



Bi-directional corneal accommodation in alert chicks with experimentally-induced astigmatism [☆]



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ABSTRACT

This study aimed to characterize corneal accommodation in alert chicks with and without experimentally-induced astigmatism. Refraction and corneal biometry were measured in 16 chicks with experimentally-induced astigmatism (>1.00 D) and 6 age-matched control chicks (astigmatism ≤ 1.00 D). Corneal accommodation was detected using a Placido-ring based videokeratography system, by measuring changes in corneal curvature from a series of consecutive images acquired from alert chicks. The correlation between the magnitudes of corneal accommodation and astigmatism was analyzed by including data from all 22 chicks. Data from all eyes showed obvious bi-directional changes in corneal accommodation. There was no significant difference in corneal accommodative changes between the fellow eyes of the treated birds, and the right and left eyes of control birds. However, positive accommodation (PA) and maximum magnitude of PA (MPA) were significantly higher in the astigmatic vs. the fellow eyes of treated chicks (mean \pm SE: PA = $+2.24 \pm 0.44$ D vs. $+1.26 \pm 0.20$ D; MPA = $+7.53 \pm 0.81$ D vs. $+4.38 \pm 0.53$ D, both $p < 0.05$). This was not the case for negative accommodation (NA) or maximum magnitude of NA (MNA) (NA = -0.46 ± 0.15 D vs. -0.33 ± 0.04 D; MNA = -0.92 ± 0.23 D vs. -0.73 ± 0.12 D, respectively, $p > 0.05$). Furthermore, higher PA and MPA were found to be correlated with higher refractive astigmatism (both $r = 0.34$, $p < 0.05$). These results suggest that the presence of astigmatism may interfere with accommodative function in chicks.

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1. Introduction

The extent to which the cornea, the major refractive component of the eye, plays a role in accommodation is controversial. Although previous studies found 0.40–0.72 D of corneal accommodation in humans aged between 20 and 40 years old (Pierscionek, Popiolek-Masajada, & Kasprzak, 2001; Yasuda & Yamaguchi, 2005; Yasuda, Yamaguchi, & Ohkoshi, 2003), negative results have also been reported (Bannon, 1946; Buehren, Collins, & Carney, 2003; He et al., 2003; Read, Buehren, & Collins, 2007; Rosenfield & Gilmartin, 1987). These inconsistent results may be due to methodological differences or difficulties in detecting subtle changes in corneal curvature. In contrast to the findings in humans, there is stronger evidence for corneal accommodation in several avian species,

including the chicken, which has been proposed as a good model for studying corneal accommodation, because of its prominent amplitude of corneal accommodation (Glasser, Troilo, & Howland, 1994; Troilo & Wallman, 1985). Previous studies showed significant corneal steepening accompanied with lenticular accommodation (Glasser, Troilo, & Howland, 1994; Murphy, Glasser, & Howland, 1995; Ostrin et al., 2011; Schaeffel & Howland, 1987; Troilo & Wallman, 1987) and the total accommodation (i.e., lenticular plus corneal accommodations) can be over 25.00 D (Glasser, Troilo, & Howland, 1994; Schaeffel, Glasser, & Howland, 1988). Indeed, corneal deformation has been estimated to contribute 40.0–50.0% (about 6.00–9.00 D) of the ocular accommodation (Glasser, Troilo, & Howland, 1994; Schaeffel & Howland, 1987; Troilo & Wallman, 1987). Nevertheless, some studies could not detect any corneal accommodation in chicks (Beer, 1892; Sivak et al., 1986).

Corneal accommodation in chicks has been reported to occur due to the contraction of a longitudinal Crampton's muscle (Walls, 1942). This muscle is the anterior portion of the striated ciliary muscle which originates at the sclera, with the scleral occiscle acting as a supporting base (Glasser, Troilo, & Howland, 1994;

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Murphy, Glasser, & Howland, 1995). A direct connection of the muscle to the corneal inner lamella creates a circumferential tension that alters corneal curvature upon muscle contraction. In empirical studies, changes in chick corneal curvature have been measured either by an infrared photokeratometer (García de la Cera et al., 2007; Schaeffel & Howland, 1987; Troilo & Judge, 1993) or by a modified keratometer (Irving, Sivak, & Callender, 1992; Troilo & Wallman, 1987). Ocular accommodation was induced either pharmacologically by treatment with nicotine (Glasser, Troilo, & Howland, 1994; Schmid & Wildsoet, 1997; Troilo & Wallman, 1987), or electrophysiologically by stimulation of the Edinger–Westphal nucleus (Glasser, Troilo, & Howland, 1994; Troilo & Wallman, 1987). However, the extent to which experimental manipulations to stimulate corneal accommodation mimic the natural action of the system is still unclear.

Astigmatism is a refractive error frequently associated with myopia (or “nearsightedness”) and hyperopia (or “farsightedness”) in humans (Read, Collins, & Carney, 2007) and animal models (monkeys: Kee et al., 2005; chicks: Kee & Deng, 2008). It has been hypothesized that the presence of astigmatism may facilitate the accuracy of accommodative response by utilizing the contrast cues associated with the two principal refractive meridians (Howland, 1982); thus the significant astigmatism found in infants could potentially interfere with the eye’s focusing strategy and signaling pathway during early eye growth. However, despite the high prevalence of astigmatism found across different nations (see a summary figure in Kee, 2013), the functional role, if any, of astigmatism during normal and abnormal refractive development remains unclear (Kee, 2013). The present investigation had two key aims. First, we investigated whether we could detect corneal accommodation in chicks under natural viewing conditions: that is with no artificial stimulation, anesthesia, nor the use of lid retractors. Second, we sought to test the hypothesis that corneal accommodation in chicks is influenced by the level of either refractive or corneal astigmatism.

