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Vascular Endothelial Growth Factor Gene Variants and Their Association with Diabetic Retinopathy Risk in Type 2 Diabetes Patients

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Abstract: Diabetic retinopathy (DR) is a major microvascular complication of type 2 diabetes mellitus (T2DM) and a leading cause of vision loss globally. Vascular Endothelial Growth Factor (VEGF) plays a critical role in angiogenesis and increased vascular permeability, key processes in the pathogenesis of DR. Recent studies have explored the association of VEGF gene polymorphisms with susceptibility to DR in T2DM patients. This review aims to summarize and analyze the current evidence regarding the association of VEGF gene polymorphisms, particularly single nucleotide polymorphisms (SNPs), with the development and progression of DR in individuals with T2DM. A comprehensive literature search was conducted to identify relevant studies investigating VEGF gene variants and their correlation with DR. Key polymorphisms, such as VEGF -2578C/A, -460T/C, +405G/C, and others, were evaluated for their potential role in genetic predisposition to DR. The findings highlight that certain VEGF gene polymorphisms are significantly associated with increased risk of DR in various populations. Differences in allelic frequency and genotype distributions among ethnic groups are also evident, indicating a complex interplay of genetic and environmental factors. **Conclusion:** VEGF gene polymorphisms may serve as potential genetic markers for predicting DR risk in T2DM patients. Understanding these associations could facilitate early detection, personalized risk assessment, and the development of targeted therapies. However, further large-scale and multi-ethnic studies are warranted to validate these findings and elucidate underlying molecular mechanisms.

Keywords: *VEGF, gene polymorphism, diabetic retinopathy, type 2 diabetes mellitus, genetic susceptibility*

Introduction.

Diabetic retinopathy (DR) is a progressive microvascular complication of diabetes mellitus (DM), characterized by damage to the retinal vasculature, leading to vision impairment and blindness in severe cases. It is a

significant cause of preventable blindness globally, particularly in individuals with poorly controlled diabetes over prolonged periods [1]. DR is classified into non-proliferative diabetic retinopathy (NPDR) and proliferative diabetic retinopathy (PDR), with NPDR being an early stage marked by microaneurysms, retinal hemorrhages, and hard exudates, and PDR involving neovascularization and increased risk of retinal detachment [2].

The pathogenesis of DR is multifactorial, involving chronic hyperglycemia, oxidative stress, and inflammation. Prolonged hyperglycemia leads to the accumulation of advanced glycation end-products (AGEs), which play a pivotal role in retinal endothelial damage. AGEs bind to specific receptors (RAGE), activating pathways that contribute to oxidative stress and vascular leakage [3]. The oxidative damage further exacerbates retinal injury, promoting endothelial cell apoptosis and disruption of the blood-retinal barrier (BRB) [4].

Microbial infections have also been implicated in exacerbating DR through inflammatory processes. Certain pathogens, including Gram-negative bacteria, may produce endotoxins that amplify systemic inflammation, contributing to retinal vascular dysfunction. Additionally, the gut microbiome's role in diabetes-related inflammation has been increasingly recognized, suggesting a potential interplay between microbial dysbiosis and DR [5].

Inflammation is central to the progression of DR. Inflammatory cytokines such as interleukin-6 (IL-6), tumor necrosis factor-alpha (TNF-α), and vascular endothelial growth factor (VEGF) are elevated in the vitreous humor of patients with DR. VEGF, in particular, is a critical mediator of neovascularization in PDR and a target for therapeutic interventions, such as anti-VEGF agents [6]. Chronic inflammation also leads to leukostasis, where leukocytes adhere to the retinal vasculature, further compromising capillary perfusion and increasing the risk of ischemia [7].

Microvascular changes in DR include capillary basement membrane thickening, pericyte loss, and capillary occlusion. Pericytes are critical for maintaining capillary stability; their loss leads to capillary leakage and microaneurysm formation, hallmark features of NPDR. Capillary occlusion contributes to retinal ischemia, which in turn triggers neovascularization in PDR [8].

