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Polarimetric interferometry to objectively evaluate the optical properties of corneal stroma

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Abstract

A new non-invasive method, based on the interferometric analysis of diffractive and polarizing effects related to the birefringent properties of corneal collagen fibrils, has been developed to objectively evaluate the optical properties of the stroma. The new method shows a relevant impact on corneal surgeries specifically for lamellar transplantation where, due to the polarizing properties of the stroma, the alignment between collagen fibrils of donor corneas with patient collagen fibril orientation has shown an improvement of visual acuity postoperatively. Further studies on the regularity of the corneal isogyre pattern are showing this new method has a strong impact in early-stage diagnosis of corneal disease.

Keywords: corneal cross, isogyre, melatope, birefringence

1. Introduction

The cornea has a dual function, optical and mechanical. The human cornea provides two-thirds of the refractive power of the eye and the requisite durability to maintain its shape, notwithstanding the action of the extraocular muscles and the internal force of the ciliary muscle. The latter mediates the change in lens shape in order

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to adjust the focusing power of the eye, and has been shown to have a very small and optically negligible effect on corneal shape.[1] A fundamental characteristic for corneal function is that of transparency. The cornea is classically accepted as a five-layered structure: an epithelial and an endothelial layer at the anterior and posterior surfaces respectively; two membranes: Bowman's under the epithelium and Descemet's anterior to the endothelium; and a central and predominant stroma composed of collagen fibrils, which represent approximately 90% of corneal thickness.

The stroma is composed of approximately two successively stacked lamellae of type I collagen fibrils with a diameter of around 25-30 µm.[2] Within each lamella, the collagen fibrils run parallel to each other and show a regular interfibrillar spacing.[2-5] The orientation of the fibrils is constant within each lamella, but varies throughout successive layers.[6-9] The regular arrangement of the fibrils within each layer is considered to be responsible for the transparency of the tissue.[2,10,11] It is also an important factor for determining the mechanical properties of the cornea[12,13] as well as maintaining its shape.[14,15]

Such an organization contributes to characteristic patterns that are linked to highly ordered structures, such as crystals, which possess the property of birefringence.

These characteristic patterns seen when the structure is illuminated between crossed polarizers are a dark cross, the arms of which have been called isogyres, and colored rings or isochromatics. Whilst these formations, which are caused by the separation of refracted rays into ordinary and extraordinary components that travel at different velocities through the crystal depending on the atomic spacing in the plane of travel, can be explained in relation to crystal structure,[16] similar features have been noted over 200 years ago in eye lenses.[17-18] More recently, they have been noted in the cornea.[7,8,19-24] This cannot be explained on the same basis as crystals, given that ocular tissue is not highly ordered, but rather accords with the descriptions for elongated structures that arise in biology, such as cell layer arrangements.[25,26] The lamellar organization of the corneal stroma is akin to a Wiener body,[27] which gives rise to form birefringence due to the directional variations in refractive index: *i.e.*, differences along single fibers and across fiber layers.[27] It has also been shown that isogyres can be formed in curved structures made of amorphous materials that do not possess any birefringent properties.[21,28] The number of studies that have considered corneal birefringence show differences in findings,[19-24] and indeed it has been suggested that the random orientation of the central layers of the stroma results in no birefringence effects at the corneal apex.[8] Such variation is to be expected in a biological tissue that differs in shape and thickness between individuals and changes with age and pathologies. There has been a paucity of investigation into the use of polarization optics for the study of stromal structure. Stromal orientation is important for optical quality and differences between individuals may suggest that the particular orientation of lamellae is optimized

for image quality. In cases of corneal transplantation, the orientation of the donor cornea should therefore be considered. This study presents results of measurements on *in-vivo* corneal tissue using a new device that can determine the polarization properties of the cornea *in vitro* or *in vivo*. The instrument applies specialized software to determine the orientation of the corneal fibrils, offering the prospect of a detailed structural analysis which may have applications for clinical studies.

2. Scope

Development of a new non-invasive method to objectively evaluate the optical properties of the stroma, based on the interferometric analysis of diffractive and polarizing effects related to the birefringent properties of the stroma.

3. Method

Comparison between polarimetric interferometry image obtained by the interference between polarized light and stromal structure of human corneas using a new patented[29] medical device called Lumaxis® (Phronema SRL; Bari, Italy) with data published in the literature and obtained by x-ray, and second and third harmonic generation technique (SGH eTGH).

