Available online at www.sciencedirect.com

SciVerse ScienceDirect

Current Opinion in Pharmacology

Altering the way the optic nerve head responds to intraocular pressure—a potential approach to glaucoma therapy

Nicholas G Strouthidis[1,2] and Michael JA Girard[3]

Over the past decade, engineering principles have been used to explain why a mechanical load, intraocular pressure, can lead to the development of glaucomatous optic neuropathy. This has led to the 'biomechanical theory' of glaucoma, which posits that the behavior of optic nerve head connective tissues (specifically within the peripapillary sclera and lamina cribrosa) in response to intraocular pressure (regardless of its magnitude) can directly and indirectly influence the physiology and pathophysiology of the optic nerve head. Given that the biomechanics of the sclera and lamina cribrosa probably influence retinal ganglion cell loss in glaucoma, the idea that altering biomechanical behavior might be protective against glaucoma is an appealing notion. There is some evidence to suggest that stiffening the peripapillary sclera may be protective against the development of glaucoma in an animal model. It is technically possible to stiffen the sclera *in vivo* using collagen cross-linking techniques already applied *in vivo* to the cornea in the treatment of keratoconus. It has yet to be established whether scleral cross-linking is safe in humans and that it confers anything more than a theoretical advantage in terms of reducing the risk of glaucomatous damage.

Addresses
[1] NIHR Biomedical Research Centre at Moorfields Eye Hospital NHS Foundation Trust and UCL Institute of Ophthalmology, 162 City Road, London, EC1 V 2PD, UK
[2] Singapore Eye Research Institute, 11 Third Hospital Avenue, Singapore, 168751, Singapore
[3] Department of Bioengineering, National University of Singapore, 9 Engineering Drive 1, Block EA #03-12, Singapore 117576, Singapore

Corresponding author: Strouthidis, Nicholas G (nick.strouthidis@moorfields.nhs.uk)

Current Opinion in Pharmacology 2013, **13**:83–89

This review comes from a themed issue on **Neurosciences**

Edited by **Carlo Nucci**, **Nicholas G Strouthidis** and **Peng Tee Khaw**

For a complete overview see the Issue and the Editorial

Available online 19th September 2012

http://dx.doi.org/10.1016/j.coph.2012.09.001

Introduction

Although the definition of the glaucomas – a group of conditions in which there is a progressive optic neuropathy with development of associated characteristic visual field deficits – does not include elevated intraocular pressure (IOP), it is clear that elevated IOP is a key risk factor in the development of glaucoma. Furthermore, there is evidence to suggest that lowering of IOP may retard glaucomatous conversion in ocular hypertensives and glaucomatous progression in both high-tension and normal-tension forms of open angle glaucoma [1–3]. Given these observations, it is reasonable to infer that IOP, regardless of its magnitude, may influence the development of glaucomatous optic neuropathy.

How does IOP – either directly or indirectly – influence damage to the retinal ganglion cell (RGC) axons and therefore the development of glaucoma? There is much evidence to suggest that a key site of injury to the RGC is at the level of the lamina cribrosa (LC) within the optic nerve head (ONH) [4–7]. From a mechanical perspective, the ONH is essentially a 'weak spot' as it is a discontinuity within the otherwise robust corneo-scleral coat. The LC, and adjacent peripapillary sclera serve as the chief load-bearing connective tissue structures of the ONH, deriving their 'strength' from the constituent collagen. The LC is a complex three-dimensional structure comprising of a series of interconnected connective tissue beams and spaces, bridging the 'gap' within the termination of the peripapillary sclera. The 'pores' or spaces within the LC architecture allow the passage of RGC axon bundles as they leave the globe to continue within the retrobulbar optic nerve. It is within the passage through the LC that RGC axons are presumed to be vulnerable to IOP-related insult in glaucoma – perhaps transmitted through direct damage to the supporting connective tissues with interruption of axoplasmic flow and disruption of nutrient supply to the axons.

Within the last 15 years, the principles of biomechanics have been applied to the study of the relationship between the ONH and IOP [8–10]. Biomechanics refers to the application of mechanical principles to biological systems. The ONH may be treated as a biomechanical structure, with a complex 3D load-bearing connective tissue architecture (the LC) subjected to stress (a measure of internal force per unit area) induced by IOP. By studying the response of ONH connective tissues to IOP, we may elucidate: (1) why certain eyes are predisposed to developing glaucomatous optic neuropathy with high IOP and others are not; (2) why certain eyes develop glaucoma with a statistically normal IOP and (3) how ageing may influence the development of glaucoma through alterations in connective tissue biomechanical behavior.