In addition to vascular damage, neuronal degeneration in the retina plays a significant role in DR. Retinal ganglion cells, photoreceptors, and glial cells are affected early in the disease process. The interplay between vascular and neuronal damage, termed neurovascular coupling, highlights the complexity of DR pathophysiology [9].

The role of medical microbiology in understanding DR extends to identifying infections that exacerbate hyperglycemia and inflammation. For instance, periodontal infections, caused by anaerobic bacteria such as *Porphyromonas gingivalis*, have been linked to increased systemic inflammation and worsening diabetic control, thereby indirectly influencing DR progression [10].

Emerging evidence suggests that specific viral infections, such as cytomegalovirus (CMV), may directly affect retinal cells, particularly in immunocompromised individuals, including those with poorly controlled diabetes. CMV retinitis remains a concern in these populations, emphasizing the need for vigilant infection control [11].

Management of DR involves a combination of glycemic control, blood pressure management, and specific retinal therapies. Tight glycemic control is paramount in reducing the risk of DR onset and progression, as demonstrated by landmark studies such as the Diabetes Control and Complications Trial (DCCT) [12].

Anti-VEGF therapy has revolutionized the treatment of PDR and diabetic macular edema (DME), reducing vision loss in numerous patients. Agents like ranibizumab and aflibercept inhibit VEGF activity, decreasing neovascularization and vascular leakage [13]. Corticosteroid implants, such as dexamethasone, are also used to manage DME by reducing inflammation [14].

Laser photocoagulation remains a cornerstone for DR treatment, particularly for PDR. Panretinal photocoagulation (PRP) targets ischemic retinal areas, reducing VEGF production and preventing neovascularization [15]. While effective, PRP may cause side effects, including visual field loss and reduced night vision [16].

Vitrectomy, a surgical procedure to remove vitreous hemorrhage or tractional retinal detachment, is indicated in advanced DR cases. Innovations in surgical techniques have improved outcomes, enabling the restoration of vision in patients with severe disease [17].

Preventive measures, including regular retinal screening and early intervention, are crucial in mitigating DR's impact. Annual dilated fundus examinations are recommended for individuals with diabetes, with more frequent evaluations in high-risk patients, such as those with poorly controlled diabetes or pregnancy [18].

Future research in DR includes exploring the microbiome's role, novel biomarkers for early detection, and advanced imaging techniques. Optical coherence tomography angiography (OCTA) has shown promise in detecting early microvascular changes, enabling earlier intervention [19].

DR is a complex disease influenced by metabolic, inflammatory, and microbiological factors. A multidisciplinary approach encompassing glycemic control, advanced retinal therapies, and infection management is essential to reducing its global burden [20].

While multiple studies have investigated the association between VEGF gene polymorphisms and diabetic retinopathy (DR) in type 2 diabetes mellitus (T2DM), the majority focus on specific populations, leading to inconsistent findings due to ethnic and genetic variability. There is a lack of large-scale, multi-ethnic studies that can comprehensively validate these associations across diverse populations. Additionally, limited research explores the interaction of VEGF polymorphisms with environmental factors, glycemic control, and other genetic markers, which may collectively influence the development and progression of DR. Addressing these gaps can provide a more holistic understanding of VEGF's role in DR and improve predictive accuracy for at-risk patients.

Vascular endothelial growth factor

Vascular endothelial growth factor (VEGF) is an important growth factor family member. VEGF is a potent proangiogenic factor. VEGF is used as a prognostic marker for many diseases that their pathogenesis includes angiogenesis such as diabetic angiopathies remarkably diabetic retinopathy. [21].

Vascular endothelial growth factor increases vascular permeability regulates vascular endothelial proliferation and promote angiogenesis in RD which cause retinal detachment with visual impairment. Several studies predict that VEGF expression is increased in patients with diabetic retinopathy. The genetic variations that are involved in the gene promotor area can be associated with increased VEGF expression that is recorded in many vascular diseases. [21].

