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REVIEW ARTICLE

Implantable anti-angiogenic scaffolds for treatment of neovascular ocular pathologies

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Abstract

The retinal physiology can accrue oxidative damage and inflammatory insults due to age and metabolic irregularities. Two notable diseases that involve retinal and choroidal neovascularization are proliferative diabetic retinopathy and wet age-related macular degeneration. Currently, these diseases are mainly treated with anti-VEGF drugs (VEGF = vascular endothelial growth factor), generally on a monthly dosage scheme. We discuss recent developments for the treatment of these diseases, including bioactive tissue-engineered materials, which may reduce frequency of dosage and propose a path forward for improving patient outcomes.

Keywords Pathological neovascularization · Age-related macular degeneration · Diabetic retinopathy · Self-assembly · Hydrogel · Peptide nanofibers · Anti-angiogenic materials

Introduction

The retina is the only part of the central nervous system (CNS) that can be non-invasively imaged via simple photography and hence offers fascinating insight into CNS health. In spite of the relative ease of detection of retinal features and the adoption of techniques such as spectral domain optical coherence tomography (SD-OCT) [1] in clinical practice, patients often seek medical attention only after significant progression of retinal diseases. There may be several factors to potentially explain this delay in presentation. As patients use binocular vision to interpret their surroundings, reduced central vision in one eye may not be readily noticed unless the normal eye is covered. In the case of diabetic retinopathy, affected patients are usually of working age and treatment burden may impact the decision to seek medical attention. These patients may not easily be able to take a day off work every month for treatment. Finally, access to care may limit a patient's ability to reach a retina specialist. Often these patients must drive long distances, and for a patient with compromised vision, especially elderly patients, a caretaker/family member generally has to accompany the patient. Thus, there is an unmet clinical need to reduce the frequency of treatment and visit burden for patients. To understand some of the current clinical options employed to treat these diseases, it is instructive to explore the root causes.

Scope

Retinal diseases such as wet age-related macular degeneration (wAMD) and progressive proliferative diabetic retinopathy (PDR) are major causes of loss of visual acuity (and ultimately, blindness) [2, 3]. The retina in the posterior segment of the eye is the only part of the central nervous system exposed to direct environmental exposures such as radiation. It contains an extraordinary concentration of photoreceptive neurons in the center of the retina called macula. The center of the macula contains the fovea, which mainly has cone photoreceptors that enables high-resolution color vision [4].

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Metabolic disorders such as hyperglycemia, dyslipidemia, and protein misfolding can lead to inflammation, vasculopathy, edema, and neuropathy in the retina [5] and the underlying vascular layer (choroid) [6]—these pathological changes are most noticeable in the macula. Many resultant posterior segment diseases of the eye share a few core causes and symptoms (Fig. 1) but may differ in the mechanistic details and, of course, in the location of the affected tissue microenvironment. For example, different aspects of the complement system is involved in the progression of diabetic retinopathy and wet AMD [7]. In this review, we focus on neovascular posterior segment diseases and strategies to treat and manage the conditions (Fig. 1).

Pathophysiology of wAMD

Blood vessels in the choroid nourish the retinal pigment epithelium (RPE) and the overlying photoreceptors. There are three distinct layers in the choroid: capillaries under the Bruch's membrane (choriocapillaris), arterioles/venules under choriocapillaris (Sattler's layer), and arteries/veins underneath (Haller's layer). Among these layers, the choriocapillaris is highly fenestrated and lacks complete pericyte coverage, perhaps making it most susceptible to age-related metabolic damage. Over time, extracellular aggregates (drusen) build up between the Bruch's membrane and the retinal pigment epithelium (RPE) cells [8]. At a later stage of the disease, severe oxidative stress and resultant inflammatory cascades in the RPE layer disrupt cellular homeostasis of photoreceptors and can lead to their apoptosis [5, 6]. Hypoxia in the RPE microenvironment may lead to the upregulation of angiogenic growth factors such as vascular endothelial growth factor (VEGF) and downregulation of anti-angiogenic factors such as pigment epithelium–derived factor (PEDF), causing formation of immature vasculature in the choriocapillaris, which are prone to leakage and clotting [6]. The fluid build-up and associated immune reaction may disrupt the basement membrane under RPE layer (Bruch's membrane) and the extracellular fluid may enter the photoreceptor layer (Fig. 1c).