In this review, the biomechanical paradigm of glaucoma will be explained as well as the key developments over the

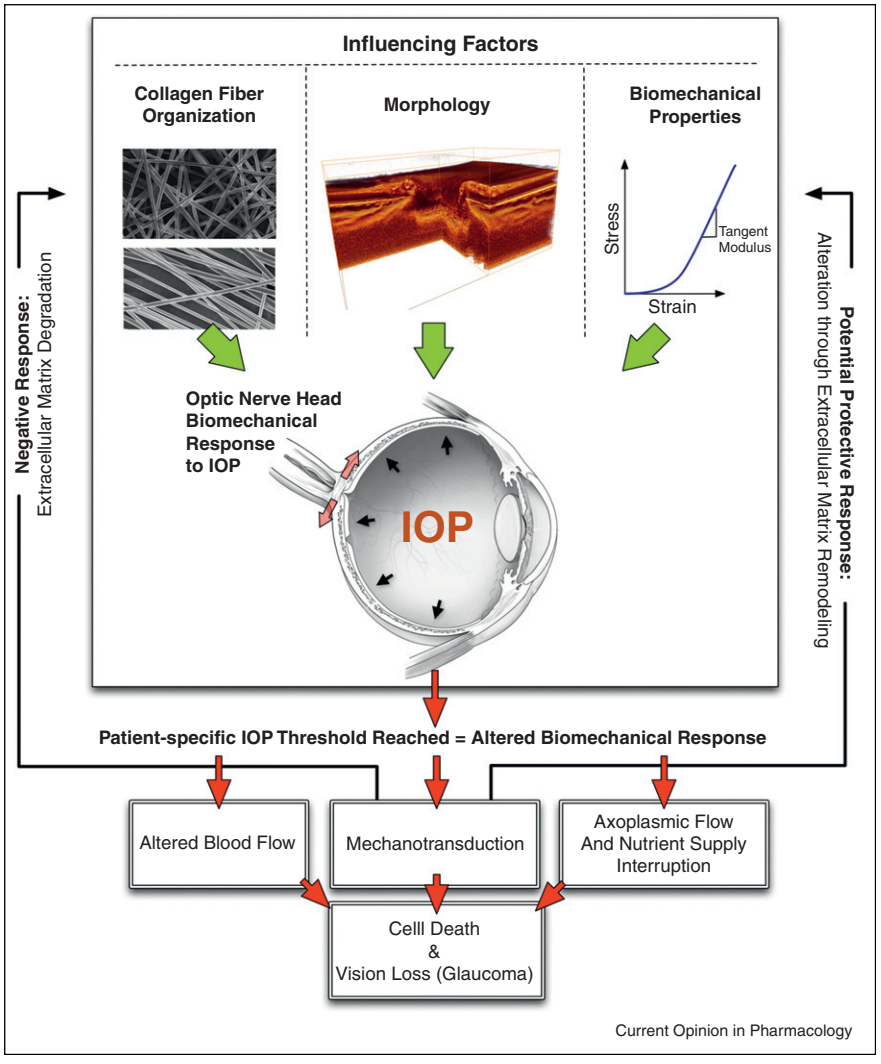
past 5 years that have increased our understanding of how the connective tissues of the ONH respond to biomechanical insult. We will speculate upon methods that may be used to alter the biomechanical behavior of the ONH in order to prevent or slow the development of glaucoma.

The biomechanical theory of glaucoma

The biomechanical theory of glaucoma proposes that ONH biomechanics may explain how IOP-induced stress and strain (a measure of tissue deformation) of the load bearing tissues of the ONH (sclera and LC) influence their physiology and pathophysiology, and of the adjunctive tissues (astrocytes, glia, endothelial cells, vascular pericytes and their basement membranes) and the RGC axons [8,11,12]. ONH biomechanics is the 'link' by which IOP can influence such apparently non-IOP factors such as ischemia, inflammation, autoimmunity, astrocytic and glial cell biology. The interplay between IOP, biomechanics and retinal ganglion cell loss is outlined in schematic form in Figure 1.

Multiple biomechanical factors are likely to influence an individual ONH's susceptibility to glaucoma, including, but not limited to: (1) the magnitude and variation of IOP; (2) collagen fiber orientation in the peripapillary sclera – a highly-aligned collagen fiber ring may protect the ONH against IOP-related insult [13,14]; (3) LC and scleral stiffness – a stiff sclera may shield the ONH from IOP-related insult; 4) LC and scleral geometry – a thick lamina may protect against mechanical damage [15•]. IOP-induced deformations of connective tissues, and thus biomechanics, can also affect the volume flow and perfusion pressure of blood within the laminar capillaries, which will in turn influence the diffusion of nutrients to

Figure 1



Schemata describing the mechanisms by which optic nerve head biomechanics may result in retinal ganglion cell loss in glaucoma. 'Mechanotransduction' refers to the conversion of mechanical stimuli to chemical signals at a cellular level.

astrocytes. Finally there may be features within the RGC itself that will result in an increased susceptibility to apoptosis in response to localised distress [16].