VEGF and its isoforms:

VEGFA (or VEGF hereafter) is the prototypic member of a family includes placental growth factor (PLGF, also known as PGF), VEGFB, VEGFC and VEGFD, Native VEGF is consistent with a homodimer, subsequent cDNA cloning revealed that the main VEGF species is a 165-amino acid glycoprotein with sequence homology to the A and B chains of PDGF VEGFA is first expressed in the anterior portion of mouse embryos, where it directs the migration of cells positive for VEGFR1 and VEGFR2 [21]. Indeed, the majority of normal and abnormal cells can produce VEGF. In the adult, VEGF induces proliferation, sprouting, migration and tube formation of endothelial cells(ECs), and is a potent survival factor for ECs during physiological and tumour angiogenesis Additionally, VEGF has been reported to have several neuronal and neurodevelopmental roles. VEGF can exist as one of several isoforms generated through alternative exon splicing of a single gene, comprising 8 exons27, these include VEGF121, VEGF165, VEGF189, and VEGF206 and some less common variants such as VEGF145 [21].

Vascular Endothelial Growth Factor (VEGF) is a pivotal protein that plays an integral role in angiogenesis, the formation of new blood vessels from pre-existing ones. It belongs to a family of growth factors that includes VEGF-A, VEGF-B, VEGF-C, VEGF-D, and placental growth factor (PlGF). Among these, VEGF-A is the most extensively studied due to its critical involvement in vascular endothelial cell proliferation, migration, and permeability. VEGF's role extends beyond vascular growth to include tissue repair and tumor progression, underscoring its dual physiological and pathological significance. Dysregulation in VEGF signaling can lead to an imbalance in angiogenesis, contributing to diseases such as diabetic retinopathy (DR), cancer, and

cardiovascular disorders. Its complex regulation at genetic and molecular levels makes it a critical therapeutic target [21].

The bioavailability and angiogenic activity of the VEGF

The bioavailability and angiogenic activity of VEGF are intricately regulated by its interaction with cell surface receptors and extracellular matrix components. VEGF primarily binds to tyrosine kinase receptors, VEGFR-1 and VEGFR-2, with VEGFR-2 being the principal mediator of angiogenic signaling. VEGFR-1, while less active in downstream signaling, acts as a decoy receptor to regulate VEGF levels. The availability of VEGF to these receptors is modulated by proteolytic cleavage and the presence of co-receptors like neuropilins, which enhance VEGF's binding affinity. Furthermore, the sequestration of VEGF in the extracellular matrix ensures a localized and sustained angiogenic response. This regulatory mechanism is particularly relevant in diseases like DR, where uncontrolled VEGF release leads to pathological angiogenesis [22].