Pathophysiology of PDR

The American Diabetes Association estimates that about 10% of Americans have diabetes [9]. Among American diabetic patients above 40 years of age, more than 25% suffer from retinopathy [10]. In these patients, hyperglycemia leads to

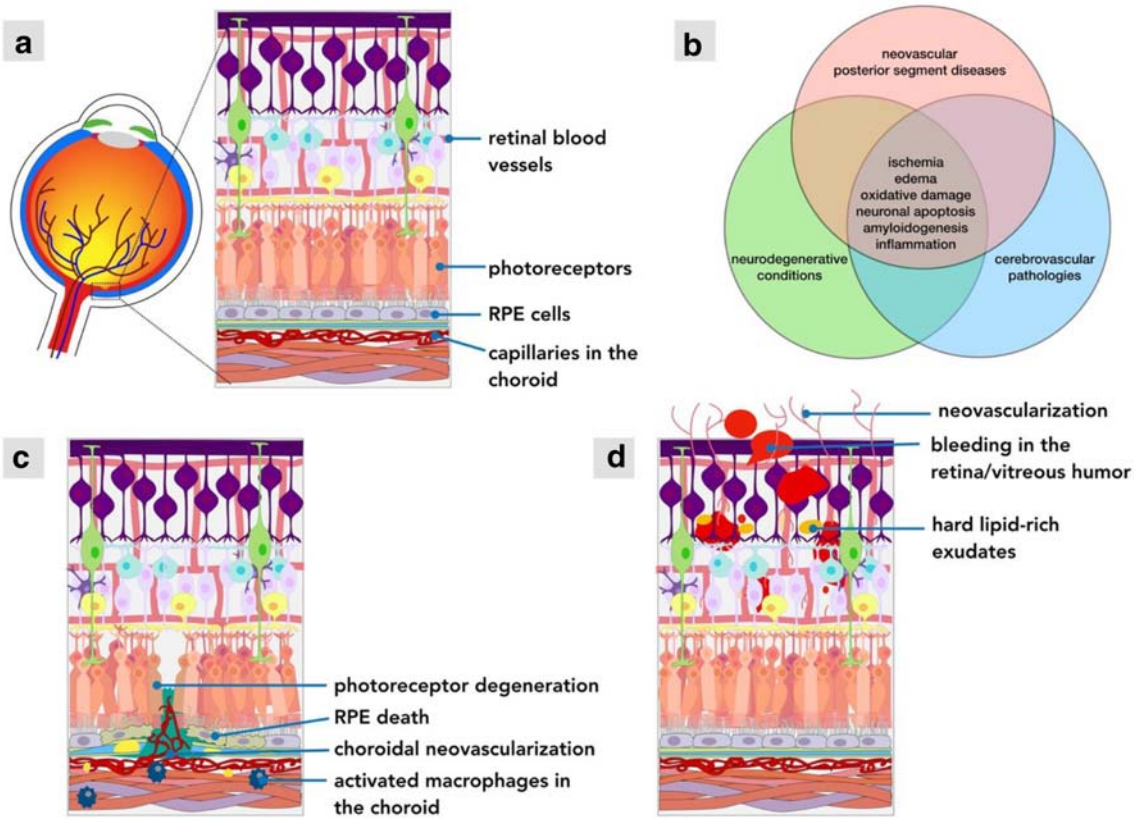

Fig. 1 **a** Retinal anatomy (RPE: retinal pigment epithelium). **b** The confluence of pathological factors culminating in neovascular posterior segment diseases are similar to other neurodegenerative diseases and cerebrovascular diseases. Pathophysiological features of **c** wet age-related macular degeneration and **d** diabetic retinopathy

upregulation of vascular endothelial growth factor (VEGF) [11]. Patients progress through various stages of non-proliferative retinopathy and some eventually develop proliferative disease characterized by the development of immature blood vessels on the retina that are prone to leakage, leading to distortion of the retinal tissue and even tractional retinal detachment (Fig. 1d) [12]. Diabetic retinopathy is characterized by microaneurysms, blot hemorrhage, aberrant neovascularization, and the presence of hard exudates in the retina. Hyperglycemia also results in macular edema and disruption of the blood-retinal barrier (Fig. 2) [5]. If left untreated, these retinal changes lead to reduced vision.

Hyperglycemia leads to the activation of several pathways that cause the accumulation of advanced glycated end (AGE) products, capillary constriction, and hypoxia in the retinal microenvironment [5, 13]. These conditions signal the upregulation of VEGF that leads to retinal neovascularization [14, 15]. These anchored fibrotic vessels are also prone to intravitreal hemorrhage; hence, they are sometimes referred to as leaky vessels [5]. Moreover, inflammation, pyroptosis of Müller cells, and apoptosis of retinal neurons also contribute to the progression of diabaetic retinopathy [16–18]. Pro-inflammatory molecules and pathways involved in diabetic retinopathy are topics of ongoing research [19–22]. A crucial step in the development of PDR involves the loss of pericytes around the endothelial cells [23] and damaged endothelial tight junctions, which lead to the breakdown of the blood-retinal barrier [24–26]. Resulting vascular fluid accumulation in the interstitial space in the macula (macular edema) can severely affect central vision [27, 28]. Soft exudates and retinal leukostasis accompany observable circulatory alterations within the retina [29].