Measuring the anatomical response to acute changes in IOP

A first essential process in the study of ONH biomechanics has been to examine the response of ONH tissues in response to manipulations in IOP. Several investigators have studied the 'compliance' (the inverse of stiffness) of the normal ONH in response to an acute elevation of IOP using techniques of increasing complexity. These include: X-ray photography of cadaveric non-human primate eyes with fine platinum wires inserted into the peripapillary sclera and optic disc [17], laser doppler velocimetry of normal human autopsy eyes [18], conventional histology of human eyes [19], 2D [20,21], and 3D histomorphometric reconstructions of post mortem normal monkey eyes [22]. Various imaging modalities have been used *in vivo* in normal monkey eyes [23,24]. Numerical models have also been used [25,26]. These reports were all consistent (to a greater or lesser degree) in finding a posterior movement of the optic disc surface in response to an acute elevation of IOP.

Post mortem studies, performed both in human and monkey eyes, have indicated that the LC and scleral canal wall deform after acute IOP elevation [19–21,27]. The effect of acute IOP elevation upon monkey ONH connective tissues was initially explored using 2D histomorphometry by comparing a series of eyes perfusion fixed at IOP 10 mmHg with eyes that had been immersion fixed at IOP 0 mmHg [21]. In that study, the lamina was found to be thinner and more anterior, and the scleral canal diameter larger in the IOP 10 mmHg eyes. These observations suggested that, with acute IOP elevations at a low native IOP, the scleral canal expands resulting in a tauter, thinner, more anteriorly placed lamina. Subsequently, 2D histomorphometry was performed in normal young adult monkeys perfusion fixed at 10 mmHg in one eye and 30 or 45 mmHg in the fellow eye [20]. In that study, a posterior laminar deformation of 10–23 μm in the high IOP eyes compared to their contralateral fellow eyes was demonstrated. Most recently, ONH connective tissue deformation has been characterised in 3D histomorphometric ONH reconstructions of eyes perfusion fixed after acute IOP elevation to 30 or 45 mmHg compared to 10 mmHg in their fellow eyes [22]. Minimal to modest regional laminar thinning and posterior bowing of the peripapillary sclera, thinning and expansion of the scleral canal was observed in most of the high IOP eyes. The minimal posterior laminar displacement in response to acute IOP elevation was also noted in a series of *in vivo* spectral domain OCT imaging studies conducted both in monkeys and in humans, although scleral canal expansion was not measured [28,29]. Modeling has demonstrated that the LC is likely to be subject to significant increases in stress and strain, even in the absence of posterior deformation, because of the transfer of tensile stretch caused by the expansion of the scleral canal [26,30,31].

The effect of chronic IOP in the development of glaucomatous optic neuropathy

Given that only minimal posterior laminar displacement has mostly been observed following acute IOP elevations, it is therefore likely that the characteristic 'cupping' identified in glaucomatous optic neuropathy follows on from connective tissue and extracellular matrix remodeling in response to a chronic IOP insult (whether elevated or within normal range). In 3D histomorphometric studies of monkey eyes with early onset experimental glaucoma, thickening and posterior bowing of the LC was detected [32–34]. Furthermore, the proportion of horizontally orientated laminar beams was found to increase, leading to a suggestion that retrolaminar septal beams become 'recruited' into the connective tissue LC in response to chronic IOP elevation [32]. An alternative or additional hypothesis, supported by findings in normal human eyes [35], is that there is gradual posterior migration of the laminar insertion such that it inserts into the pia. It is expected that the astrocytes and lamina cribrosa cells mediate the extracellular remodeling of the LC in response to chronic IOP [36].