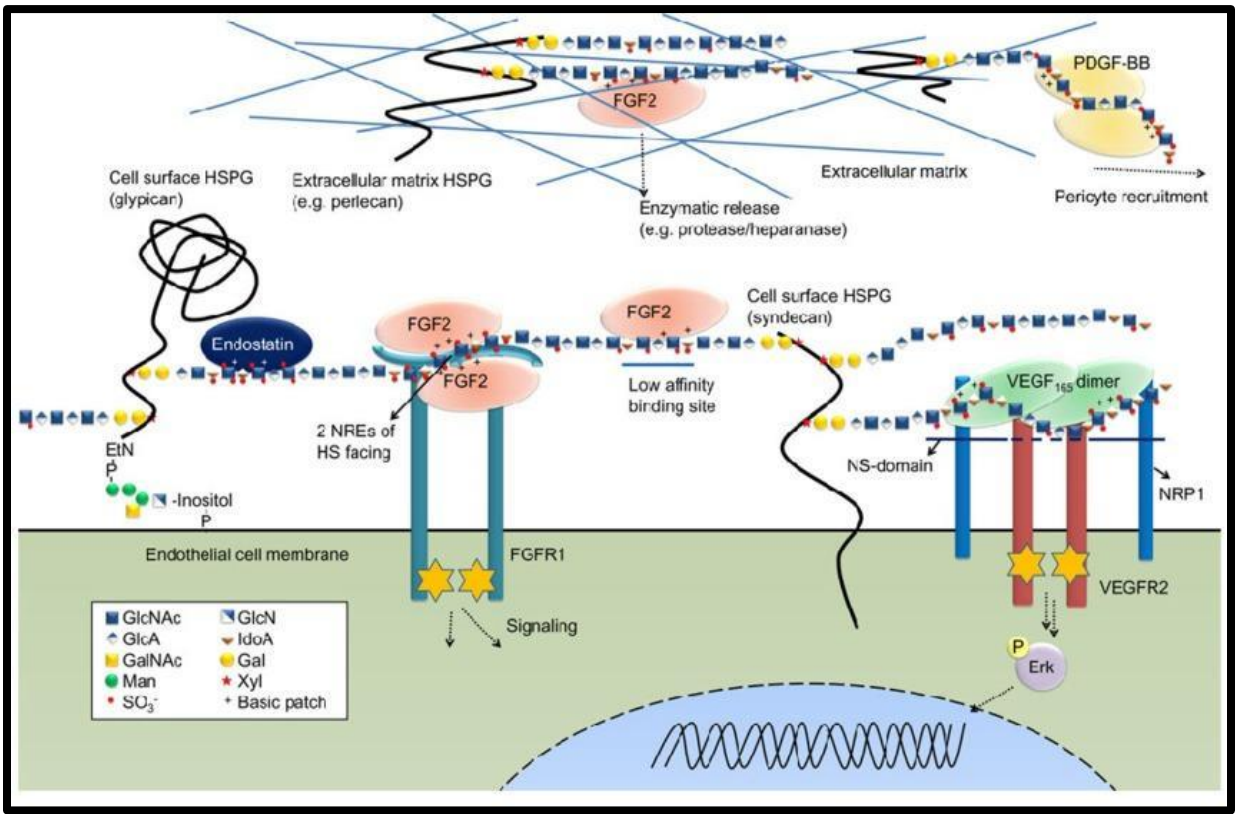

Figure 1: Mechanism of action of VEGF in angiogenesis [22].

VEGF's angiogenic activity is a tightly controlled process crucial for maintaining vascular homeostasis. In response to hypoxia or other stimuli, VEGF expression is upregulated via the hypoxia-inducible factor-1 (HIF-1) pathway. This upregulation triggers endothelial cell proliferation and migration, resulting in the formation of new capillaries. In diabetic retinopathy, chronic hyperglycemia induces oxidative stress and hypoxia in

retinal tissues, activating the HIF-1α/VEGF signaling pathway. Dysregulation of this pathway results in increased VEGF expression, promoting excessive neovascularization and vascular leakage in the retina [23].
VEGF polymorphisms are genetic variations that can influence its expression and activity. Single nucleotide polymorphisms (SNPs) within the VEGF gene have been associated with altered susceptibility to various diseases, including cancer, diabetic retinopathy, and cardiovascular disorders. These polymorphisms can affect VEGF production, receptor binding, and downstream signaling, thereby modulating disease progression and therapeutic responses. For instance, VEGF gene polymorphisms have been strongly linked to the severity of diabetic retinopathy in type 2 diabetes patients, as VEGF is a key mediator of retinal neovascularization [24].

One of the most studied VEGF polymorphisms is located in the promoter region of the VEGF gene. This polymorphism affects the transcriptional activity of the gene, leading to variable VEGF expression levels. For instance, the −257C>A SNP has been linked to altered VEGF secretion, influencing angiogenesis and disease susceptibility. In type 2 diabetes patients, these variations in VEGF gene expression play a crucial role in the development and progression of diabetic retinopathy. Elevated VEGF levels have been detected in the vitreous humor and serum of patients with proliferative diabetic retinopathy (PDR), further supporting its involvement [25].

