VEGF Paradoxically Reduces Cerebral Blood Flow in Alzheimer’s Disease Mice

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ABSTRACT: Vascular dysfunction plays a critical role in the development of Alzheimer’s disease. Cerebral blood flow reductions of 10% to 25% present early in disease pathogenesis. Vascular Endothelial Growth Factor-A (VEGF-A) drives angiogenesis, which typically addresses blood flow reductions and global hypoxia. However, recent evidence suggests aberrant VEGF-A signaling in Alzheimer’s disease may undermine its physiological angiogenic function. Instead of improving cerebral blood flow, VEGF-A contributes to brain capillary stalls and blood flow reductions, likely accelerating cognitive decline. In this commentary, we explore the evidence for pathological VEGF signaling in Alzheimer’s disease, and discuss its implications for disease therapy.

KEYWORDS: Alzheimer’s disease, cerebral blood flow, VEGF-A signaling, blood-brain barrier

Introduction

Vascular dysfunction plays a vital role in the pathogenesis of Alzheimer’s disease. Not only are hypertension, diabetes, and atherosclerotic disease primary risk factors for Alzheimer’s disease,1 patient-level meta-analysis of multiple genome-wide association studies have highlighted the involvement of cerebrovascular disease-related pathways in Alzheimer’s disease.2 Alzheimer’s disease-related vascular changes are characterized, in part, by impairment of autoregulation and reduced cerebral blood flow.3-5 Importantly, these changes precede cognitive decline, making room for potential therapeutic interventions.6

Cerebral blood flow reductions of 10% to 25% present early in disease pathogenesis.5 Mechanisms driving such drastic changes have largely remained unestablished. Recent studies employing in vivo microscopy have implicated cellular changes in mouse models of the disease, including pericyte constriction of capillaries,7 vascular obstructions secondary to hypercoagulability,8 and leukocyte capillary stalling.9,10 Specifically, neutrophil adhesion to the cortical microvasculature leads to 17% reduced cerebral blood flow and cognitive deficits in the APP/PS1 and 5xFAD mouse models.9 Detecting and addressing this phenomenon in humans may be critical in developing new treatment strategies.

Aberrant VEGF Signaling in Alzheimer’s Disease

The vascular endothelial growth factor (VEGF-A) is involved in a broad array of signaling pathways contributing to angiogenesis,11 neurogenesis,12 and neuroprotection.13 Physiologically, angiogenesis addresses local and global hypoxia. States of global cerebral hypoxia, like Alzheimer’s disease, show evidence of increased VEGF-A levels13-15 and capillary density.16 However, vascular integrity is impaired, including the formation of vascular loops, glomeruloid structures, aberrant branching patterns, and irregular basement membranes, ultimately leading to insufficient oxygenation of brain tissue and neuronal dysfunction.17 In the end, though VEGF may induce neo-angiogenesis, it also contributes to vascular hyperpermeability and brain edema, which paradoxically contributes to diminished blood flow, reduced nutrient delivery, and entry of restricted molecules into the brain,18 likely accelerating Alzheimer’s progression.

We recently found that upregulated VEGF-A signaling contributes to cerebral blood flow reductions through capillary stalls in the APP/PS1 model of Alzheimer’s disease (Figure 1A–C). Specifically, expression of the VEGF-A-associated tight junction protein occludin was downregulated in occluded capillaries.19 The capillary stalling hypothesis suggests pathological VEGF-A/occludin-associated blood-brain barrier hyperpermeability activates local inflammatory markers in endothelial cells, recruiting leukocytes to the site of injury, increasing the incidence of stalled capillaries, and ultimately leading to cerebral blood flow reductions (Figure 1D). We targeted this pathway using an anti-VEGF-A antibody. Injection of the antibody immediately improved the integrity of the blood–brain barrier, leading to a reduction in stalled capillaries, and restoring cerebral blood flow. Longitudinally inhibiting VEGF-A through the vascular lumen specifically could also address the deleterious effects of pathological angiogenesis without compromising pathways of neuroprotection, such as neurogenesis, on the other side of the blood–brain barrier. Indeed, a recent perspective linked blood-brain barrier breakdown to cognitive impairment in Alzheimer’s disease patients.20 Overall, these publications indicate a critical role of a dysregulated blood-brain barrier in cognitive impairment and dementia.
Interestingly, recent evidence suggests mutations of VEGF-A protect against Alzheimer’s disease. Two epistatic interactions, each between VEGF-A related single nucleotide polymorphisms identified in large-scale GWAS studies (143 Alzheimer’s disease cases and 180 controls), were the strongest protective factors against Alzheimer’s disease in the absence of ε4 APOE allele, which remained the most significant genetic predisposition. This study suggests that VEGF-A signaling may play a more significant role than previously indicated in the pathogenesis of Alzheimer’s disease.