The role of sclera

The peripapillary sclera appears to exert a large influence over the biomechanics of the ONH; as stated earlier, the circumferential IOP-induced 'hoop stress' is transferred to the LC via the sclera and scleral canal expansion will drive some of the strains experienced within the LC. *In vivo* studies conducted in normal subjects and subjects with glaucoma have estimated that ocular rigidity (a crude measure of scleral stiffness) increases with the development of glaucoma [37]. These findings support *ex vivo* work conducted in the monkey eye that suggests that scleral stiffness increases with both ageing and with moderate elevations in IOP, although the latter may be preceded by a period of scleral hypercompliance [38,39••]. A recent inflation study of enucleated normal and glaucomatous human eyes found that glaucomatous eyes had a stiffer meridional strain response in the peripapillary region than in normal eyes, although it is unclear whether this characteristic represents a consequence of, or a predisposition towards, the development of glaucoma [40•]. It is possible that the sclera may stiffen and does so as a protective mechanism against an increase in IOP through mechanotransduction (the cellular 'conversion' of mechanical stumuli into biochemical responses) of scleral fibroblasts and extracellular matrix remodeling. If this were the case, such a response could act to slow glaucomatous progression in eyes with higher IOPs and/or more compliant scleral shells at baseline.

There is, in fact, evidence to suggest that a stiffer sclera may be protective against the development of glaucoma, at least in the mouse eye [41••]. Mice with a mutation to collagen 8 (Aca23 mice), which have longer and wider eyes than wild type mice, were found to develop proportionally less globe enlargement and significantly less RGC loss than wild type mice with the onset of experimentally induced glaucoma. It is tempting, if not unreasonable, to assume that the difference in susceptibility to experimental glaucoma may be due to alterations in scleral biomechanical behavior.

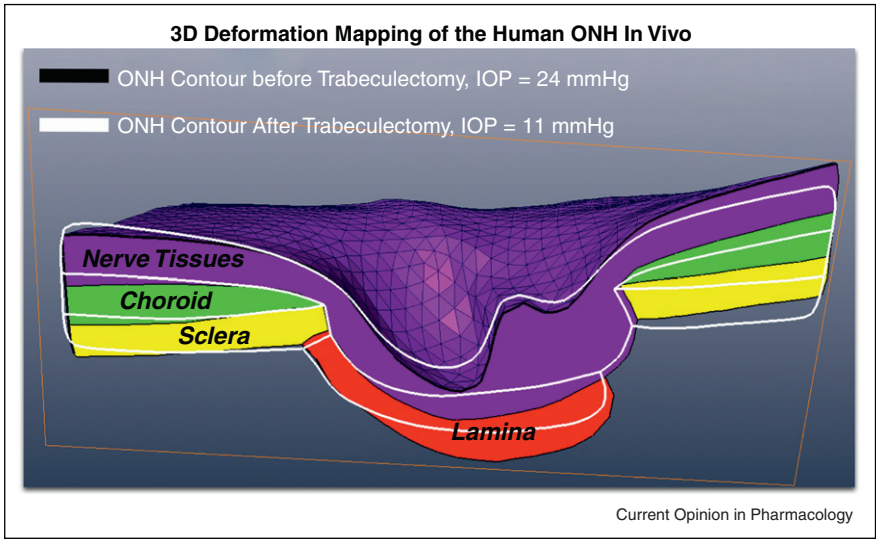
Monkey eyes with initially stiff or thick sclera have been shown to be less prone to biomechanical changes in response to chronic IOP elevation [39••], which suggests that stiff eyes have a lower sensitivity to IOP. The notion that a stiff scleral shell may be an advantage for patients with glaucoma appears to be paradoxical when one considers that the sclera stiffens with age [38] and susceptibility to glaucoma increases with age [42]. To reconcile these contradictory observations, it should be noted that collagen fibers become more brittle with age [43]; it is therefore possible that scleral stiffening with age is a 'suboptimal' mechanism resulting in a biomechanically unstable ONH.

Altering biomechanical behavior as potential therapy?

Having posited that the biomechanical behavior of ONH connective tissues influences the pathophysiology of RGC loss in glaucoma, a natural extension might be to consider whether manipulating ONH biomechanics might serve as a potential therapeutic avenue in glaucoma. It is certainly technically possible to 'stiffen' the sclera and this has been mooted as a potential treatment for progressive myopia. This has been achieved *in vivo* in human subjects by juxtascleral injection of a liquid polymer 'strengthening' agent and by 'scleroplastic' surgery whereby donor scleral strips or other biomaterials are sutured to the sclera to increase its tensile strength [44]. Although the proof of concept (in other words, some reduction in the rate of axial lengthening) has been demonstrated using posterior polar scleral buckling techniques [45], high complication rates have also been reported with other scleroplastic surgical techniques [46]. As such these techniques have not yet been widely adopted for the treatment of progressive myopia.