Similarity of wAMD and PDR with other CNS diseases

PDR, wAMD, and other notable retinal diseases such as retinal vein occlusion [30] share common characteristics, such as chronic inflammation, oxidative stress, upregulation of vascular endothelial growth factor, aberrant neovascularization, and edema [31], although they occur in different tissue microenvironments [32]. As these neovascular pathologies share some common causal pathways, anti-angiogenic drugs can be broadly effective against these diseases. Age is a predominant risk factor for wAMD, as is the case for many neurodegenerative diseases [33, 34]. Age-related macular degeneration and Alzheimer's disease share risk factors (hypertension, hyperlipidemia, etc.) and pathological mechanisms (oxidative stress, inflammation, complement activation, neurodegeneration, amyloid deposition, disruption of blood-brain barrier, etc.)—although it remains to be determined whether they share etiologic roots [35–37]. Both diabetic retinopathy and age-related macular degeneration may also be considered as sub-types of cerebrovascular diseases, i.e., diseases associated with microvascular abnormalities in the central nervous system (Fig. 1b). Pathologic alterations in the cerebral microvasculature also leaves its imprint on the retinal structure; hence, retinal imaging may be used in the future as a cost-effective screen for diseases such as Alzheimer's, frontotemporal

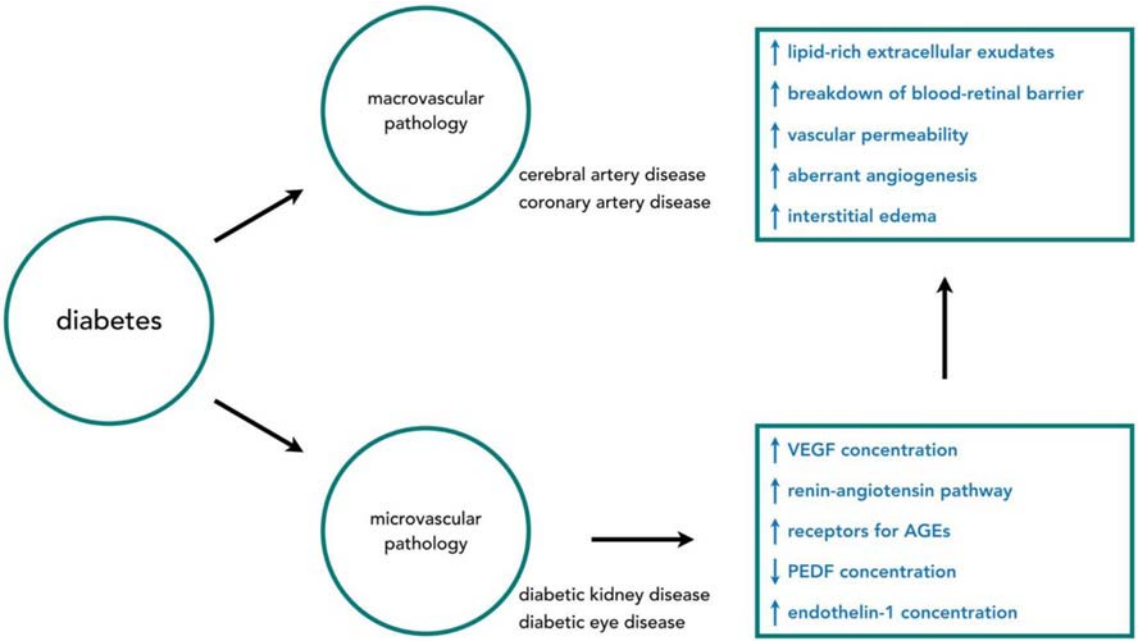

Fig. 2 Pathological disease progression for proliferative diabetic retinopathy. VEGF vascular endothelial growth factor, AGE advanced glycated end-products, PEDF pigment epithelium-derived factor

dementia, and amyotrophic lateral sclerosis [37–39]. As the retina is the only part of the central nervous system directly exposed to light, photo-induced apoptosis of relevant cells may also be a prominent source of risk [40].

