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Vascular endothelial growth factor inhibition and proliferative diabetic retinopathy, a changing treatment paradigm?

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Abstract:

Prior to the development of panretinal photocoagulation (PRP) in the 1970s, proliferative diabetic retinopathy (PDR) was the most common cause of blindness in diabetic patients. The diabetic retinopathy study demonstrated that PRP could decrease severe visual loss from PDR by 50%. Since then and for the past four decades, PRP has been the treatment of choice for eyes with PDR. In the past decade, vascular endothelial growth factor (VEGF) inhibition has become the treatment of choice for diabetic macular edema (DME). When treated intensively with anti-VEGF drugs, about one-third of eyes with DME experience an improvement in their diabetic retinopathy severity scale. Randomized clinical trials comparing ranibizumab to PRP and aflibercept to PRP have shown that VEGF inhibitors cause regression of intraocular neovascularization but need to be given on a fairly regular basis. Despite these promising results, concerns about treatment adherence have surfaced. Patients with PDR that are treated solely with anti-VEGF drugs and somehow interrupt their treatment are at a high risk of developing irreversible blindness. Combination treatment of PRP plus an anti-VEGF drug may be the treatment of choice for PDR.

Keywords:

Aflibercept, bevacizumab, diabetic retinopathy severity scale, intravitreal injection, panretinal photocoagulation, pattern scanning laser photocoagulation, proliferative diabetic retinopathy, ranibizumab, vascular endothelial growth factor

Introduction

Diabetes mellitus has been recognized for almost two millennia, yet intraocular complications were not described until the invention of the ophthalmoscope in the 1850s.[1] The invention of the ophthalmoscope in the 1850s allowed the detection and description of the clinical features of diabetic retinopathy. Early on it was recognized that proliferative diabetic retinopathy (PDR) if left untreated led to severe visual loss. In 1876, Manz provided the first description and illustrations of PDR, which he named retinitis proliferans. His drawings portrayed fibrovascular proliferations along the vascular arcades, the optic nerve, and the posterior pole.[2]

The discovery of insulin in 1921 by Best and Banting prolonged the lifespan of diabetic patients. Paradoxically, this increased longevity allowed diabetic complications to occur in more patients. One of these complications was PDR and for decades not much could be done to alleviate visual loss from PDR. In 1930, Houssay and Biasotti[3] demonstrated that hypophysectomy improved glycemic control in pancreatectomized diabetic dogs. As corticosteroids were not available at the time, this observation was not pursued further. In 1953, Poulsen[4] noted that a

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diabetic patient who suffered from a hemorrhagic infarct of her pituitary had the regression of her retinopathy. Pituitary ablation became an option for patients with PDR, once hormone replacement was available. However, the side-effects profile was not insignificant as it included some that were life-threatening.[5]

The ancient Greeks were well aware that direct sungazing during an eclipse was harmful. Theophilus Bonetus (1610–1689) was the first to describe a central scotoma following a retinal solar burn. Once the ophthalmoscope became an indispensable instrument during routine clinical practice, descriptions of the ophthalmoscopic features of retinal solar burns became well known. Since then, several researchers investigated on how to harness solar energy for retinal therapeutical uses.[6] One of these researchers was Meyer-Schwickerath who in 1946 invented a sunlight photocoagulator that focused sunlight through a telescope. The main limitation of this device was that treatment could only be performed during a sunny day. Meyer-Schwickerath continued experimenting with several light sources, and in 1950, the xenon (Xe) arc photocoagulator was born. This eliminated the need for sunlight and produced a stronger beam for photocoagulation.[7]

Aiello *et al*.[8] noticed that diabetic patients with eyes with unilateral chorioretinitis developed much less severe diabetic retinopathy than the fellow nonuveitic eye. They, as well as other research groups, speculated that perhaps causing chorioretinal scars might help regress PDR. At the same time, the first ophthalmic lasers were being developed.