In the past decade, corneal cross-linking has been introduced as an *in vivo* method that stiffens the cornea and retards the progression of keratoconus [47]. This is achieved by photo-polymerisation to induce increased cross-linking between collagen fibrils leading to a stiffer fiber network. In clinical practice, topical riboflavin is used as the photosensitizing agent, with UVA illumination acting as the photo-stimulus. A number of investigators have demonstrated that the same cross-linking technique may increase scleral stiffness both *ex vivo* in human and porcine eyes [48,49] and *in vivo* using rabbit eyes [50], with a sustained effect for up to 8 months in the latter. Unfortunately, this technique was also found to be highly cytotoxic to the outer retina of the rabbit eye and this was presumably related to the dosing of UVA [50]. An alternative *in vivo* chemical cross-linking method using glyceraldehyde was found to have a similar biomechanical effect on rabbit sclera, although without any observed retinal toxicity [51]. There is, however, some evidence to suggest that increasing scleral cross-linking may result in

Figure 2



3D OCT images acquired *in vivo* in a glaucoma subject before and after IOP lowering by trabeculectomy. The 3D displacement in the images in response to the change in IOP may be used to extract tissue strains in the imaged regions of the optic nerve head.

greater scleral permeability that theoretically could lead to unwanted sequelae [52]. Scleral cross-linking has not yet been applied to an animal model of glaucoma.

It may be that a 'safe' technique that stiffens the sclera might protect against glaucoma by limiting IOP-induced strain in the ONH tissues, as predicted by computational models [30,31]. However, at the time of writing, one cannot yet be certain whether stiffening of the sclera (or indeed the LC) will be protective in glaucoma; it is possible that in some individuals the effect may be harmful and in effect an increasing of ONH compliance may be therapeutically advantageous. Our understanding of the biomechanical characteristics that predispose clinically to the development of glaucoma (and perhaps might be modified in a therapeutic approach) will improve as *in vivo* biomechanical testing of the ONH begins to become possible using high-resolution spectral domain OCT technology (Figure 2) to capture IOP-induced deformations of the sclera and LC.

Conclusions

The biomechanical behavior of the connective tissues of the optic nerve head – specifically the lamina cribrosa and peripapillary sclera – probably influences the physiology and pathophysiology of the optic nerve head. The biomechanical theory of glaucoma may help to explain how certain eyes are predisposed to the development of glaucomatous optic neuropathy either at high IOP or at normal levels of IOP, and some eyes are not. Furthermore, age-related changes in biomechanical behavior may help to explain the increasing predisposition to glaucoma with ageing. Given the likely role of scleral and lamina cribrosa biomechanics, an ability to manipulate their biomechanical behavior may serve as a potential therapeutic target. It is already technically possible to strengthen and stiffen the sclera both *ex vivo* and *in vivo* in animals using collagen cross-linking techniques. Whilst these undoubtedly have the potential to stiffen the sclera, these techniques are not yet known to be safe and there is no proof that they will be of benefit in glaucoma. It is possible that scleral stiffening may not be advantageous in all individuals with, or at risk of, glaucoma. Our understanding of optic nerve head biomechanics *in vivo* will need to increase so that we can appreciate what characteristics predispose to glaucomatous damage; only then can we begin to design appropriate biomechanical modification therapies.

Conflict of interest

None.

Acknowledgements

NGS is supported by the National Institute for Health Research (NIHR) Biomedical Research Centre based at Moorfields Eye Hospital NHS Foundation Trust and UCL Institute of Ophthalmology. The views expressed are those of the author(s) and not necessarily those of the NHS, the NIHR or the Department of Health.

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. Heijl A, Leske MC, Bengtsson B, Hyman L, Hussein M: **Reduction of intraocular pressure and glaucoma progression: results from the Early Manifest Glaucoma Trial**. *Arch Ophthalmol* 2002, **120**:1268-1279.

2. AGIS Investigators: **The Advanced Glaucoma Intervention Study (AGIS): 7. The relationship between control of intraocular pressure and visual field deterioration. The AGIS Investigators**. *Am J Ophthalmol* 2000, **130**:429-440.

3. Kass MA, Heuer DK, Higginbotham EJ, Johnson CA, Keltner JL, Miller JP, Parrish RK, Wilson MR, Gordon MO: **The Ocular Hypertension Treatment Study: a randomized trial determines that topical ocular hypotensive medication delays or prevents the onset of primary open-angle glaucoma**. *Arch Ophthalmol* 2002, **120**:701-713.

4. Anderson DR, Hendrickson A: **Effect of intraocular pressure on rapid axoplasmic transport in monkey optic nerve**. *Invest Ophthalmol Vis Sci* 1974, **13**:771-783.