Diagnosis and standard of care

The primary goal of therapies is to increase best correlated visual acuity (BCVA) in patients, compared with the baseline before beginning the treatment. For secondary measurements of improvement, SD-OCT (reduction in sub-retinal thickness and decrease in the volume of accumulated retinal fluid) and fluorescein angiography (shrinkage of visible lesion area) can be useful techniques to document disease progression or regression. Adverse events associated with the treatment (e.g., photophobia, intraocular inflammation, endophthalmitis, and pain) may limit the appeal of a clinical option such as intravitreal injection. Current therapeutic approaches include laser photocoagulation [41], intraocular injections of steroids (e.g. dexamethasone and triamcinolone acetonide) [42, 43], anti-VEGF drugs [44–46], and in extreme cases, vitrectomy (Table 1). Of these options, anti-VEGF therapies have rapidly become the front-line therapies used by retina specialists as they, unlike steroids, are not associated with the risks of increased intraocular pressure and cataract formation [47].

When injected into the vitreous humor, anti-VEGF drugs attenuate neovascularization and reduce retinal edema [48]. Three FDA approved anti-VEGF drugs are available for intraocular injection: pegaptinib [46, 49], ranibizumab [50], and aflibercept [51]. Ranibizumab is an antibody fragment whose active domain is similar to another widely used monoclonal antibody, bevacizumab. Ophthalmologists often use bevacizumab as an anti-VEGF intraocular therapeutic [44, 45] since it has a similar activity profile as ranibizumab and is less expensive per injection [52]. Additionally, anti-inflammatory agents can be used to mitigate intraocular inflammation and edema. Notably, intravitreal administration of the steroids (such as triamcinolone acetonide) shows efficacy in reducing edema associated with DR. [53, 54] Semi-permanent intravitreal implants of steroids (such as Allergan's Ozurdex© and Alimera's Iluvien®) have also been successful in treating edema associated with PDR [55]. Similar implants for anti-VEGF drugs are not on the market—however—Genentech has developed a port-delivery device for long-term release of ranibizumab, which is currently in clinical trials [56].

Pitfalls of current treatment options

Laser photocoagulation therapy may cause side effects, such as the formation of blind spots and scars on the retina [57]. Moreover, it is less effective than intravitreal anti-VEGF therapy in treating macular edema [58]. On the other hand, intravitreal injections carry the risks of endophthalmitis, retinal detachment, and cataract formation—and these risks increase with dosing frequency [59]. Lack of convenience is another major issue for patients. For example, a healthcare professional must administer the intraocular injection of bevacizumab on a monthly schedule, which requires topical anesthesia [44, 60, 61]. The high frequency of dosage (monthly) is needed due to the need to maintain a steady level of anti-angiogenic therapeutics in the vitreous humor, leading to poor patient comfort. A major limitation of current technologies is that they manage the pathological outcome (vascular leakage, neovascularization) but not the root causes (hypoxia, oxidative imbalance, and cellular apoptosis). Even if an anti-VEGF therapy is fully successful, it does not aim to regenerate RPE cells or damaged photoreceptors. Thus, the optimal outcome of these therapies is *maintenance* of remaining vision, rather than *restoring* vision. This is where new stem cell and gene therapy–based options may be most appealing. FDA has already approved Spark Therapeutics' Luxturna (voretigene neparvovec) for biallelic RPE65 mutation-associated retinal dystrophy, which

Table 1 Selected non-steroidal intravitreal drugs targeting neovascular posterior segment diseases such as wAMD and PDR

Drug	Company	Target	Dose interval	Stage
Bevacizumab	Genentech	VEGF-A	4 weeks	On market
Ranibizumab	Genentech	VEGF-A	4 weeks	On market
Aflibercept	Regeneron	VEGF-A, VEGF-B, PlGF	4 weeks	On market
Brolucizumab	Novartis	VEGF-A	8/12 weeks	FDA-approved
Faricimab	Genentech	VEGF-A & Ang2	8/12 weeks	Phase III
Conbercept	Chengdu Kanghong	VEGF-A, VEGF-B, PlGF	8/12 weeks	Phase III
Abicipar	Allergan	VEGF-A	8/12 weeks	Phase III
Risuteganib	Allegro	Integrins	N/A	Phase II
Sunitinib	Graybug	VEGF receptors	N/A	Phase I/II
AR-13503	Aerie	Rho kinase & protein kinase C	N/A	Pre-clinical

may help dictate a road map for the development of such once-and-done therapeutic options.

Technologies in the pipeline

Anti-VEGF drugs smaller than monoclonal antibodies (mAbs), such as Novartis's brolucizumab (a humanized single-chain variable fragment recently approved by the FDA) [62] and Allergan's abicipar (a designed ankyrin repeat protein), have shown potential in the clinical trials in treating neovascular ocular diseases over extended periods compared with current anti-VEGF drugs (Fig. 3; Table 2) [63–66]. Such next-generation anti-VEGF molecules have a much lower molecular weight compared with monoclonal antibodies (e.g., 26 kDa for brolucizumab, compared with 150 kDa for mAbs) and high affinity for the target growth factors, which may facilitate tissue penetration and enhanced intraocular retention (Table 1). Bispecific antibodies (such as Genentech's faricimab) that can bind to more than one biological target may be useful for blocking multiple pathologic pathways at once.