More than half a century ago, investigators interested in diabetic retinopathy got together at Airlie House, Virginia. Important outcomes of this meeting were the decision to run the diabetic retinopathy study (DRS), the design of a standard classification of diabetic retinopathy, and the diabetic retinopathy severity scale (DRSS).[9]

The DRS was conducted in the 1970s. Patients with PDR in at least one eye or severe nonproliferative diabetic retinopathy (NPDR) in both eyes were randomized to either panretinal photocoagulation (PRP) or indefinite deferral of treatment. Eyes that were randomized to the photocoagulation arm could be treated with the Xe arc or argon laser photocoagulation. Patients were seen every 4 months, and photocoagulation could be repeated if retreatment criteria were met. Eyes treated with the argon laser also had direct treatment to the new vessels even if they were on the disc or elevated. In contrast eyes treated with the Xe arc did not undergo direct treatment of elevated new vessels or new vessels on the disc. The primary endpoint was severe visual loss on two consecutive visits. Severe visual loss was defined as a best-corrected visual acuity of $\leq 5/200$. PRP of eyes with high-risk characteristics decreased severe visual loss by 50%. Based on these results, PRP has been the treatment of choice for over 40 years in patients with PDR.[10-12]

In 1948, Michaelson[13] hypothesized that a diffusible and soluble factor was responsible for retinal vascular growth during the development and disease states such as PDR. Since then, the search for this elusive factor X garnered the attention of researchers worldwide. It was not until 1989 that Ferrara and Henzel[14] isolated a growth factor specific for vascular endothelial cells from pituitary follicular cells. They named this new growth factor vascular endothelial growth factor (VEGF). VEGF is currently widely accepted as Michaelson's X factor.

In the past decade, VEGF inhibitors have largely replaced macular laser photocoagulation (MLP) as the treatment of choice of diabetic macular edema (DME). Because of the central role that VEGF plays in the pathogenesis of PDR, these same VEGF inhibitors have recently been studied in the context of PDR.

Pathophysiology

Brownlee[15] has proposed that chronic hyperglycemia leads to the overproduction of superoxide by the mitochondrial electron chain. These reactive oxygen species activate several metabolic pathways that are characterized by advanced glycation end products, activation of protein kinase C, increased hexosamine pathway flux, and an increased polyol pathway flux. The initial clinical manifestations of this metabolic disarray are microvascular alterations such as microaneurysms that progress to retinal ischemia, an increase in retinal vasopermeability, DME, and retinal neovascularization.[16,17] PDR is characterized by intraocular neovascularization, which is a response to retinal ischemia. At the molecular level, PDR is characterized by angiogenesis. Even though other molecules may be involved, the main driver of angiogenesis is VEGF, which is markedly upregulated by hypoxia.[18]

Once VEGF is produced by the ischemic retinal cells, it diffuses toward the retinal vascular endothelial cells. The retinal endothelial cells express several VEGF tyrosine kinase receptors on its surface. VEGF receptor 2 (VEGFR-2) is the major mediator of the angiogenic and vascular permeabilizing effects of VEGF.[18] Binding of VEGF to VEGFR-2 leads to its dimerization and autophosphorylation of intracellular tyrosine residues, which initiates the signal transduction that leads to endothelial proliferation, endothelial survival, transcriptional activation, endothelial migration, and vascular leakage.[19]

Further evidence of VEGF's role in PDR comes from experimental and clinical studies. Animal models of retinal ischemia demonstrate that VEGF mRNA expression is temporally and spatially correlated with retinal neovascularization.[20] Inhibition of VEGF in these animal models inhibited intraocular neovascularization. Furthermore, intravitreal injections of VEGF in normal nonhuman primate eyes cause retinopathy, characterized by intraretinal hemorrhages, retinal edema, venous beading, and microaneurysms.[21] Intravitreal VEGF levels are elevated in patients with PDR.[22,23] Several groups have reported that intravitreal bevacizumab causes rapid regression of retinal and iris neovascularization in eyes with PDR.[24-26]