5. Quigley H, Anderson DR: **The dynamics and location of axonal transport blockade by acute intraocular pressure elevation in primate optic nerve**. *Invest Ophthalmol Vis Sci* 1976, **15**:606-616.

6. Quigley HA, Addicks EM, Green WR, Maumenee AE: **Optic nerve damage in human glaucoma. II. The site of injury and susceptibility to damage**. *Arch Ophthalmol* 1981, **99**:635-649.

7. Minckler DS, Bunt AH, Johanson GW: **Orthograde and retrograde axoplasmic transport during acute ocular hypertension in the monkey**. *Invest Ophthalmol Vis Sci* 1977, **16**:426-441.

8. Burgoyne CF, Downs JC, Bellezza AJ, Suh JK, Hart RT: **The optic nerve head as a biomechanical structure: a new paradigm for understanding the role of IOP-related stress and strain in the pathophysiology of glaucomatous optic nerve head damage**. *Prog Retin Eye Res* 2005, **24**:39-73.

9. Sigal IA, Ethier CR: **Biomechanics of the optic nerve head**. *Exp Eye Res* 2009, **88**:799-807.

10. Downs JC, Roberts MD, Burgoyne CF: **Mechanical environment of the optic nerve head in glaucoma**. *Optom Vis Sci* 2008, **85**:425-435.

11. Fechtner RD, Weinreb RN: **Mechanisms of optic nerve damage in primary open angle glaucoma**. *Surv Ophthalmol* 1994, **39**:23-42.

12. Burgoyne CF, Morrison JC: **The anatomy and pathophysiology of the optic nerve head in glaucoma**. *J Glaucoma* 2001, **10**:S16-S18.

13. Girard MJ, Dahlmann-Noor A, Rayapureddi S, Bechara JA, Bertin BM, Jones H, Albon J, Khaw PT, Ethier CR: **Quantitative mapping of scleral fiber orientation in normal rat eyes**. *Invest Ophthalmol Vis Sci* 2011, **52**:9684-9693.

14. Pijanka JK, Coudrillier B, Ziegler K, Sorensen T, Meek KM, Nguyen TD, Quigley HA, Boote C: **Quantitative mapping of collagen fiber orientation in non-glaucoma and glaucoma posterior human sclerae**. *Invest Ophthalmol Vis Sci* 2012, **53**:5258-5270.

15. • Grytz R, Sigal IA, Ruberti JW, Meschke G, Downs JC: **Lamina cribrosa thickening in early glaucoma predicted by a microstructure motivated growth and remodeling approach**. *Mech Mater* 2012, **44**:99-109.

In this study, a numerical model of growth and remodeling is used to demonstrate that the lamina cribrosa will thicken by increase in collagen fibril mass with the onset of early glaucoma. Furthermore, the growth and remodeling algorithm can also demonstrate the physiological need for a connective tissue lamina cribrosa in order to maintain biomechanical homeostasis at normal levels of IOP in the human eye.

16. Burgoyne CF, Downs JC: **Premise and prediction – how optic nerve head biomechanics underlies the susceptibility and clinical behavior of the aged optic nerve head**. *J Glaucoma* 2008, **17**:318-328.

17. Levy NS, Crapps EE: **Displacement of optic nerve head in response to short-term intraocular pressure elevation in human eyes**. *Arch Ophthalmol* 1984, **102**:782-786.

18. Zeimer RC, Ogura Y: **The relation between glaucomatous damage and optic nerve head mechanical compliance**. *Arch Ophthalmol* 1989, **107**:1232-1234.

19. Yan DB, Coloma FM, Metheetrairut A, Trope GE, Heathcote JG, Ethier CR: **Deformation of the lamina cribrosa by elevated intraocular pressure**. *Br J Ophthalmol* 1994, **78**:643-648.

20. Bellezza AJ, Rintalan CJ, Thompson HW, Downs JC, Hart RT, Burgoyne CF: **Deformation of the lamina cribrosa and anterior scleral canal wall in early experimental glaucoma**. *Invest Ophthalmol Vis Sci* 2003, **44**:623-637.

21. Bellezza AJ, Rintalan CJ, Thompson HW, Downs JC, Hart RT, Burgoyne CF: **Anterior scleral canal geometry in pressurised (IOP 10) and non-pressurised (IOP 0) normal monkey eyes**. *Br J Ophthalmol* 2003, **87**:1284–1290.

22. Yang H, Downs JC, Sigal IA, Roberts MD, Thompson H, Burgoyne CF: **Deformation of the normal monkey optic nerve head connective tissue after acute IOP elevation within 3-D histomorphometric reconstructions**. *Invest Ophthalmol Vis Sci* 2009, **50**:5785-5799.

