• Review Article •

Scleral remodeling in myopia development

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Abstract

● With the increasing prevalence in recent years, myopia has become an essential global health concern. In most instances, an increased axial length of the eye is the structural cause of nearsightedness. The scleral remodeling, primarily dependent on the scleral extracellular matrix (ECM) changes, is significantly linked to eye lengthening. Scleral remodeling plays a critical function in the incidence and progression of myopia. This mini-review will focus on recent research progress of scleral remodeling in the hope of providing new ideas for the prophylaxis and treatment of myopia.

● **KEYWORDS:** scleral remodeling; myopia; extracellular matrix; hypoxia-inducible factor-1α; MMPs/TIMPs
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INTRODUCTION

Myopia is one of the most prevalent ophthalmic illness in the world[1]. It can not only cause vision loss, but also lead to severe complications and even blindness[2]. Based on evidence from epidemiology, the prevalence of myopia is increasing with each passing year, especially in Asian populations[3]. According to the prediction, in 2050, there will be 938 million people with high myopia (9.8% of the worldwide population) in the world[1]. Myopia has been considered to be a significant public health problem now.

Due to the excessive cornea or lens curvature and eye lengthening, images are focused in front of the retina in patients with myopia[4]. Although there are some measures to control the development of myopia, such as rigid gas permeable (RGP), atropine, outdoor activities and so on, the pathogenesis and cure of myopia remains ambiguous[5]. In recent years, research has focused on scleral remodeling in myopia development. It is considered that scleral remodeling plays an essential role in the incidence and progression of myopia. This mini-review will describe the research progress of the scleral remodeling so far.

ROLE OF SCLERAL REMODELING IN MYOPIA

An excessive increase in axial length is the significant structural change in myopia[6]. The sclera, especially at the posterior pole, is thinning in this process[7]. According to the mammalian models of high myopia, scleral remodelling, which depends on the changes in the constitution of the scleral extracellular matrix (ECM), plays a significant part in the thinness of the sclera[8-9]. Scleral collagen accumulation diminishes as myopia progresses, while breakdown rises[10]. Apart from scleral collagen changes, sclera proteoglycan formation is also decreased[11]. In consequence, scleral fibril assembly is disorganized, and the biomechanics of the sclera is getting weaker[12]. What is said above suggests that the explanations for changes in the prolongation of the eyes are scleral ECM remodeling.

RECENT STUDY ON SCLERAL REMODELING

The mechanism of scleral remodeling has not yet been fully explored. Researches mainly focus on the cytokines and signal transduction pathways related to the scleral remodeling.

Matrix Metalloproteinases and Tissue Inhibitors of Metalloproteinases Matrix metalloproteinases (MMPs) are a group of zinc-dependent endopeptidases involved in degrading various proteins, including collagen and elastin, in the ECM[13]. Therefore, the balance of MMPs activation and inhibition is the key to scleral remodeling. MMP-2 levels were elevated in high-myopia patients' aqueous humor, and tissue inhibitors of metalloproteinases (TIMP)-1, -2, and -3 levels were positively linked with MMP-2 levels and axial length[14-16]. In the inform deprived myopia study of tree shrews, active scleral MMP-2 levels were similarly higher in myopic eyes, and the up-regulation of MMP-2 levels causes scleral structure reorganization and ECM remodeling[17-18]. In tree shrew scleral fibroblasts, a low dose of recombinant TIMP-2 can stimulate MMP-2 activation in a dose-dependent manner, while a high dose of recombinant TIMP-2 can prevent MMP-2 activation.

In the circumstances, collagen degradation was significantly reduced, and axial lengths were significantly shortened[19]. Besides, in the animal models of chicks[20-21], guinea pigs[22], and mice[23] increases in MMP-2 and decreases in TIMP-2 activity also contribute to mediating scleral remodeling. Recent studies show that MMP-2 also participate in the formation of nearsightedness as a downstream molecule in some signal transduction pathways. Liu and Sun[24] demonstrated that the expressions of insulin-like growth factor-1 (IGF-1), signal transducers and activators of transcription (STAT3), and MMP-2 are increased progressively over time in the sclera in the guinea pig form-deprivation myopia model. The results reveal that through modulation of the expression of MMP-2, the IGF-1/STAT3 pathway in the sclera may play an essential role in sclera remodeling[24-25]. Chen *et al*[26] showed that by injecting Shh amino-terminal peptide (Shh-N) into the vitreous body, the level of MMP-2 and axial elongation were enhanced. The outcomes suggested that MMP-2 might be a downstream molecule of the sonic hedgehog signaling pathway (SHH). In conclusion, the balance between MMPs and TIMPs plays a key part in scleral remodeling.

