Title
Chapter Fourteen Scleral Mechanisms Underlying Ocular Growth and Myopia

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Publication Date
2015

DOI
10.1016/bs.pmbts.2015.05.005

Peer reviewed

HHS Public Access

Author manuscript

Prog Mol Biol Transl Sci. Author manuscript; available in PMC 2016 June 12.

Published in final edited form as:

Prog Mol Biol Transl Sci. 2015 ; 134: 241–248. doi:10.1016/bs.pmbts.2015.05.005.

Scleral Mechanisms Underlying Ocular Growth and Myopia

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Abstract

In the regulation of ocular growth, scleral events critically determine eye size and thus the refractive status of the eye. Increased scleral matrix remodeling can lead to exaggerated eye growth causing myopia and additionally increased risk of ocular pathological complications. Thus, therapies targeting these changes in sclera hold potential to limit such complications since sclera represents a relatively safe and accessible drug target. Understanding the scleral molecular mechanisms underlying ocular growth is essential to identifying plausible therapeutic targets in the sclera. This section provides a brief update on molecular studies that pertain to the sclera in the context of ocular growth regulation and myopia.

1. INTRODUCTION

The sclera, the outermost layer of the eye, performs many key functions. Apart from offering protection to the interior ocular structures, it determines the final shape and size of the eye, serves as the anchor for extraocular muscles, and provides support for the ciliary muscle, which subserves lenticular accommodation. It also provides channels for blood vessels and nerves serving intraocular structures and allows for the exchange of fluids, including aqueous humor entering the choroid via the uveoscleral pathway. The sclera is predominantly made up of collagen, with interspersed fibroblasts that produce and maintain its extracellular matrix (ECM). It undergoes several changes during the development and progression of myopia, which are at first subtle at the gross anatomical level, but may give way to more substantial changes accompanied by scleral thinning and weakening, leading to pathological complications involving the normally protected retina and choroid—such as maculopathies, retinal schisis, and detachment, with highly myopic eyes being most at risk. A thinner and biomechanically weaker peripapillary sclera in myopia can affect the biomechanics of the lamina cribrosa explaining the increased susceptibility for glaucomatous optic nerve damage.[1,2]

2. STRUCTURAL AND BIOMECHANICAL CHANGES IN MYOPIA

Structural and biomechanical changes in the myopic sclera of human eyes are well documented; apart from being thinner than normal, its glycosaminoglycan and collagen contents are reduced and its fibril assembly disorganized, rendering it biomechanically weaker.[3–6] Humans with moderate as well as high or pathological myopia have been shown to have thinner than normal scleras, with reductions in the thickness of the posterior sclera

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up to 31% of the normal mature human sclera being reported in pathological myopia. [4,7] The most often studied biomechanical property of the sclera is creep rate, which represents the extension of the sclera over time when a constant load is applied. Increases in creep rate are consistent with decreases in biomechanical stability, and for sclera tissue from myopic eyes, increases of over 200% compared to values for normal eyes have been documented.[8,9] Such biomechanical changes in the sclera facilitate elongation of the eye in myopia, and in their more exaggerated form, may lead to posterior staphylomas, corresponding to localized mechanical failure of the sclera. Scleral reinforcement surgery, which currently represents the only treatment option for such complications, is not without risk of complications.[10–12] A seemingly and increasingly plausible option would be to inhibit the scleral changes underlying the excessive elongation in myopia. Indeed, the main rationale behind the research directed at understanding scleral molecular mechanisms underlying myopia development has been to devise less invasive therapies to promote natural matrix deposition, and to improve scleral strength by way of resisting ocular elongation and slowing myopia progression. Research directed at the visual and retinal events that trigger such anomalous growth and the nature of the signal cascades generated is discussed elsewhere in this chapter. Relevant scleral studies are summarized here.

3. MOLECULAR CHANGES IN MYOPIA

The structural and biomechanical changes in the myopic sclera alluded to above are products of biochemical and molecular changes in the sclera. Detailed characterization of molecular changes in the sclera has been possible with the establishment of animal models for myopia (reviewed in detail in Ref. 13). Note that much of this research has focused on mammalian models, mostly the tree shrew, because their sclera is most like the human (primate) sclera, comprising a single fibrous layer as opposed to the bilayered sclera of the chick. In addition to confirming findings from studies in humans of decreased glycosaminoglycan and collagen content in myopia,[4] such studies have linked scleral changes in myopic eyes to altered expression of a number of genes, including collagen (predominantly type-I), matrix metalloproteases (MMPs), tissue inhibitors of MMPs (TIMPs), FGF receptor-1, TGFβ, and integrins (reviewed in detail in Ref. 14).[15–17] Studies involving the tree shrew model have shown reduced scleral hydroxyproline and levels of sulfated glycosaminoglycans at the posterior pole of myopic eyes, suggesting reduced collagen accumulation and proteoglycans, respectively. [18] This reduction in scleral collagen content is accompanied by reduction in collagen fibril diameter.[19,20] Collagen constitutes 85–90% of the total scleral protein, with collagen type-I showing by far the highest expression (>99%) of the numerous collagen subtypes.[21] Nonetheless, other collagen subtypes, such as type III, V, and VI, have also been identified in human sclera,[22] and as many as 11 collagen subtypes (I, III, V, VI, VII, VIII, IX, XIII, XV, XVI, and XVII) have been reported in tree shrew sclera.[23] The various collagen subtypes do not appear to be uniformly affected in myopia. However, a reduction in the collagen subtype ratio (V/I) has been linked to myopia, with speculation that it may be important in determining alterations in fibril diameter in myopia.[23] In tree shrews, higher levels of the active form of MMP-2, an enzyme associated with breakdown of collagen and proteoglycans, and decreased levels of TIMP-1 have also been reported in myopic scleras.[24,25] In addition, downregulation of collagen binding integrin subunits α1, α2, and