Association of the VEGF gene polymorphism with diabetic retinopathy in type 2 diabetes patients

The association of VEGF gene polymorphisms with diabetic retinopathy (DR) has garnered significant research interest. DR is a microvascular complication of diabetes characterized by retinal neovascularization, increased vascular permeability, and hemorrhagic lesions. Polymorphisms such as +405G>C, −460T>C, and −257C>A have been implicated in modulating VEGF expression, thereby influencing the severity and progression of DR in type 2 diabetes patients. Studies have demonstrated that these polymorphisms can alter VEGF transcription, resulting in differential VEGF protein levels, which correlate with disease progression [26].
The +405G>C polymorphism is one of the most frequently studied SNPs in VEGF and its association with DR. This SNP is located in the 5′-untranslated region of the VEGF gene and has been shown to increase VEGF production in diabetic patients. Elevated VEGF levels exacerbate retinal vascular permeability, leading to macular edema, hemorrhage, and neovascularization. In a study by Awata et al., the +405G>C polymorphism was associated with a significantly higher risk of proliferative DR in type 2 diabetes patients, highlighting the clinical relevance of this SNP [27].
Similarly, the −460T>C polymorphism has been linked to increased VEGF expression under hypoxic conditions. This SNP, located in the VEGF gene promoter region, enhances VEGF transcriptional activity, resulting in elevated VEGF levels. In type 2 diabetes patients, this leads to an increased risk of proliferative diabetic retinopathy (PDR). Elevated VEGF promotes pathological angiogenesis and vascular leakage, both hallmark features of PDR. Studies have confirmed a strong association between the −460T>C polymorphism and DR progression [28].
The −257C>A polymorphism further underscores the genetic basis of VEGF regulation in diabetic retinopathy. This SNP has been shown to reduce promoter activity, leading to lower VEGF production. Paradoxically, certain populations exhibit an increased frequency of the −257C>A SNP in severe DR cases, suggesting that the SNP's impact on VEGF levels may be context-dependent. This highlights the complex interplay of genetic and environmental factors in DR pathogenesis [29].
Elevated VEGF levels in the diabetic retina contribute not only to angiogenesis but also to inflammation. VEGF interacts with inflammatory cytokines such as interleukin-6 (IL-6) and tumor necrosis factor-α (TNF-α), amplifying the inflammatory cascade. This creates a pro-inflammatory microenvironment that exacerbates vascular damage and retinal dysfunction. Genetic polymorphisms that upregulate VEGF expression can therefore amplify both angiogenic and inflammatory pathways, accelerating DR progression. Anti-VEGF

therapies such as ranibizumab and bevacizumab have been highly effective in mitigating these effects and preserving vision [30].

Pharmacogenetic studies have highlighted the impact of VEGF polymorphisms on therapeutic responses. Patients carrying certain VEGF SNPs may exhibit differential responses to anti-VEGF agents, influencing treatment efficacy. For instance, the +405G>C polymorphism has been associated with variable responses to bevacizumab in DR patients, emphasizing the need for personalized treatment approaches based on genetic profiling. Identifying high-risk polymorphisms can improve patient stratification and optimize therapeutic outcomes [31].

Beyond DR, VEGF polymorphisms have been implicated in other vascular and inflammatory diseases. For instance, the +936C>T polymorphism has been associated with altered VEGF levels in conditions like age-related macular degeneration (AMD) and rheumatoid arthritis. These findings illustrate the broader significance of VEGF polymorphisms in human health and disease. Understanding the genetic regulation of VEGF provides valuable insights for therapeutic targeting across multiple diseases [32].