2. Methods

2.1. Animal subjects

Twenty-two White Leghorn chicks (*Gallus gallus domesticus*) were hatched and raised in a temperature- and light-controlled animal room at The Hong Kong Polytechnic University. The light/dark cycle was 12 h/12 h (7:00 am to 7:00 pm) and the illumination level was about 100 lux at the chicks’ eye level. Food and water were provided *ad libitum*. Care and use of the animals were in compliance with the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research and the protocol was reviewed and approved by the Animal Subjects Ethics Sub-committee of the university.

2.2. Manipulations

Sixteen chicks treated by optical manipulations (see below) that developed >1.00 D of corneal astigmatism were included in this study. Six age-matched untreated chicks served as controls. To induce astigmatism, the right eyes of the treated birds were covered, from day 5 to day 12 post-hatching, with a crossed-cylinder lens (+4.00 DS/−8.00 DCx45, $n = 3$; +4.00 DS/−8.00 DCx90, $n = 3$; +2.00 DS/−4.00 DCx180, $n = 3$), a slit aperture (0.5 mm width \times 10 mm height; horizontal slit, $n = 3$; vertical slit, $n = 2$), or a spherical spectacle lens (+15 D, $n = 1$; −15 D, $n = 1$). The fellow eyes were left untreated (we refer to these eyes as, “untreated fellow eyes”). Each lens or slit aperture was first glued to a Velcro ring with Norland Optical Adhesive (Norland Products Inc., New Brunswick,

NJ, USA) and later attached to the Velcro ring’s adhesive mate, which was glued (Pattex leather contact adhesive, Dusseldorf, Germany) to the feathers around the right eye. During the treatment period, the devices were cleaned every morning. All measurements were performed at 12 days of age.

2.3. Measurements

Refractive status was measured under anesthesia with a modified Hartinger refractometer as described previously (Chu, Deng, & Kee, 2012). After refractometry, corneal parameters were measured in alert chicks using a custom-made videokeratography system under dim illumination without using lid retractors. To avoid the potential influence of diurnal effects (Campbell et al., 2008; Johnson et al., 2004), the refractions and corneal curvature measurements were performed between 9:00 am to 11:00 am and 1:00 pm to 5:00 pm, respectively. The components of refractive errors (i.e., M, spherical equivalent; MMM, most myopic meridian; MHM, most hyperopic meridian; RA, refractive astigmatism; R-J0 and R-J45, the two astigmatic components of RA) and corneal curvature parameters (i.e., MK, mean corneal curvature; FK, flattest corneal curvature; SK, steepest corneal curvature; CA, corneal astigmatism; C-J0 and C-J45, the two astigmatic components of CA) were decomposed using power vector analysis (Thibos, Wheeler, & Horner, 1997).

2.3.1. Videokeratography system (VKS)

A Placido-ring videokeratography system (VKS) was custom-built for chick eyes. The instrument comprised of a dome (80 mm in radius) with an inner aperture of 12 mm diameter to house a telecentric imaging system (CCD camera: Guppy AVT F-046, Edmund Optics, NJ, USA). The dome surface has 16 concentric bright rings around the inner aperture (see Fig. 1A). Unlike a previous version (Xu et al., 2009), the current system used a series of white LEDs (illumination LEDs), instead of a circular fluorescent light, to provide even illumination for the Placido-rings (see Fig. 1A). To align the center of Placido-rings with the subject’s pupil center, four infrared LEDs were installed at the outer perimeter of the dome to illuminate the pupil (Fig. 1A, “iris LED”). These LEDs can be switched off independently after a good alignment was achieved (Fig. 1 B and C). Another four red LEDs were installed near the inner aperture to serve as fixation targets to attract chick’s attention (Fig. 1A, “Fixation LED”). Once the image was aligned and focused at a working distance of 80 mm, the iris LEDs were switched off and a series of images were captured in multiple-shot mode (frame rate = 49.4 frame per second) using the software (AVT Fire Package version 3.0) provided by the CCD camera.

To derive the common corneal biometric parameters, images of good quality (sharply focused with good alignment) were selected and analyzed via a user interface written in MatLab software (see Appendix A for details). All corneal parameters were calculated from the central 2.8 mm diameter because the instrumental noise was the lowest (0.18 D) when compared to smaller diameters (see Appendix A for details).

2.3.2. Corneal accommodation

When the chick’s attention was directed to the fixation LEDs, only the iris LEDs were switched off (i.e., the fixation LEDs were still switched on) and a series of continuous frames were captured using the multiple-shot mode as described above (500–1500 frames, 10.1 and 30.3 s duration, respectively). The fixation LEDs, located at 80 mm working distance (i.e., 12.5 D), were the only stimuli for positive accommodation; no stimulus was used to stimulate the negative accommodation. This procedure was performed on each eye consecutively for all birds. After excluding all distorted or disrupted images from the 500–1500 frames acquired from each

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