β1, as well as TGFβ isoforms, particularly TGFβ1, and upregulation of FGF receptor-1 have been linked to myopia development.[16,26–28] Apart from these studies tying growth factors to scleral remodeling, recent studies in the guinea pig model have implicated two second messengers, cyclic AMP and cyclic GMP; specifically, both scleral cyclic AMP and cyclic GMP levels were found to be increased in form deprivation myopia and subconjunctival injections of activators of these second messengers induced myopic shifts in normal animals, while their inhibitors inhibited form deprivation myopia.[29,30]

A point of relevance is that most of the molecular changes in the sclera discussed above are reversible. For example, it has been shown that when myopia-inducing experimental treatments are terminated, the expression levels of several of these molecules generally return to baseline in eyes undergoing recovery from induced myopia.[14,25,31] Furthermore, biomechanical testing showed that scleral strips from tree shrew eyes recovering from induced myopia were less extensible than those from myopic eyes, implying some recovery of biomechanical properties as well.[8] Together these observations provide strong rationale for intervening at a molecular level to “rescue” the myopic sclera.

4. RECENT ADVANCES IN MOLECULAR STUDIES

The studies summarized above heavily relied on real-time PCR as well as standard biochemical assays. However, with rapid advances in technologies, it is now possible to study genome-wide gene expression profiles. Thus, related studies in human donor tissue have further opened up the scleral molecular landscape, identifying many new genes in the sclera that have never been studied thus far, and signifying that although collagen occupies the most “real estate” (90%), there may be several other important players participating in maintaining normal scleral health.[32–34] Likewise, recent studies involving the tree shrew model have identified many new candidate genes belonging to families such as cell signaling/matrix-degrading/structural proteins in myopia development.[26,35] Genes reported to be upregulated in the sclera during myopia development include TGFβ receptor-3, TGFβ-induced protein ig-h3, and MMP14, while TGFβ1, TGFβ2, thrombospondin-1, tenascin, osteonectin, osteopontin, TIMP-3, and a disintegrin and metalloproteinase with thrombospondin motifs 5 (ADAMTS5) were downregulated.

Across many fields, the roles of micro (mi)-RNAs, which represent small noncoding RNA molecules, are attracting increasing interests as potential therapeutic targets. Thus far, they have been profiled in the human sclera and implicated in regulation of early (fetal) ocular growth.[36] Specifically, several collagen-specific miRNAs show increased expression in fetal eyes exhibiting active growth (enlargement). Drawing parallels between fetal and myopic eye growth—they both involve active growth, it seems reasonable to posit roles for these small molecules in the regulation of myopic eye growth, by modulating the expression levels of various scleral mRNAs (genes). Understanding the roles of miRNAs in myopia awaits characterization of scleral genome-wide expression profiles in a suitable animal model of myopia. Such studies will also offer a rich resource for identifying key gene signaling pathways and exploring specific molecular mechanisms, with the potential for identifying scleral gene targets for novel myopia therapies. Preliminary findings from scleral miRNA as well as mRNA profiling studies in the mouse myopia model indicate that

miRNAs of the let-7 family, previously implicated in matrix remodeling in other tissues, are upregulated in eyes exposed to form deprivation. Notable signaling pathways showing overrepresentation of genes include intermediate filament organization, scaffold protein binding, detection of stimuli, calcium ion, and G-protein pathways.[37]

5. POTENTIAL THERAPEUTIC APPROACHES

Thus far, research into drug therapies aimed at the sclera for myopia prevention or retardation is still nascent, and to-date no single molecule has been proven conclusively to act via a known/established scleral mechanism. Nonetheless, the sclera is listed among possible sites of action of the only two pharmacological treatments in current clinical use for myopia, topical atropine, which is widely used in East Asia, and oral 7-methylxanthine, which is approved for use in Denmark.[38–40] Other exploratory treatments targeting the sclera involve collagen cross-linking procedures, of which some are already being applied clinically to stabilize corneas, and biopolymers designed as tissue scaffolds to support new ECM production and/or alter scleral remodeling.[41,42]