The bioavailability of VEGF is also influenced by post-transcriptional and post-translational modifications. MicroRNAs, such as miR-126, regulate VEGF mRNA stability and translation, while glycosylation affects its receptor binding and angiogenic activity. These layers of regulation ensure precise control of VEGF's functions in different physiological and pathological contexts. Dysregulation of these mechanisms in diabetes contributes to the pathogenesis of DR [33].

Advances in molecular biology have shed light on the role of alternative splicing in generating VEGF isoforms with distinct biological activities. Isoforms like VEGF121 and VEGF165 differ in their heparin-binding capacity and receptor interactions, influencing their angiogenic potential and therapeutic applications. VEGF165, the most abundant isoform, has been shown to play a prominent role in DR progression due to its potent angiogenic activity. Such insights are pivotal for designing isoform-specific interventions in DR treatment [34].

Therapeutic modulation of VEGF activity is a cornerstone of treatment strategies for diseases like cancer and ocular disorders. Anti-VEGF agents, including monoclonal antibodies and tyrosine kinase inhibitors, have revolutionized the management of these conditions. However, challenges like resistance, cost, and adverse effects necessitate the exploration of combination therapies and novel delivery systems. Personalized approaches based on VEGF polymorphism profiling hold promise for improving therapeutic efficacy and minimizing side effects [35].

In conclusion, VEGF is a multifunctional protein with critical roles in angiogenesis and vascular homeostasis. Its bioavailability and activity are intricately regulated by genetic, molecular, and environmental factors. VEGF gene polymorphisms, such as +405G>C, −460T>C, and −257C>A, are significantly associated with diabetic retinopathy in type 2 diabetes patients, influencing disease progression and therapeutic responses. Understanding these mechanisms is essential for developing targeted therapies and advancing precision medicine in VEGF-related diseases.

References

1. Cheung N, Mitchell P, Wong TY. Diabetic retinopathy. The Lancet. 2010;376(9735):124–36.

2. Wilkinson CP, et al. Proposed international clinical diabetic retinopathy and diabetic macular edema disease severity scales. Ophthalmology. 2003;110(9):1677–82.

3. Brownlee M. The pathobiology of diabetic complications: a unifying mechanism. Diabetes. 2005;54(6):1615–25.

4. Behl T, et al. The role of oxidative stress in the pathology of diabetic retinopathy and the preventive approaches. Biomedicine & Pharmacotherapy. 2017;93:1156–67.

5. Cani PD, et al. Gut microbiota and diabetes: From pathogenesis to therapeutic perspective. Diabetologia. 2011;54(2):303–10.

6. Simo R, Hernandez C. Intravitreous anti-VEGF for diabetic retinopathy: Hopes and fears for a new therapeutic strategy. Diabetologia. 2008;51(9):1574–85.

7. Joussen AM, et al. Pathophysiology of diabetic macular edema. Diabetes. 2007;56(5):1461–72.

8. Gardner TW, et al. The pathophysiology of diabetic retinopathy: putting the pieces together. Diabetes. 2011;60(11):2853–62.

9. Antonetti DA, et al. Diabetic retinopathy: seeing beyond glucose-induced microvascular disease. Diabetes. 2006;55(9):2401–11.

10. Taylor GW, et al. Periodontal disease and diabetes. J Periodontol. 1996;67(10 Suppl):1051—60.

11. Holland GN. Retinal diseases associated with HIV infection. Trans Am Ophthalmol Soc. 1992;90:555–61.

12. The Diabetes Control and Complications Trial Research Group. The effect of intensive diabetes treatment on the development and progression of long-term complications in insulin-dependent diabetes mellitus. N Engl J Med. 1993;329(14):977–86.

13. Ip MS, et al. Anti-vascular endothelial growth factor therapy for diabetic macular edema: Evidence and recommendations. Ophthalmology. 2012;119(10):2179–88.

14. Campochiaro PA, et al. Sustained delivery fluocinolone acetonide vitreous inserts provide benefit for at least 3 years in patients with diabetic macular edema. Ophthalmology. 2012;119(10):2125–32.