Panretinal Photocoagulation

Over the past four decades, several studies have shown the value of PRP on PDR.[27] However, PRP is inherently destructive. The reduction in severe visual loss following PRP comes with a price of a loss of peripheral visual field, night vision loss, exacerbation of DME, contrast sensitivity loss, and dyschromatopsia.[28-31] PRP causes destruction of the retinal pigment epithelium (RPE) and adjacent photoreceptors, leaving the inner retina intact. Since photoreceptors are one of the most metabolically active cells in the body, a reduction of photoreceptors will result in a lowered oxygen consumption, which, in turn, will alleviate the ongoing ischemia.[32] The end result is the downregulation of VEGF secretion, which results in PDR regression.[33] In those eyes that respond to PRP and have PDR regression, the effect is usually long-lasting and permanent.[34,35] Despite PRP, up to 15% of eyes continue to lose vision.[36-38] In the DRS presumably, the loss of vision was secondary to continued progression of PDR. In the CLARITY and Protocol S studies, it was a mix of DME and progression of PDR.

Since the landmark study of the DRS, laser technology has evolved. Several lasers with different wavelengths have been used in the past, including the ruby (694 nm), krypton (647 nm), dye, and argon (488 and 514 nm) lasers. Currently, the diode solid-state, aluminum green frequency-doubled neodymium yttrium aluminum garnett (532 nm), and the yellow (577 nm) lasers are most commonly used in ophthalmic practices. The laser beam is targeted on the RPE where the hemoglobin and melanin in the RPE and choroid absorb the laser energy. Laser energy absorption leads to heat generation in the RPE and choroid. The biological response to laser irradiation depends on the laser pulse duration, laser wavelength, and laser irradiance. This response is mathematically described by the Arrhenius integral, which quantifies the tissue damage caused by the laser pulse. This effect is an exponential function of temperature and a linear function of pulse duration.[39]

The incorporation of scanning galvanometers into modern ophthalmic lasers renders them capable of generating a pattern of multispots. However, certain modifications had to be implemented. To precisely place the multi-spot array of burns in the intended site and not be at the mercy of ocular movements, each laser pulse duration had to be shortened. In the ETDRS and DRS, each laser pulse lasted between 100 and 200 ms. With pattern scanning, each laser pulse lasts between 10 and 30 ms.[39] The obvious advantage of pattern scanning lasers is that it permits ophthalmologists to complete PRP quicker. The shorter pulses also produce burns that primarily affect the RPE, the photoreceptors, and outer retina unlike longer-lasting pulses that usually affect full-thickness retina. As the burns do not reach the choroid, patients experience less pain as well. Clinically, these burns appeared smaller and more uniform than those of conventional monospot photocoagulation.[39,40] These differences are clinically important, and treatment parameters need to be modified. Since the pattern scanning laser produces smaller burns, in order to treat the same total retinal area the grid density and the total number of burns delivered need to increase by the reciprocal of the square of the spot size diameter.[41] Chappelow *et al*.[42] highlighted the differences between conventional monospot photocoagulation and pattern scanning laser photocoagulation in the treatment of high-risk PDR. In a retrospective comparative case series of 82 eyes, they showed that using traditional laser settings in the pattern scan laser underperformed conventional monospot laser photocoagulation.[42] When modified parameters were utilized, pattern scanning laser photocoagulation was just as efficient as conventional monospot laser photocoagulation in high-risk PDR.[40]

Anti-Vascular Endothelial Growth Factor

VEGF inhibition has been reported to cause regression of intraocular neovascularization in eyes with PDR.[24-26] The Pan American Retina Collaborative Retina Study Group recently reported the 24-month outcomes of 97 eyes with PDR that were treated with intravitreal bevacizumab.[25] The best-corrected visual acuity and central macular thickness improved significantly from baseline. On average, patients were injected four times per eye (range 1–8 injections) over almost 30 months. Sixty eyes had previously undergone PRP, and of these, 73% had complete regression of neovascularization, 15% had partial regression, and 12% had no regression at all. In the 37 eyes that had no prior PRP, almost half underwent combined PRP and intravitreal bevacizumab. The other half was treated solely with intravitreal bevacizumab. Of these eyes, about 60% required PRP or vitrectomy to control the PDR. Forty percent of these treatment-naive eyes had complete resolution of their PDR with just intravitreal bevacizumab.[25] In eyes with

more advanced PDR caution is required as tractional retinal detachments have been reported following VEGF inhibition.[43]

Comparative Trials

González *et al*.[44] conducted a short-term study where they compared intravitreal pegaptanib sodium, a VEGF 165 antagonist, injected every 6 weeks for 30 weeks to PRP. The pegaptanib-treated eyes fared better in terms of neovascularization regression and visual acuity compared to those eyes treated with PRP.