However, recent failures of high-profile candidates against wet AMD (e.g., Ophthotech's Fovista [67]) and other retinal diseases (Genentech's lampalizumab [68]) have somewhat tempered investor confidence in the drug candidates using newer approaches such as complement inhibition [69]. Notably, Rosenfeld and Feuer have called for limiting the use of retrospective subgroup analyses from phase II trials for the design of expensive phase III clinical trials [70] as the biases involved in the data selection may suggest a rosier view of the chances of success in the phase III [71].

Topical eye drops have so far not been a clinical option due to the difficulty of targeting the aberrant vessels in the posterior segments. However, currently, eye-drop drugs such as PAN-90806 and pazopanib have demonstrated potential for treatment of neovascular posterior segment diseases [72, 73].

Gene therapies, such as Luxturna, can potentially be transformative for the retinal disease landscape [74, 75]. Immune reaction to the viral vectors in this context may not be a major problem as the vitreous humor and the retina are relatively immuno-privileged. However, the chance of off-site immunogenicity cannot be completely ruled out as the blood-retinal barrier can be compromised in diseases such as wAMD and PDR. Another complication is that wAMD and PDR can be polygenic in nature and can potentially be tricky to target with gene therapy.

Embryonic stem cells (ESCs) [76–78] or induced pluripotent stem cells (iPSCs) [79, 80] can be used to create suspensions or patches of RPE cells that can be implanted subretinally for improving physiologic healing responses [81, 82]. The surgical procedure for the implantation can be risky, and there is some risk of retinal detachment and adverse immunologic reaction—but a major advantage of the treatment is that it can potentially be used to treat both dry and wet phenotypes of retinal diseases. The therapy targets a causal factor of retinal diseases: oxidative damage to and apoptosis of RPE cells. The cellular reprogramming of autologous somatic cells into iPSCs involves risk of introducing oncologic mutations, which can be overcome [83]. Allogeneic iPSCs, unlike autologous iPSCs, have an additional route of graft failure: through immune rejection mediated by microglia, T cells, and B cells [84].

Tissue-engineered scaffold–based alternatives

There is a clinical need for developing functionalized peptide-, protein-, or polymer-based scaffolds to create injectable (or otherwise easily implantable in the vitreous humor) biomaterial matrices to attenuate neovascularization in vivo. These matrices need to be anti-angiogenic themselves [85] or should be able to release sequestered anti-angiogenic therapeutics in a temporally controlled fashion (Fig. 3) [86]. If a material can be easily syringe-aspirated, injected, and reassembled in the vitreous humor, a sustained and site-specific response for disease management can be engineered. The matrix can have multimodal efficacy if it contains therapeutics that have different mechanisms of action. Even for delivering stem-cells into the subretinal space, a biodegradable scaffold may afford improved cellular integration with host cell microenvironment [83].

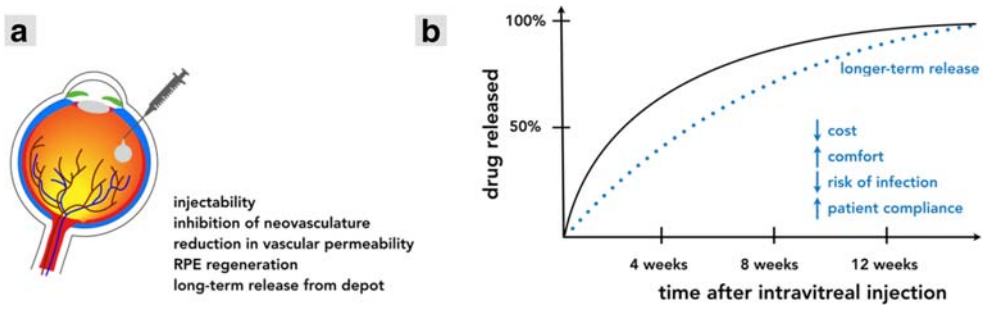

Fig. 3 Desirable features for intra-vitreally injected drugs/bioactive materials. Efficacy aside, longer durability of drugs or materials (blue dotted line, as compared with burst release in case of solid black line) in the vitreous humor enables improved patient comfort and reduces patient compliance issues

Table 2 Selected active patents on wAMD and PDR targeting VEGF or VEGF receptors

Patent	Drug	Technology platform	Inventor
WO2000075319A1	Aflibercept	Modified chimeric polypeptides	Papadopoulos et al.
WO1998045331A2	Ranibizumab	Humanized monoclonal antibody fragment	Baca et al.
WO1998018480A1	Pegaptanib	PEGylated RNA aptamer	Janjic et al.
WO2007112675A8	Conbercept	VEGF receptor fusion protein	Yu et al.
WO2016073918A1	Brolucizumab	Single chain antibody fragment (scFv)	Sallstig et al.
WO2015069668A1	Abicipar	Designed ankyrin repeat protein (DARPin)	Hohman et al.

