells, often called Treg cells, are an essential group of CD4+ T cells. Various preclinical and clinical studies have shown that Treg cells are a predominant kind of immunosuppressive cells observed in malignancies [32]. They inhibit the process of immune surveillance to counteract cancer in individuals with favorable medical conditions. They impede the ability of patients with tumors to develop anti-tumor solid immunity, which leads to the formation and advancement of different types of malignant tumors, such as NSCLC [33]. The expression of VEGF-A in cancer patients was found to relate strongly with the levels of intratumoral Tregs [34]. VEGF-A can promote the development of regulatory T cells (Tregs) by increasing the population of immature dendritic cells (DCs) [35]. In addition, VEGF-A can directly control the recruitment of Treg cells in the tumor microenvironment (TME) by binding to VEGFR2. This interaction boosts the proliferation of Treg cells and enhances their immunosuppressive activity [32,35,36]. Tumor-associated macrophages (TAMs) are versatile cells that can adopt various polarization states. They play a crucial role in the initiation and advancement of cancer [37]. Tumor-associated macrophages (TAMs) are found at every stage of tumor formation, making them the most prevalent immune cells in the tumor microenvironment (TEM) [38]. There are two distinct phenotypes of TAMs, namely M1 and M2. The M1 phenotype has tumor-suppressing actions, while the M2 phenotype facilitates tumor advancement [39]. Tumor-associated macrophages (TAMs) produce cytokines, chemokines and growth factors that induce immunosuppression, and activate the suppressive immunological checkpoint proteins in T cells [40]. Hwang et al. demonstrated that M2 tumor-associated macrophages (TAMs) significantly increased VEGF-A and VEGF-C expression levels in non-small cell lung cancer (NSCLC) cells. On the other hand, M1 TAMs only increased the expression levels of VEGF-A in NSCLC cells. This indicates that TAMs play a significant role in the development of blood vessels and lymphatic vessel formation, promoting the advancement of NSCLC [41].

A type of cell called dendritic cells has the highest potential to present antigens compared to other cells. They can produce cytokines and facilitate the development of effector T and NK cells [42,43]. Dendritic cells (DC) can be separated from the first phase of hematopoietic progenitor cell (HPC) and VEGF-A may contribute to this mechanism by binding to HPC CD34+ cells through VEGFR-1 and thus suppressing the activity of nuclear factor-κB (NF-κB), that activates transcription

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factors in these cells. As a result, the differentiation and maturation of DC are inhibited [44,45]. VEGF can potentially hinder the function of dendritic cells by increasing the expression of PD-1. Blocking the development of dendritic cells decreases the infiltration of T cells into tumors and has an immunosuppressive impact. Recent data reveal that VEGF might impair mature DCs' migratory ability and immunological activity through the VEGFR-2-mediated RhoA-cofilin1 pathway [46].

Elevated levels of immature DCs in cancer patients are correlated with heightened levels of VEGF, which play a role in facilitating the malfunction of DCs [44]. In addition, the findings of a clinical trial examining the connection between DC infiltration and VEGF expression in NSCLC (132 primary NSCLC patients who underwent surgery) revealed that the average number of infiltrating DCs in the group with high VEGF expression was lower than that in the group with low expression [47]. This suggests that VEGF might control the infiltration of DCs into NSCLC tumors.

VEGF-A is a factor that can enhance the proliferation of myeloid-derived suppressor cells (MDSCs). The MDSC population comprises diverse and varied immature myeloid cells, which serve as progenitor cells for macrophages, dendritic cells (DC), or granulocytes. MDSCs are defined by their origin in the bone marrow, immature state, and ability to suppress the immune response [48]. These factors can enhance the survival of tumor cells, stimulate the growth of new blood vessels (angiogenesis), facilitate the invasion of tumor cells, and accelerate the spread of cancer to other parts of the body (metastases). In addition, MDSCs can promote immunological tolerance and decrease the activity of effector T cells and NK cells, hence stimulating immune responses [48,49]. Furthermore, MDSCs can hinder the proliferation of T cells specific to tumors and facilitate the formation of regulatory T cells (Tregs), which are crucial in suppressing the immune response and evading the immune system. MDSCs are also implicated in the process of Treg cell development. An elevation of myeloid-derived suppressor cells (MDSC) in the bloodstream of individuals with cancer leads to a reduction in the number of fully developed dendritic cells (DCs) [50]. Many studies have indicated that MDSCs play a significant role in modulating a range of tumor-related immunosuppressive activities and tumor immune escape, including NSCLC [32].

Natural Killer (NK) cells are a specific subset of cytotoxic innate lymphoid cells within the innate immune system. They possess a distinct ability to eliminate tumor cells effectively. VEGF can impede the development of NK cells by obstructing the maturation of DCs [44,51]. In addition, VEGF can enhance the quantity of MDSCs and suppress the activity of NK cells, resulting in immunological escape [32].