The Diabetic Retinopathy Clinical Research (DRCR) Network Protocol S aimed to evaluate the effectiveness of ranibizumab compared to PRP in eyes with PDR. In this study, patients were randomized to ranibizumab 0.5 mg intravitreal injection monthly for 3 months. At 4 months, patients were reevaluated and were followed monthly without further injections if neovascularization had completely resolved. If any neovascularization was still present, additional injections were given at months 4 and 5. Beginning at month 6 of the treatment protocol, injections were performed as needed, ceasing further treatments if retinal neovascularization had completely resolved or if no improvement was noted after two consecutive injections. Any patient who developed progressive retinopathy despite monthly injections was allowed to receive PRP. At 2 years, ranibizumab provided better visual acuity outcomes, less visual field loss, fewer vitrectomies were required, and less development of center-involved DME when compared with the PRP group. The advantages of PRP were fewer visits, fewer injections, and greater cost-effectiveness in eyes without DME initially. More than half of the patients in the PRP group needed supplemental laser during the first 2 years of the study (51% vs. 14%).[38] At 2 years, the chances of worsening PDR were higher in the PRP-treated eyes as compared to ranibizumab-treated eyes (42% vs. 34%).[45] Regardless of treatment assignment, a worse baseline DRSS was associated with increased risk of worsening (64% in high-risk PDR or worse vs. 23% in moderate PDR or better). In the PRP-treated eyes, pattern scan laser treatment was associated with PDR worsening (60%), independently of the total number of spots placed, when compared to conventional monospot PRP (39%). In eyes with PDR without DME, there was less-PDR worsening in the eyes treated with ranibizumab (31%) than those treated with PRP (45%).[45] The visual acuity results at 5 years were similar in both groups as were the changes in the letter scores. The mean changes in the visual acuity over the course of the study in the eyes with baseline DME indicated an early benefit for the ranibizumab group that disappeared at 5 years. In the eyes without DME at baseline, little difference was seen in the visual acuity over the course of the study. Visual field preservation was significantly greater in the ranibizumab group compared with the PRP group, but that difference began to decrease at 2 years. At 5 years, the ranibizumab benefit was lower but still greater than PRP. A major concern was the number of patients that were lost to follow-up, excluding deaths only 66% of patients completed the 5-year visit.[46] Despite the inherent destructive nature of PRP, surprisingly the patient-reported outcomes were similar between PRP and ranibizumab.[47] The results of Protocol S are summarized in Table 1.

CLARITY was a multi-center phase 2b, single-blind, randomized, noninferiority trial that compared aflibercept to PRP. It included 232 participants that had active PDR. Patients were randomly assigned to intravitreal aflibercept or PRP. Patients in the aflibercept arm received three consecutive monthly injections and were then followed on a monthly basis with injections performed as needed. Patients in the PRP arm underwent PRP and were assessed every 8 weeks. At 52 weeks, aflibercept was not only noninferior to PRP but also superior to PRP in terms of the visual outcomes. New-onset center involved DME (29% vs. 11%), vitreous hemorrhage (18% vs. 9%), need for vitrectomy (6% vs. 1%), and visual loss (10% vs. 5%) were more likely to occur in eyes treated with PRP than with aflibercept. Furthermore, patient satisfaction scores favored intravitreal aflibercept over PRP. These results were achieved, with a mean of four aflibercept injections. Based on these results, the authors recommended that intravitreal aflibercept might be considered as an alternative treatment for PDR in compliant patients. The main limitation of this study was the limited follow-up of 52 weeks, which precludes the assessment of long-term treatment adherence.[37] The outcomes of CLARITY are summarized in Table 2.