Specific to ophthalmologic applications, a multi-modal release platform can decrease the frequency of injection while targeting multiple mechanisms of disease progression. Decreased frequency of in-patient visits would improve patient compliance and long-term efficacy, and lower the chances of deleterious side effects (e.g. endophthalmitis and retinal detachment). There are distinct burdens associated with intraocular drug delivery: solubility, pH, viscosity, buffer capacity, ocular toxicity, and proper CMC (chemistry, manufacturing, and control). To mitigate risk and increase the likelihood of success, researchers need to of course demonstrate efficacy of the implants in vitro *and* in vivo in the preclinical phase before beginning clinical trials. These experiments may inform the iterative design and optimization of the therapeutic approach.

Broadly, the anti-angiogenic tissue-engineered scaffolds may impact other areas of disease management where the sequential release of agents can potentiate tailorable wound healing, drug delivery, and hemostasis.

The primary topics we aim to address in the following sections are: (a) modular scaffold design, (b) anti-angiogenic scaffolds, (c) scaffolds for tailorable multi-modal release of anti-angiogenic molecules, (d) anti-inflammatory scaffolds, and (e) challenges for clinical translation of tissue-engineered solutions. Our goal is to establish a blueprint for combinatorial therapies for drug-device combinations useful for improving the clinical outcome and comfort of patients suffering from neovascular retinal diseases.

Modular scaffold design

A tissue-engineered scaffold is a fibrous material architecture that resembles extracellular matrix and can affect the tissue environment when implanted in vivo. Such scaffolds can either be prepared from biological sources (e.g., decellularized tissue scaffolds) or be prepared synthetically (e.g., polymer- and peptide-based scaffolds). Such scaffolds can be modular or multi-component, where different components determine aspects of the material and biochemical properties of the scaffold (Table 3). Such components can often be divided into structural (*medium*) and functional (*message*). Examples of functional components include sequestered small molecules or biomolecules, as well as cells supported in the scaffold.

An interesting class of multicomponent scaffolds is the group of self-assembled peptide-based scaffolds containing biofunctional domains [89, 93, 95]. The functional moiety is directly built into the primary structure of the building block of the scaffold. As the building blocks self-assemble into biofunctional three-dimensional scaffolds, the *message* carried by the functional domain becomes immobilized onto the *medium*, resulting in sustained display of functional epitopes attenuated only by the biodegradation or disassembly of the scaffold [100]. Such functionalized scaffolds have been used for angiogenesis promotion [89, 93, 101] and inhibition [85], chondrogenesis [102], and dentinogenesis [95].

The self-assembling peptide platform is suitable for a variety of biological applications through attachment of functional moieties at the termini, which preserves the nanofibrous self-assembly of the conserved core fibrillizing domain. Functional moieties mimicking the active sites of a relevant protein can attach to either terminus of the peptide in the design process. This platform model ensures that one can make hydrogels with excellent material properties and desirable signals embedded in the peptide. Not only can cells attach to the nanofibrous hydrogel but they can also receive specifically designed cues from the signaling domain of the peptide. Thus, the attachment of an anti-angiogenic sequence [103] to the base peptide sequence yields an anti-angiogenic peptide hydrogel that can persist for months in the host tissue [85]. The anti-angiogenic sequence may be chosen from natural proteins and peptides such as Kringle (domain 5) [104–107], laminin-1 [108], and histidine-proline-rich glycoprotein [109].

Potential of combinatorial therapy

In addition to the covalently attached functionality, one can aim to exploit the interaction of non-covalent sequestration of ant-angiogenic and the anti-inflammatory molecules within the amphiphilic nanofibrous hydrogel. Such combinatorial approach may complement the slow dissociation/release of the anti-angiogenic peptides from the nanofibers with a more rapidly delivered anti-angiogenic or anti-inflammatory therapy

Table 3 The toolkit for rational design of self-assembling peptides

Desired trait	Feature of primary sequence	References
Self-assembly	Central β-sheet forming domain (CFD)	ref. [87–92]
Functionality	Flanking epitope domains (FED)	ref. [85, 89, 93–95]
Biodegradability	Domains cleavable by matrix proteases	ref. [89, 93]
Biocompatibility	Peptide sequence, low immunogenicity	ref. [85, 89, 94–96]
Material property	Ratio of the lengths of CFD and FED	ref [87, 90]
Drug delivery	Charged amphiphilic nanofibrous matrix	ref [96–99]

that synergistically counteracts neovascularization and edema in the retina. Self-assembling peptide platforms has been used for the controlled release of small molecule drugs, growth factors, and cytokines [96–99]. The delivered compound can be varied in regard to charge, amphiphilicity, and size [96–99].