23. Coleman AL, Quigley HA, Vitale S, Dunkelberger G: **Displacement of the optic nerve head by acute changes in intraocular pressure in monkey eyes**. *Ophthalmology* 1991, **98**:35-40.

24. Quigley HA, Pease ME: **Change in the optic disc and nerve fiber layer estimated with the glaucoma-scope in monkey eyes**. *J Glaucoma* 1996, **5**:106-116.

25. Sigal IA, Yang H, Roberts MD, Burgoyne CF, Downs JC: **IOP-induced lamina cribrosa displacement and scleral canal expansion: an analysis of factor interactions using parameterized eye-specific models**. *Invest Ophthalmol Vis Sci* 2011, **52**:1896-1907.

26. Roberts MD, Liang Y, Sigal IA, Grimm J, Reynaud J, Bellezza A, Burgoyne CF, Downs JC: **Correlation between local stress and strain and lamina cribrosa connective tissue volume fraction in normal monkey eyes**. *Invest Ophthalmol Vis Sci* 2010, **51**:295-307.

27. Albon J, Purslow PP, Karwatowski WS, Easty DL: **Age related compliance of the lamina cribrosa in human eyes**. *Br J Ophthalmol* 2000, **84**:318-323.

28. Agoumi Y, Sharpe GP, Hutchison DM, Nicolela MT, Artes PH, Chauhan BC: **Laminar and prelaminar tissue displacement during intraocular pressure elevation in glaucoma patients and healthy controls**. *Ophthalmology* 2011, **118**:52-59.

29. Strouthidis NG, Fortune B, Yang H, Sigal IA, Burgoyne CF: **Effect of acute intraocular pressure elevation on the monkey optic nerve head as detected by spectral domain optical coherence tomography**. *Invest Ophthalmol Vis Sci* 2011, **52**:9431-9437.

30. Sigal IA, Flanagan JG, Tertinegg I, Ethier CR: **Modeling individual-specific human optic nerve head biomechanics. Part I: IOP-induced deformations and influence of geometry**. *Biomech Model Mechanobiol* 2009, **8**:85-98.

31. Sigal IA, Flanagan JG, Tertinegg I, Ethier CR: **Modeling individual-specific human optic nerve head biomechanics. Part II: influence of material properties**. *Biomech Model Mechanobiol* 2009, **8**:99-109.

32. Roberts MD, Grau V, Grimm J, Reynaud J, Bellezza AJ, Burgoyne CF, Downs JC: **Remodeling of the connective tissue microarchitecture of the lamina cribrosa in early experimental glaucoma**. *Invest Ophthalmol Vis Sci* 2009, **50**:681-690.

33. Yang H, Downs JC, Bellezza AJ, Thompson H, Burgoyne CF: **3-D histomorphometry of the normal and early glaucomatous monkey optic nerve head: prelaminar neural tissues and cupping**. *Invest Ophthalmol Vis Sci* 2007, **48**:5068-5084.

34. Yang H, Downs JC, Girkin C, Sakata L, Bellezza A, Thompson H, Burgoyne CF: **3-D histomorphometry of the normal and early glaucomatous monkey optic nerve head: lamina cribrosa and peripapillary scleral position and thickness**. *Invest Ophthalmol Vis Sci* 2007, **48**:4597-4607.

35. Sigal IA, Flanagan JG, Tertinegg I, Ethier CR: **3D morphometry of the human optic nerve head**. *Exp Eye Res* 2010, **90**:70-80.

36. Hernandez MR: **The optic nerve head in glaucoma: role of astrocytes in tissue remodeling**. *Prog Retin Eye Res* 2000, **19**:297-321.

37. Hommer A, Fuchsjager-Mayrl G, Resch H, Vass C, Garhofer G, Schmetterer L: **Estimation of ocular rigidity based on measurement of pulse amplitude using pneumotonometry and fundus pulse using laser interferometry in glaucoma**. *Invest Ophthalmol Vis Sci* 2008, **49**:4046-4050.

38. Girard MJ, Suh JK, Bottlang M, Burgoyne CF, Downs JC: **Scleral biomechanics in the aging monkey eye**. *Invest Ophthalmol Vis Sci* 2009, **50**:5226-5237.

39. Girard MJ, Suh JK, Bottlang M, Burgoyne CF, Downs JC:
•• **Biomechanical changes in the sclera of monkey eyes exposed to chronic IOP elevations**. *Invest Ophthalmol Vis Sci* 2011, **52**:5656-5669.