Research has demonstrated that natural killer (NK) cells are capable of releasing vascular endothelial growth factor-A (VEGF-A) when exposed to low oxygen circumstances, which is a distinguishing feature of the tumor microenvironment (TME) [52]. In settings of low oxygen levels (hypoxia), the release of VEGF is temporary. This is because when NK cells return to the bloodstream, this occurrence can be reversed. Hypoxia plays an essential part during cancer treatment by causing an imbalance in the signaling between pro- and antiangiogenic variables and physical compression. This results in abnormal blood vessels and substantially decreased blood flow in tumors. The increasing heterogeneity in blood flow, which worsens over time, differs depending on the stage and location of tumor growth. This leads to cancer cells evading the immune system, enhancing their ability to move in and spread to other body parts, and exerting selected survival pressures. By relieving hypoxia, it is possible to alter the characteristics of macrophages, making them more supportive of tumor growth and suppressing the immune response, improving the efficiency of cancer treatment [53].

4. VEGF and Immune Checkpoints in NSCLC

VEGF not only stimulates tumor growth by facilitating the formation of new blood vessels but also affects different immune cells in the tumor microenvironment, suppressing the immune response. Thus, while treating NSCLC, choosing VEGF-VEGFR-targeted medications can impede tumor growth.

VEGF is upregulated in non-small cell lung cancer (NSCLC), with higher expression levels observed in the tumor tissue compared to the adjacent normal lung tissue [54]. The elevated Vascular

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Endothelial Growth Factor (VEGF) expression is associated with tumor recurrence, reduced survival rate, metastasis, and mortality [54,55]. VEGF is essential for tumor progression and immunosuppression. Hence, specific medications that hinder the VEGF pathway, such as monoclonal antibodies against VEGF and tyrosine kinase inhibitors (TKIs), are employed to treat NSCLC.

An immunological checkpoint is a protein that can induce immunosuppression, hence modulating the immune response. Monoclonal antibodies that inhibit the binding of cytotoxic T lymphocyte-associated antigen-4 (CTLA-4) and programmed cell death protein (PD-1) or its ligand PD-L1 have received clinical approval. PD-1 and PD-L1 are predominantly present in immune cells, specifically NK cells, DC, CD4+, and CD8+ T cells [56,57]. PD-1 binds with its ligand PD-L1 to suppress the activation and reproduction of T cells, resulting in the evasion of the immune system. Strong connections were found between the levels of PD-L1 expression and the levels of angiogenic factors, including VEGFA and HIF-1α [58]. CTLA-4 is an external protein found on the surface of cells that can regulate immunological suppression. Its primary function is to activate T cell receptors, which play a crucial role in the immune response [59].

The first-line therapy, now approved as immune checkpoint inhibitors, can be classified into three primary categories: Anti-PD-1, Anti-PD-L1, and Anti-CTLA-4. VEGF-A suppresses immune activation and promotes immunosuppression by influencing different immune cells inside the tumor microenvironment (TME). Thus, the suppression of immunological escape can be achieved by decreasing the impact of VEGF, followed by the combination of immune checkpoint inhibitors for the treatment of NSCLC. Initially, anti-angiogenic medications have the ability to restore normalcy to the blood vessels within tumors, resulting in an increase of tumor immune cells, specifically tumor-infiltrating lymphocytes, in cases of non-small cell lung cancer (NSCLC). Immune checkpoint inhibitors alleviate the suppression of PD-1 and PD-L1 on T cells, and the combined impact of these two factors result in improved treatment outcomes for solid malignancies [60].

A restricted number of studies have investigated the importance of plasma vascular endothelial growth factor (VEGF) levels in predicting the therapeutic outcomes of anti-angiogenesis together with anti-PD-L1 approaches. Tozuka et al. indicated that patients who had a reduction in their post-treatment plasma VEGF-A concentrations in comparison to their pre-treatment levels exhibited a markedly extended progression-free survival (PFS) in contrast to those whose post-treatment plasma VEGF-A concentrations either increased or remained stable. Within the responder cohort, a larger fraction of patients demonstrated a persistent decline in their plasma VEGF-A levels across the treatment period [61].

4. Discussion

Vascular endothelial growth factor (VEGF) facilitates tumor proliferation by enabling the formation of new blood vessels and controls various immune cells inside the tumor microenvironment, therefore inhibiting immune response. Upregulation of VEGF in non-small cell lung cancer (NSCLC) is linked to increased expression levels, which in turn are connected with tumor recurrence, decreased survival rate, metastasis, and death. Monoclonal antibodies targeting VEGF and tyrosine kinase inhibitors (TKIs) are drug therapies used to treat non-small cell lung cancer (NSCLC) by inhibiting the VEGF transport route. Levels of plasma vascular endothelial growth factor (VEGF) have been shown to indicate the effectiveness of anti-angiogenesis and anti-PD-L1 therapies. Significantly prolonged progression-free survival (PFS) was seen in patients with decreased post-treatment plasma VEGF-A levels. Further studies are needed for the evaluation of different VEGF factors and their potential roles as prognostic and predictive factors for NSCLC.

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