The combinatorial formulations may yield valuable insights into treatment paradigms that focus not just on control of neovascularization but also on the modulation of inflammation. Such a bi- or multi-modal therapeutic strategy may reduce the need for frequent intraocular injections, and enable targeting of disease population non-responsive to current standard-of-care treatment protocols.

Choice of animal models

The choice of an appropriate animal model to evaluate the efficacy of the therapeutic strategies is crucial. While hypoxia-induced models result in rapid development of retinopathy [110, 111], they lack the reproducibility of chemical or laser-induced pathology. Pancreatectomy or treatment with streptozotocin/alloxan can cause marked induction of a diabetic phenotype with the manifestation of DR-like pathologies in genetic mouse or rat models; however, lack of animal welfare may be a potential issue with this model. For mimicking generic retinal degeneration, Royal College of Surgeons (RCS) rat model has been used with some success. The RCS rat has a defective Mertk gene [112] that limits the ability of ability of RPE cells to phagocytose and recycle segments of photoreceptors that are degraded normo-physiologically [76]. Arguably, one of the most refined models is laser-induced photocoagulation (LIPC) of the central retinal vein. This model in mice, rats, pigs, and monkeys has gained favor in creating defined and repeatable lesions on the retina. LIPC in the retina of multiple species has proved to be a reproducible model. If the model is used in genetically obese and hyperglycemic animal models (such as Otsuka Long-Evans Tokushima Fatty, OLETF, rats), the relatively lower cost involved may allow researchers to test more combinations of scaffolds and drugs delivery vehicles prior to tests in large animals. PDR-like pathological phenotypes (e.g. aberrant angiogenesis, retinal edema, inflammation, and vascular leakage) has been observed in the LIPC retinal vein occlusion model [113]. Porcine laser-induced RPE injury model recapitulates AMD-like pathological phenotype but involves significantly higher cost and complexity [83]. Potentially, the optimal model for mimicking retinal pathologies are non-human primate models (such as rhesus monkeys) as they have a similar macular and foveal physiology compared with humans (similar thickness of retinal layers, for example) [84]. However, there are very few centers equipped to carry out the experiments in a humane manner, and this scarcity limits the application of the model. For the interest of space, we have described only the LIPC rat model in detail below (for broader discussions on animal models of wAMD and PDR, readers are referred to prior reviews on the topic [110, 111, 114–116]).

LIPC rat model The model is useful for the evaluation of a variety of ocular therapeutics [117–125]. Notably, the laser-induced retinal disease model shares the following similarities to the pathologic burden in humans: (a) rapid neovascularization of the retina and (b) development of inflammation and retinal edema. Following laser treatment, the rats can receive an intravitreal injection of a therapeutic into the vitreous humor via the pars plana immediately following confirmation of retinal neovascularization and retinal edema by confocal scanning laser ophthalmoscopy (cSLO) and SD-OCT. [121, 126] Prior to starting anti-angiogenic treatment with the injectable formulations, it is important to monitor the onset and progression of clinical symptoms of disease progression. Fundoscopy, fluorescein angiography, and SD-OCT may be used to image neovascularization induced in the posterior segment [121, 122, 126–138]. Imaging is performed on anesthetized rats prior to laser treatment and post-injection. The total area of nonperfusion (dark) and leakage (bright) on wide-field angiography can be valuable indicators of vascular permeability. Total retinal volume centered on the optic nerve (measured from the internal limiting membrane [ILM] to the retinal pigmented epithelium [RPE]) and maximal retinal thickness (ILM to RPE) also provides measurable clinical outcomes. Finally, the animals are sacrificed for histological staining of enucleated eyes to determine cellular infiltration, matrix

deposition, the overall volume of implants, and their intraocular degradation [96].

Challenges for clinical translation

The choice of investing in an early-stage biotech company in the ophthalmology space is riddled with risks. In the intraocular diseases area, failure of multiple biologics in the last few years has raised questions about due diligence in the preclinical stage and the design of clinical trials. However, even in the best of circumstances, there is an inherent chance of unforeseen outcomes after a drug is approved, perhaps best illustrated by the post-approval failure of Affymax [139]. Hence, it is perhaps essential for biotech entrepreneurs to invest time in formulating the optimal series of experiments and market analyses for de-risking a technology (Fig. 4).