The scleral biomechanics of eight pairs of monkey eyes with unilateral chronic IOP elevation by pressurising the posterior scleral shells and measuring 3D displacements by speckle interferometry. Finite element models incorporating stretch-induced stiffening and multidirectionality of collagen fibrils were then matched to the experimentally measured scleral displacements in order to extract scleral biomechanic properties. The posterior scleral shells had inhomogenous, anisotropic and non-linear biomechanical properties. In most cases sclera stiffened with moderate levels of chronic IOP elevation although this may be preceded by scleral hypercompliance.

40. Coudrillier B, Tian J, Alexander S, Myers KM, Quigley HA,
• Nguyen TD: **Biomechanics of the human posterior sclera: age- and glaucoma-related changes measured using inflation testing**. *Invest Ophthalmol Vis Sci* 2012, **53**:1714-1728.

In this study the biomechanical response of human posterior sclera was tested *in vitro* by pressure inflating cadaveric normal and glaucoma eyes and computing the strain response from digital image correlation displacements. Sclera was found to be stiffer in older eyes and glaucomatous eyes were stiffer in the meridional orientation in the peripapillary sclera.

41. Steinhart MR, Cone FE, Nguyen C, Nguyen TD, Pease ME, Puk O,
•• Graw J, Oglesby EN, Quigley HA: **Mice with an induced mutation in collagen 8A2 develop larger eyes and are resistant to retinal ganglion cell damage in an experimental glaucoma model**. *Mol Vis* 2012, **18**:1093-1106.

This study tested the hypothesis that eyes with a collagen mutation (in collagen 8, Aca23 mouse) that result in a larger globe than wild type eyes would have an altered biomechanical response to raised intraocular pressure. By inducing an experimental glaucoma model using microbead injection in both wild type and Aca23 mice, the investigators were able to demonstrate proportionally less ocular expansion in the Aca23 eyes as well as less retinal ganglion cell loss. This represents strong evidence, at least in the mouse eye, that altering scleral biomechanics may be protective against glaucoma.

42. Friedman DS, Wolfs RC, O'Colmain BJ, Klein BE, Taylor HR, West S, Leske MC, Mitchell P, Congdon N, Kempen J: **Prevalence of open-angle glaucoma among adults in the United States**. *Arch Ophthalmol* 2004, **122**:532-538.

43. Bailey AJ, Paul RG, Knott L: **Mechanisms of maturation and ageing of collagen**. *Mech Ageing Dev* 1998, **106**:1-56.

44. Avetisov ES, Tarutta EP, Iomdina EN, Vinetskaya MI, Andreyeva LD: **Nonsurgical and surgical methods of sclera reinforcement in progressive myopia**. *Acta Ophthalmol Scand* 1997, **75**:618-623.

45. Ward B, Tarutta EP, Mayer MJ: **The efficacy and safety of posterior pole buckles in the control of progressive high myopia**. *Eye (Lond)* 2009, **23**:2169-2174.

46. Curtin BJ, Whitmore WG: **Long-term results of scleral reinforcement surgery**. *Am J Ophthalmol* 1987, **103**:544-548.

47. Goldich Y, Marcovich AL, Barkana Y, Mandel Y, Hirsh A, Morad Y, Avni I, Zadok D: **Clinical and corneal biomechanical changes**

after collagen cross-linking with riboflavin and UV irradiation in patients with progressive keratoconus: results after 2 years of follow-up. *Cornea* 2012, **31**:609-614.

48. Thornton IL, Dupps WJ, Roy AS, Krueger RR: **Biomechanical effects of intraocular pressure elevation on optic nerve/lamina cribrosa before and after peripapillary scleral collagen cross-linking**. *Invest Ophthalmol Vis Sci* 2009, **50**:1227-1233.

49. Wollensak G, Spoerl E: **Collagen crosslinking of human and porcine sclera**. *J Cataract Refract Surg* 2004, **30**:689-695.

50. Wollensak G, Iomdina E, Dittert DD, Salamatina O, Stoltenburg G: **Cross-linking of scleral collagen in the rabbit using riboflavin and UVA**. *Acta Ophthalmol Scand* 2005, **83**:477-482.

51. Wollensak G, Iomdina E: **Long-term biomechanical properties after collagen crosslinking of sclera using glyceraldehyde**. *Acta Ophthalmol* 2008, **86**:887-893.

52. Stewart JM, Schultz DS, Lee OT, Trinidad ML: **Exogenous collagen cross-linking reduces scleral permeability: modeling the effects of age-related cross-link accumulation**. *Invest Ophthalmol Vis Sci* 2009, **50**:352-357.