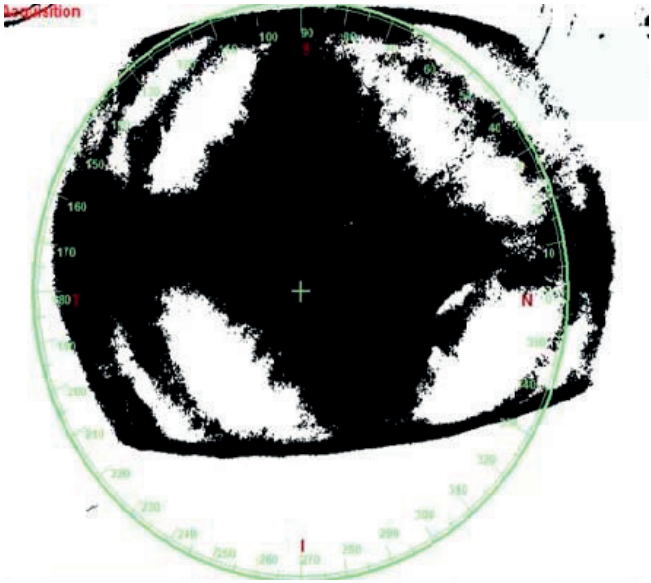

Fig. 1. Lumaxis isogyre acquisition.

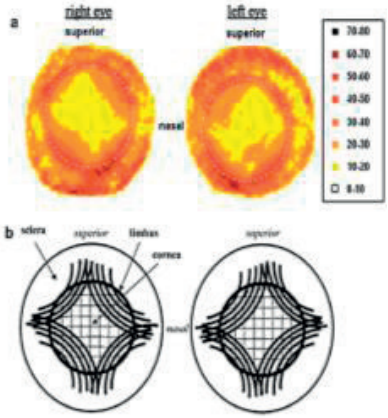

Fig. 2. (*a*) Contour maps of aligned collagen x-ray scatter from a left/right pair of normal human corneas . Note the high degree of body-line mirror symmetry. (*b*) Theoretical model of fibrillar arrangement based on (*a*). Reproduced from Boote *et al.*[30]

4. Results

Data obtained by the polarimetric interferometry showed a cross-like pattern (isogyre), shown in Figure 1, which perfectly correlates with the pattern obtained by the x-ray, THG, and SHG analysis (Figs. 2 and 3), confirming that deep stroma lamellae have two preferential alignments along the superior-inferior and nasal-temporal directions. The regular distribution of stromal lamellae allows the stroma to behave as a polarizer, which eliminates the diffractive effect of the light during its journey into the stroma.

5. Conclusions

In accordance with the importance of regularity and orientation of stromal lamellae distribution in the corneal refractive process, the importance of polarimetric interferometry as a non-invasive technique to detect such orientation as a consequence of correlation/decorrelation between probe light polarization plane angle and stromal lamellae orientation becomes evident. Information from the cross-like pattern can be used for multiple applications in ophthalmology, such as corneal transplantation and diagnosis of corneal diseases due to stromal pathologies. This new technique represents a unique method to correlate the internal stromal structure and optical properties of the cornea. Recent studies have shown a new, intrinsic axis of the stroma, which can have very important refractive applications in ophthalmology.

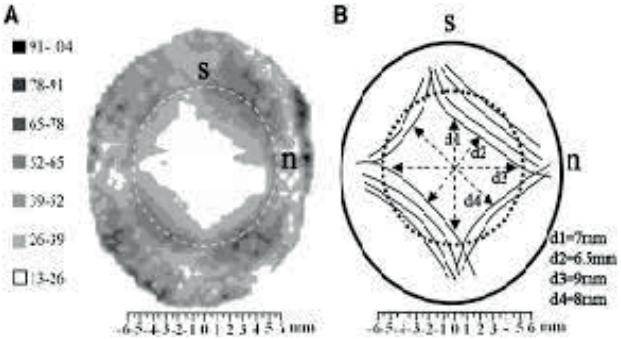

Fig. 3. (*a*) Contour map of aligned collagen x-ray scatter (a.u.) from a right human cornea. Superior, s, and nasal, n, positions are marked. Broken line denotes the limbus. Note the skewed diamond shape of the scatter contours, which displays mirror symmetry between the left and right eyes. (*b*) Proposed model of collagen fibril arrangement to explain the shape of the aligned scatter contours. The peripheral, oblique cornea is reinforced by chords of anchoring collagen of scleral origin. Figure modified from Boote *et al.*[31]

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Eugenio Lipari is the physicist who has developed this new technique; he is also CEO of Phronema SRL.

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