In retinal diseases, such strategies are further complicated by lack of gold-standard animal models and the existence of variable phenotypes and causal pathways for each disease. At the same time, intraocular therapy has a few distinct advantages as well: ease of access to the tissue through simple injection, reliable technical capabilities for detecting and cataloging disease progression/regression, and relatively lower chance of immune reaction and systemic side-effects associated with intravitreal injection due to immune privilege, low tissue flow, and the presence of blood-retinal barrier.

A real improvement in the administration of effective drugs and resultant patient comfort may come from modalities that can be used in a pharmacy (intravenous) or even at home (oral, eye drops), which may somewhat negate the aforementioned advantages. A major challenge for new entrants to the market who want to prolong administration intervals or pursue alternative routes of medication is maintaining relevant therapeutic concentration in the affected niche.

Another challenge is selection of non-promiscuous molecular targets. Targeting VEGF, VEGF-receptors, or VEGF-producing cells may have undesirable effects elsewhere in the body. Therapeutics that induce apoptosis of cells need to be confined within the diseased microenvironment. And perhaps one of the trickiest problems to solve is restoring the disrupted barriers among the distinct retinal microenvironments. Solving these problems, while maintaining safety and efficacy in a relevant disease model, then allows a startup to start navigating the tricky regulatory landscape for potential approvals that differ among the target markets. A final doublet of challenges the startup then needs to solve are deciding the price of the drug that the market can bear and convincing ophthalmologists to take up a new drug over existing "good enough" cheaper alternatives. The delay between attracting investments and the eventual recoupment of the investment may be over 5 years and hence thus to many investors they may be undesirable. We have a policy recommendation that may help attract more investments in this space: a unified regulatory landscape in developed countries so that an approval in one of the major (US/Canada/EU/Japan) markets would result in conditional approvals in other markets simultaneously.

Conclusions

Retinal diseases such as wet macular degeneration and proliferative diabetic retinopathy are multi-factorial conditions, where the patients have to comply with a strict regimen of monthly intraocular injections. Current standard-of-care drugs such as anti-VEGF antibodies are effective but suboptimal therapeutics, suffering from a high burden of compliance needed from suffering patients. Recent progress in the development of next-generation biologic drugs and implantable/

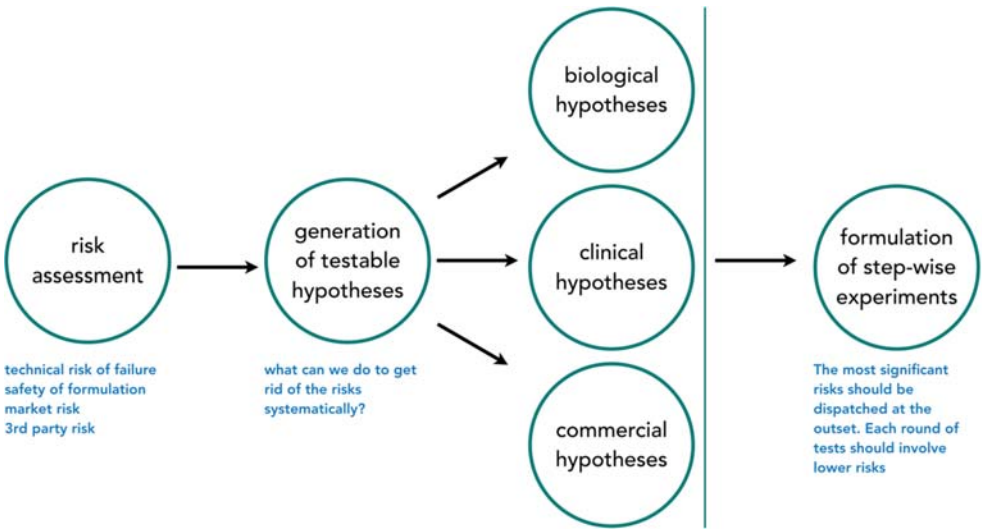

Fig. 4 A systematic appreciation of the variety of risks involved in the development of a biologic drug is crucial for increasing the chance of success and delivering the best return on investment to investors. The graphic was partly inspired by a blog by Dr. Michael Gilman

injectable biomaterial scaffolds may lead to a less frequent visits for the patients, increasing the quality of life for millions. We cover a few promising developments in "once-and-done" treatments such as stem-cell therapy and compare them against other biologics. Injectable self-assembled peptide hydrogels are noted for their potential to act as intrinsic anti-angiogenic scaffolds, as well as for their ability to sequester and deliver biologics over longer time periods—acting as a facile *active* delivery medium.

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Compliance with ethical standards

Conflict of interest V.A.K. has equity interests in start-up companies attempting to translate peptides from peptide-based technological platform.

Declaration of informed consent and animal studies Not applicable for this manuscript.

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