Models to Consider: Parkinson’s Disease and Diabetic Retinopathy

The deleterious effects of VEGF-associated blood-brain barrier hyperpermeability seem not to be restricted to Alzheimer’s disease. In post-mortem brain tissues of patients with Parkinson’s disease, elevated VEGF-A and nitric oxide levels were detected at sites of astrocytic alpha-synuclein deposition. Inhibition of VEGF-A blocked the deleterious effects of VEGF-A on the blood-brain barrier in a mouse model of Parkinson’s disease, especially among younger mice, suggesting more beneficial effects early in disease pathogenesis.

Indeed VEGF-A inhibition is the most successful treatment for multiple ocular conditions, including diabetic retinopathy. Our hypothesis mirrors the proposed pathogenesis of diabetic retinopathy. The VEGF-A/eNOS/occludin pathway seems to drive leukocyte induced capillary stalling at the blood-retina barrier of patients with diabetic retinopathy (Figure 2A and B). Inhibition of VEGF-A, by intravitreal injections, releases these pathognomonic obstructions, partially inhibiting the progression of diabetes-associated pathology. However, repetitive anti-VEGF-A injections are not without side effects.
For example, the repetitive treatment increases the risk for hypertension-induced brain hemorrhages and maybe even cognitive decline.

A study analyzed 175 patients undergoing anti-VEGF-A treatment for macular degeneration demonstrated those with 20 or more injections had a higher likelihood of mild cognitive impairment as calculated on an iPad-based brain health assessment. In our study mice were administered at most 6 anti-VEGF-A injections over the course of 2 weeks. Given the intraperitoneal mode of injection and size of the antibody used in our experiments, we largely saw effects on the vascular system rather than the brain parenchyma itself. Indeed, increased exposure to anti-VEGF-A injections could place patients at greater risk for impaired neuroprotection. Of note, those receiving 15 to 20 injections did not demonstrate a higher likelihood of cognitive impairment,

One such option could be targeting the VEGF receptor-2 (VEGF-R2) specifically. VEGF-R2 is more specific for vascular VEGF signaling and VEGF-R1 seems to be more specific for neurological signaling. In wild-type mice with microsphere-induced cortical capillary obstructions, VEGF-R2 inhibition reduced the pruning of obstructed capillaries and improved clearance of cortical capillary obstructions. Indeed, in supplemental analysis of our study VEGF-R2 levels were increased in Alzheimer’s mice as compared to their wild-type counterparts, whereas VEGF-R1 were decreased in the APP/PS1 mice, further strengthening our hypothesis on the differential effects of VEGF-A on each side of the blood brain barrier, detrimental on one and therapeutic on the other.

**Conclusion**

We recently demonstrated that luminal VEGF-A signaling contributes to leukocyte stalling in brain capillaries and reduced cerebral blood flow in a mouse model of Alzheimer’s disease. Targeting luminal VEGF-A signaling could address local inflammation, blood-brain barrier hyperpermeability, leukocyte recruitment, capillary stalling, and cerebral blood flow reductions. Though our findings may seem contradictory to evidence in the literature suggesting the beneficial effects of VEGF-A in Alzheimer’s disease, VEGF-A may simply have a deleterious effect on the luminal side of the blood brain barrier and a therapeutic effect on the brain parenchyma. We recommend continued investigation in prospective and translational studies to further delineate a difference between luminal and parenchymal VEGF signaling in Alzheimer’s disease.

**Author Contributions**

MA wrote the initial commentary and prepared the figures. OB guided the progress and contributed to the writing of this commentary.

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