The sclera is an attractive target tissue for myopia therapy for a number of reasons: as noted above, it defines the final size and shape of the eye, the changes in myopia have been relatively well characterized, and it is a relatively safe and accessible drug target. Understanding molecular mechanisms and identifying regulators of scleral remodeling are critical for devising strategies to maintain or improve scleral resistance to ocular elongation. At this time, there is still much to be explored and understood in this field, due to the complex nature of genome-wide gene regulatory networks and the as yet poorly understood role of genetic susceptibility in myopia (discussed elsewhere in this chapter) (Fig. 1). Moving forward with targeted scleral therapies will also benefit from complementary research into drug delivery systems compatible with local and sustained action.

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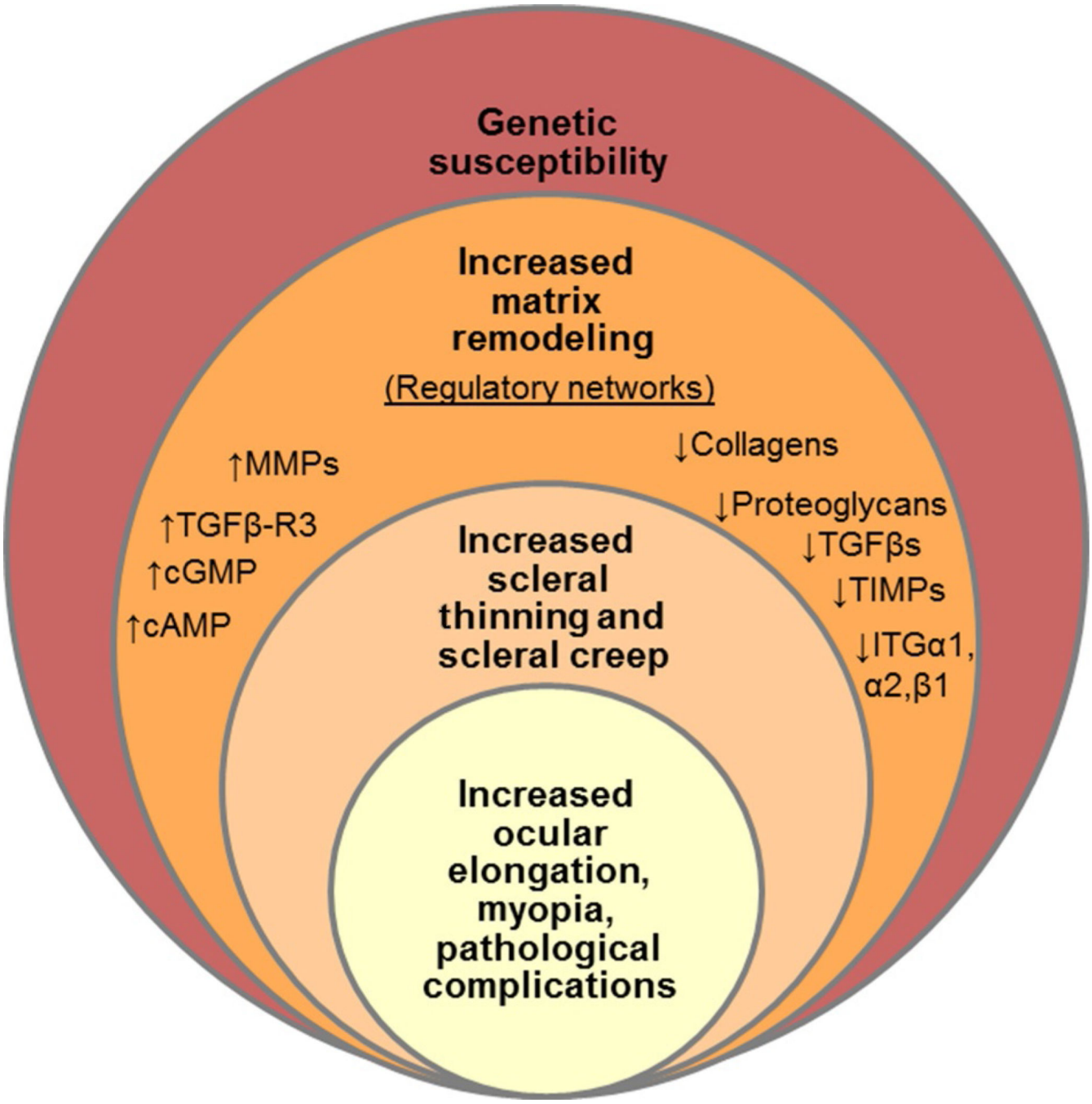

Figure 1.
Schematic of scleral remodeling mechanisms and implications in myopia.