Patients with PDR are often sick and miss scheduled medical appointments. Recent studies have shown the dangers of poor treatment adherence in PDR, particularly in eyes treated with anti-VEGF drugs.[48-50] Wubben and Johnson[50] reported the outcomes of 13 eyes of 12 patients that had their anti-VEGF treatment interrupted for different reasons, including intercurrent illness, noncompliance, and financial issues. Patients were on average absent for 12 months. Upon return, nine eyes had a vitreous hemorrhage, five eyes suffered from neovascular glaucoma, and four eyes had a tractional retinal detachment. Despite aggressive treatment of these complications, 77% of eyes lost ≥3 lines of visual acuity and 46% ended up with a visual acuity of hand motions or worse.[50] Similarly, Obeid *et al*.[49] reported 76 eyes of 59 patients that were lost to follow-up for 6 months or more. They found that there were significantly more eyes with tractional retinal detachment and iris

Table 1: Outcomes of protocol S[38,46]

	PRP (2 years), (*n*=168)	Ranibizumab (2 years), (*n*=160)	*P*	PRP (5 years) (*n*=123)	Ranibizumab (5 years) (*n*=117)	*P*
Mean VA (letters)	80	83		81	80	
Change VA (letters)	+0.2	+2.8	<0.001	3.0	3.1	0.68
≥10 letter improvement (%)	36	43	0.37	41	52	0.47
≥10 letter loss (%)	14	9	0.20	9	6	0.42
≥15 letter loss (%)	10	8	0.42	6	6	0.84
Visual field change (dB)	−422	−23	<0.001	−527	−330	0.04
Need for pars plana vitrectomy (%)	15	4	<0.001	19	11	
Development of center involved DME (%)	28	9	<0.001	38	22	<0.001

PRP: Panretinal photocoagulation, VA: Visual acuity, DME: Diabetic macular edema

Table 2: Outcomes of CLARITY[37]

	PRP (*n*=116)	Aflibercept (*n*=116)	*P*
Mean VA (letters)	79.3	82.6	
Change VA (letters)	-2.9	+1.3	<0.001
≥10 letter improvement (%)	2	5	0.45
≥10 letter loss (%)	15	5	0.009
≥15 letter loss (%)	6	5	0.72
Need for PPV (%)	6	1	0.066
Development of center involved DME (%)	8	3	

PPV: Pars plana vitrectomy, VA: Visual acuity, DME: Diabetic macular edema, PRP: Panretinal photocoagulation

neovascularization in eyes that were managed solely with anti-VEGF as compared to eyes that were treated with PRP.

Several groups have assessed the effects of adding an anti-VEGF agent to PRP in the management of eyes with PDR.[26,51-54] Mirshahi *et al*.[51] compared PRP plus a single injection of bevacizumab to PRP. They reported a significantly higher regression of neovascularization in the combination arm compared to the PRP arm. Similar results were reported by Tonello *et al*.[52] The PROTEUS study compared PRP plus ranibizumab to PRP in high-risk eyes with PDR. They reported that combination therapy of PRP plus ranibizumab was superior to PRP in causing the regression of neovascularization at 12 months of follow-up.[55] A recent systematic review and meta-analysis of anti-VEGF agents in PDR supported VEGF inhibition as an adjunct of PRP.[56]

Eyes with Nonproliferative Diabetic Retinopathy without Diabetic Macular Edema

The DRSS was initially designed for the DRS and was further modified for the ETDRS.[57,58] Worsening in the DRSS correlates with the development of PDR and visual loss.[58,59]