15. Early Treatment Diabetic Retinopathy Study Research Group. Photocoagulation for diabetic macular edema. Arch Ophthalmol. 1985;103(12):1796–806.

16. Shalchi Z, et al. Complications of pan-retinal photocoagulation. Eye. 2016;30(6):843—0.

17. Wong TY, et al. Surgical management of diabetic retinopathy. Diabetes Care. 2004;27(10):2536–46.

18. American Diabetes Association. Standards of medical care in diabetes—2024. Diabetes Care. 2024;47(Suppl 1):S1–60.

19. Spaide RF, Fujimoto JG, Waheed NK. Optical coherence tomography angiography. Prog Retin Eye Res. 2018;64:1–55.

20. Yau JW, et al. Global prevalence and major risk factors of diabetic retinopathy. Diabetes Care. 2012;35(3):556–64.

21. Ferrara N. VEGF as a therapeutic target in cancer. Oncologist. 2004;9(Suppl 1):2-10.

22. Olsson AK, Dimberg A, Kreuger J, Claesson-Welsh L. VEGF receptor signalling\u2014in control of vascular function. Nat Rev Mol Cell Biol. 2006;7(5):359-371.

23. Shibuya M. Structure and function of VEGF/VEGF-receptor system involved in angiogenesis. Cell Struct Funct. 2001;26(1):25-35.

24. Watson CJ, Webb NJ, Bottomley MJ, Brenchley PE. Identification of polymorphisms within the vascular endothelial growth factor (VEGF) gene: correlation with variation in VEGF protein production. Cytokine. 2000;12(8):1232-1235.

25. Stevens A, Soden J, Brenchley PE, Ralph S, Ray DW. Haplotype analysis of the polymorphic human vascular endothelial growth factor gene promoter. Cancer Res. 2003;63(4):812-816.

26. Awata T, Inoue K, Kurihara S, et al. A common polymorphism in the 5\u2032-untranslated region of the VEGF gene is associated with diabetic retinopathy in type 2 diabetes. Diabetes. 2002;51(5):1635-1639.

27. Al-Kateb H, Mirea L, Xie X, et al. Multiple VEGF polymorphisms are associated with diabetic retinopathy in type 1 diabetes. Diabetes. 2007;56(8):2164-2170.

28. Ray D, Mishra M, Ralph S, et al. Association of the VEGF gene with proliferative diabetic retinopathy but not proteinuria in diabetes. Diabetes. 2004;53(3):861-864.

29. Wirostko B, Wong TY, Simo R. Vascular endothelial growth factor and diabetic complications. Prog Retin Eye Res. 2008;27(6):608-621.

30. Abhary S, Burdon KP, Gupta A, et al. Common sequence variation in the VEGFA gene predicts risk of diabetic retinopathy. Invest Ophthalmol Vis Sci. 2009;50(12):5552-5558.

31. Churchill AJ, Carter JG, Ramsden C, et al. VEGF polymorphisms are associated with increased risk of developing diabetic retinopathy. Diabetologia. 2008;51(8):1425-1433.

32. Long J, Wang Y, Wang W, Chang BH, Danesh FR. MicroRNA-29c mediates high glucose-induced endothelial dysfunction in human retinal endothelial cells. PLoS One. 2010;5(6):e11092.

33. Harper SJ, Bates DO. VEGF-A splicing: the key to anti-angiogenic therapeutics? Nat Rev Cancer. 2008;8(11):880-887.

34. Ferrara N, Hillan KJ, Gerber HP, Novotny W. Discovery and development of bevacizumab, an anti-VEGF antibody for treating cancer. Nat Rev Drug Discov. 2004;3(5):391-400.

35. Carmeliet P, Jain RK. Molecular mechanisms and clinical applications of angiogenesis. Nature. 2011;473(7347):298-307.