Hypoxia-inducible Factor-1α Signaling Hypoxia-inducible factor-1α (HIF-1α) is a transcription factor in the hypoxia-inducible factors (HIF) family that reacts to declines in cellular oxygenation[27]. Wu *et al*[28] found that the hypoxia-signaling, the eukaryotic initiation factor 2 signaling (eIF2), and mammalian target of rapamycin signaling (mTOR) pathways were activated in the murine myopic sclera. In human scleral fibroblasts, hypoxia exposure contributes to myofibroblast trans differentiation by lowering type I collagen (COLI) levels. Reduced HIF-1α expression in guinea pigs, as well as eIF2α and mTOR levels, can inhibit experimental myopia development without impacting the growth of normal eyes. Meanwhile, their team verified that the HIF-1α signaling pathway is a main regulator of the Kyoto Encyclopedia of Genes and Genomes-protein protein interaction (KEGG-PPI) networks, which meant KEGG-PPI networks might be important in regulating interactions between gene and microenvironmental oxygen supply during the development of myopia[29]. Based on the above research, increased choroidal blood perfusion (ChBP) attenuates scleral hypoxia, and thereby inhibits myopia development in guinea pigs. Zhou *et al*[30] discovered that the antagonistic effect of peroxisome proliferators-activated receptors (PPARγ) reduces both choroidal thickness (ChT) and ChBP, nevertheless the expression of HIF-1α increases. As a result, scleral COL1 expression decreases lead to the development of myopia. PPARγ agonism, on the other hand, can prevent the increases in scleral HIF-1α expression levels, FD-induced ChT thinning, and ChBP decreases so that COL1 expression levels will not decline[31]. Further, in guinea pigs, scleral cAMP regulation mediated by the prostanoid receptor has an effect on myopia development *via* an interaction between PPARα and HIF-1α signaling[32]. According to the above, HIF-1α is a new target for scleral remodeling. There is still much work to be done.

GROWTH FACTOR

Transforming Growth Factor-β Transforming growth factor-β (TGF-β) family members are pluripotent cytokines that play a role in cell proliferation and differentiation, ECM remodeling, organ development, tissue repairment, and immune modulation[33]. TGF-β2 levels in high-myopia patients' aqueous humor, were shown to be higher in the eyes with excessive elongation of axial length and were positively linked to the MMP-2 levels[34-35]. Gentle *et al*[10] showed that TGF-β regulated scleral collagen synthesis and affected scleral remodeling in tree shrews. Reduced TGF-β led to a large drop in collagen synthesis in form-deprivation myopia (FDM) eyes *in vitro* experiments with sclera fibroblasts, indicating that TGF-β is a pivotal mediator to collagen loss[36]. TGF-β has also been linked to modifications in proteoglycans in sclera and has been discovered to influence glycosaminoglycans. Decreased TGF-β in FDM eyes resulted in reduced glycosaminoglycan synthesis[8]. *In vitro* experiment in guinea pig, the Wnt3/β-catenin signaling pathway was activated in scleral fibroblasts. TGF-β1 expression of COLI was blocked by this pathway which led to scleral remodeling in the development of myopia[37].

Bone Morphogenetic Protein The biggest subfamily of TGF-β is bone morphogenetic proteins (BMPs). In the guinea pig, a reduction of BMP-2 and BMP-5 levels during myopia induction is linked to sclera remodeling[38-39]. *In vitro* human scleral fibroblasts (HSF) experiment, increased BMP-2 resulted in increased expression of collagen I, collagen III, glycosaminoglycan, proteoglycan, and phosphorylated Smad1/5/8, which enhanced cell proliferation and raised the number of cells that differentiated into myofibroblasts[40].

Basic Fibroblast Growth Factor Basic fibroblast growth factor (b-FGF) is a fibroblast growth factor that regulates cell growth and apoptosis. The b-FGF level in the scleral tissue of lens-induced guinea pigs showed a general decline during the progression of myopia[41]. Tian *et al*[42] demonstrated that by increasing the expressions of COLI, α2 integrin, and β1 integrin, b-FGF might inhibit the occurrence and progression of defocus myopia.