The clinical trials demonstrated the benefit of anti-VEGF drugs in the management of DME. In addition to the functional and anatomic benefits, it was noted that eyes treated with multiple anti-VEGF injections had an improvement in their DRSS.[60] RISE and RIDE studies were multicenter, randomized clinical trials studying the effectiveness of ranibizumab in eyes with DME. In RISE and RIDE, eyes were randomized to receive either 0.5 mg ranibizumab, 0.3 mg of ranibizumab, or sham monthly injections for 24 consecutive months. After month 24, the eyes in the sham arm of the study were eligible to receive 0.5 mg of ranibizumab. After 2 years, 36% of eyes treated with ranibizumab experienced an improvement of at least 2 steps in their DRSS score. In comparison, only 5% of sham-treated eyes had this improvement.[61] Stratification by DRSS demonstrated that the eyes at the highest risk, namely eyes with severe and very severe NPDR (levels 47–53 in the DRSS) of progression to PDR benefit the most. In this sub-set of eyes in the RISE and RIDE trials, 75% had at least a 2-step DRSS improvement.[62] In the VIVID and VISTA trials, eyes were randomized to a loading dose of 5 monthly intravitreal injections followed by a bi-monthly aflibercept injection, a monthly aflibercept injection or MLP. After 2 years, 29%–37% of aflibercept-treated eyes had at least a 2-step improvement in the DRSS compared to 8%–16% MLP-treated eyes.[63] Protocol T of the DRCR network compared aflibercept, ranibizumab, and bevacizumab in eyes with DME. All of the eyes received 6 monthly injections of the drug and then were reinjected according to need. At 12 months, eyes with DME and NPDR treated with bevacizumab (22%) were less likely to experience an improvement in DRSS compared to eyes treated with aflibercept (31%) or ranibizumab (38%). However, these differences vanished by the 2nd year. In eyes with DME and PDR, aflibercept (76%) was superior to ranibizumab (55%) and bevacizumab (31%) in causing improvement in the DRSS.[60] Bonnin *et al*.[64] urge caution when interpreting DRSS improvement. In a retrospective review of 18 eyes that underwent 3 consecutive monthly anti-VEGF injections, ultra-wide-field color photos and fluorescein angiograms were compared at baseline and 1 month after the last injection. They noted that the DRSS score improved by at least 1 step in 61% of eyes. However, the corresponding fluorescein angiograms

in these same eyes showed that there was no arteriole or venule reperfusion in the nonperfused areas. Eyes with DRSS improvement may still be at high risk of developing PDR.[64]

Up until now, treatment of diabetic patients was centered on dealing with the complications of diabetic retinopathy, namely DME and PDR. Given the improvement of the DRSS in eyes with NPDR/PDR with concurrent DME, the question becomes what role, if any, does VEGF inhibition have in eyes with NPDR in preventing progression to PDR or development of DME. To answer this question, there are currently two clinical trials ongoing. PANORAMA (ClinicalTrials.gov: NCT02718326) will try to assess the efficacy and safety of aflibercept for the improvement of severe-to-moderately severe NPDR by measuring the proportion of eyes that improve at least two steps in the DRSS from baseline. The study will randomize eyes into three arms: sham injections and two different dosing regimens of aflibercept. Patients will be followed for 100 weeks. Similarly, the DRCR Protocol W (ClinicalTrials.gov: NCT02634333) will study whether or not intravitreal aflibercept will be able to prevent the development of vision-threatening diabetic retinopathy in high-risk eyes. Eyes will be randomized to sham injections or aflibercept injected at baseline, month 1, month 2, month 4, and then every 4 months until year 2.[62]

Summary

It used to be that if patients with DM lived long enough, they would become blind from PDR. Over the past four decades, blindness from PDR has been significantly decreased by PRP. However, PRP is an inherently destructive procedure that causes the regression of PDR at the expense of loss of peripheral visual fields, loss of color vision, loss of night vision, loss of contrast sensitivity, and exacerbation of DME. Interest in less destructive alternative treatments has grown because of these limitations. VEGF inhibitors cause regression of intraocular neovascularization but need to be given on a fairly regular basis. Given the chronic nature of PDR and the intravitreal half-life of anti-VEGF drugs currently in use, the main disadvantage of anti-VEGF monotherapy for PDR is that these drugs need to be administered periodically for some time. Interruption of treatment can be catastrophic and lead to irreversible blindness. Combination treatment of PRP plus an anti-VEGF drug may be the treatment of choice for PDR.

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Conflicts of interest

The authors declare that there are no conflicts of interests of this paper.

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