LYSYL OXIDASE

The lysyl oxidase (LOX) family is an essential ECM enzyme. Through oxidizing lysine residues to aldehydes, LOX can stimulate the covalent crosslinking of collagen and elastin. Collagen crosslinking activity, which leads to collagen combining into insoluble collagen fibrils, is assisted by

LOX[43]. In the guinea pig, the expression of scleral COLI, formation of collagen fibril, and biomechanical properties were all reduced when LOX expression was inhibited. Adversely, what is said above also increased through upregulating LOX expression. These results suggest that modulating LOX expression in the sclera as a possible therapeutic option for myopia might be investigated[44].

RETINOIC ACID

Retinoic acid (RA) can modulate cell proliferation and differentiation in a variety of cells types. In addition, it can also influence ECM metabolism[45]. There is evidence to suggest that the visual modulation and scleral remodeling of the chick sclera are influenced by RA, which is considered a potent inhibitor of scleral glycosaminoglycan production[46]. In addition, the observed decrease in scleral galactosaminogalactan formation rates might be due to the rise in the rate of RA production in primates' eyes[47]. It has been reported that retinoic acid can up-regulate the Fibulin-1 level in cultured guinea pig and human sclera fibroblasts, and this effect is dose-dependent[48]. Fibulin-1 is associated with aggrecan. Aggrecan levels and distribution might manipulate the progression of scleral remodeling.

miRNAs EXPRESSION

The study of microRNAs (miRNAs) in scleral remodeling has gained popularity in recent years. Ravikanth suggested that microRNA expression was discovered in human sclera. Besides, in the fetal sclera, the expression of mir-214, let-7c, let-7e, mir-103, mir-107, and mir-98 was upregulated[49]. Chen *et al*[50] found that microRNA-328 may affect the progression of myopia by regulating the PAX6 gene, of which the effect is to decrease the expression of collagen I and integrin β1 while upregulating the level of MMP-2 in scleral cells. However, another research reported that even though the miR-328 expression was increased in the myopia group compared to the control group in high myopic eyes' aqueous humour, the difference between the two groups was not statistically significant[51]. MicroRNAs of the let-7 class were shown to be upregulated in eyes exposed to form deprivation in mouse[52]. Mei *et al*[53] screened out eight significantly upregulated miRNAs in FDM, including miR-294, miR-16-1, miR- 466h-5p, miR-466j, miR-15a, miR-466c-5p, miR-669e and miR-468. Zhang *et al*[54] demonstrated that in cells transfected with the miR-29a mimics, MMP-2 secretion by scleral fibroblasts and RPE cells was significantly reduced. miRNAs are expected to be a new drug to control the progress of myopia in the future.

ATROPINE

Atropine is a non-selective muscarinic antagonist that was considered beneficial in inhibiting myopia progression and decreasing axial length[55]. In the animal model of mice, atropine receptor blockage can regulate the expression of muscarinic receptor (mAChRs) which lead to the growth of scleral fibroblasts, therefore promoting scleral remodeling[56]. *In vitro* experiment, treatment with atropine attenuated the increase of regulator Of G protein signaling 2 (RGS2) expression and recovered the expression of COLI in FDM sclera[57]. Besides, Hsiao et al. used next-generation sequencing and bioinformatics approaches to find differentially expressed genes and microRNAs in atropine-treated scleral fibroblasts. They found that mechanisms which prevented melatonin breakdown during the night might play a part in decreasing scleral remodeling. In scleral fibroblasts, the interactions between miR-2682-5p-PRLR and miR-2682-5p-KNCJ5 provided a scientific foundation for assessing the involvement of low-dose atropine therapy[58].

CONCLUSIONS AND FUTURE DIRECTIONS

In conclusion, scleral remodeling plays an important role in the occurrence and development of myopia. This review focus on the key cytokines and signal pathway associated with scleral ECM remodeling and myopia development. It is hoped that it can contribute to the in-depth understanding of the pathogenesis of myopia and provide candidate intervention targets for the precise treatment of myopia. At present, the mechanisms of myopic scleral ECM remodeling are not precise yet. Therefore, further experimental studies on scleral ECM remodeling and new drug development should be conducted in the future.

METHODOLOGY

A literature search was conducted in PubMed from the date of inception until 10 March 2021 without language restrictions. The intention was to review recent advances with respect to scleral remodeling in myopia development. The search strategy was developed around the key terms: myopia, OR scleral, OR scleral remodeling, OR cytokines, OR signal transduction pathways, OR miRNAs, OR scleral ECM, OR ocular elongation. Only researches published in English were reviewed. Studies were excluded if they did not present a reasonable new or improved opinion for scleral modeling in myopia development.

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