Chapter 4: Emmetropization and optical aberrations in a myopic corneal refractive surgery chick model

Resumen capítulo 4:

Emetropización y aberraciones ópticas en un modelo de cirugía refractiva corneal en pollos.

En este capítulo estudiamos el potencial de la cirugía láser refractiva corneal para inducir miopía (alargamiento axial del ojo) y las potenciales interacciones entre las aberraciones (inherente a la propia cirugía) y el desarrollo de la miopía en pollos (Gallus domesticus). Para ello diez pollos "White Leghorn" fueron tratados con PRK (Queratometría fotorefractiva) de forma monolateral el día posterior a su nacimiento. La cirugía fue programada para generar un cambio de –9.9 D, es decir hipermetropizando el ojo. La longitud axial se midió mediante biometría de ultrasonidos, el radio de curvatura de la cornea se midió con un video-queratómetro desarrollado para este experimento y el error esférico y aberraciones de alto orden fueron medidos con un Hartmann Shack de desarrollo propio. Todas las medidas tras la cirugía se hicieron en los días 9,12, 14 y 16 de edad de los pollos. A las dos semanas de la cirugía no se aprecian diferencias significativas en los radios corneales entre ojo tratado y control. Tras el tratamiento con PRK el astigmatismo aumentó de media en un factor 2.6 y las aberraciones de tercer orden y superior en un factor 4.3 con respecto al ojo control. Ambos ojos, tratado y control, son prácticamente emétropes tras el tratamiento. Además los ojos tratados no presentan mayor longitud axial que la encontrada en los ojos control. La escasa efectividad de la cirugía refractiva para obtener reducciones significativas de la potencia de la cornea puede ser debido a las propiedades biomecánicas del ojo del pollo. Las aberraciones de alto orden medidas inducían una importante disminución del contraste (de un factor 1.7 a 4.5 ciclos/grado) en la MTF. Sin embargo, la baja calidad de imagen no parece producir una suficiente deprivación de contraste como para generar un error refractivo miope, ni alargamiento axial del ojo en ojos operados con cirugía refractiva corneal. Tras estudiar los datos de ojos normales y tratados se pude concluir que el aumento de las aberraciones oculares impuestas no parecen ser un factor de riesgo para el desarrollo de la miopía.

This chapter is based on the article by García de la Cera et al. “Emmetropization and optical aberrations in a myopic corneal refractive surgery chick model”, Vision Research,47, 2465-2472 (2007), doi:10.1016/j.visres.2007.06.005.

The contribution of Elena García de la Cera to the study was to develop the methodology to measure ocular aberrations in chicks (optical set-up, calibrations, automatic control, data processing routines), as well as the development of routines to measure corneal radius of curvature in chicks. She also performed the experimental measurements on chicks (control and post-refractive surgery) and participated in the data analysis and interpretation.

Coauthors of the study are: Guadalupe Rodríguez, Alberto de Castro, Jesús Merayo and Susana Marcos.

4.1. Abstract

We studied the potential of myopic corneal refractive laser surgery to induce myopia (axial elongation) and potential interactions between aberrations (generally resulting from the procedure) and myopia development in chicks (Gallus domesticus). Ten white Leghorn chicks were monolaterally treated one day post-hatching with photorefractive keratectomy (PRK), with a nominal dioptric change of –9.9 D (imposed hyperopia). Axial length was measured using an adapted ultrasonic biometer; corneal radius of curvature was measured using a custom-built videokeratometer and spherical error and high order aberrations were measured using custom-built Hartmann-Shack aberrometer post-operatively on days 9, 12, 14 & 16 after hatching. Two-weeks after surgery, there were no significant differences in corneal radius of curvature between treated and control eyes. Astigmatism increased on average by a factor of 2.6 and 3rd and higher order aberrations by a factor of 4.3 after PRK. Both treated and control eyes were close to emmetropia, and no axial elongation was found in the treated eyes. The inability of the refractive procedure to achieve significant reductions in the corneal power could be attributed to the biomechanical properties of the chick cornea. High order aberrations induced significant contrast decrease (by a factor of 1.7 at 4.5 c/deg). However, reduced image quality neither produced myopic refractive error nor axial elongation in the treated eyes. Both normal and treated eyes emmetropized, indicating that increased amounts of aberrations do not appear to be a risk factor for myopia.

4.2. Introduction

The quality of visual experience in early stages of post-natal development is critical for proper eye growth and normal emmetropization. In the study presented in Chapter 3 we measured optical aberrations in eyes where myopia had been achieved by severe retinal image quality degradation with diffusers (with no feedback loop) and found that increased aberrations were a cause rather than a consequence of myopia development. Also, a recent study showed that chick eyes that had undergone ciliary nerve section showed larger amounts of higher-order aberrations but did not become myopic, implying that retinal image degradation imposed by certain amounts of aberrations do not necessarily affect the emmetropization process (Tian and Wildsoet 2006). On the other hand, Campbell et al. (Kisilak et al. 2006) found that increased aberrations immediately preceded myopia development in chicks treated with negative lenses, suggesting some role of ocular aberrations in emmetropization.

An increasingly popular technique to correct refractive errors in humans is corneal refractive surgery. Corneal power is changed using excimer laser, reshaping the anterior surface of the cornea by laser ablation of corneal tissue. Corneal photorefractive keratectomy (PRK) has been shown to produce reliable refractive results in humans, with efficacies of 90%, and stability (changes in spherical equivalent less than 1 D, 6 and 12 months after surgery) of 85.8% (Tuunanen and Tervo 1998). The potential use of corneal refractive surgery to produce a permanent change in corneal power seems attractive as an alternative to current methods used to impose experimental refractive errors in laboratory animals and to study mechanisms of refractive error development. PRK has been used to alter emmetropization in the rabbit (Bryant et al. 1999) and infant Rhesus Monkeys (Zhong et al. 2004). In both cases, the axial length changed to compensate for the induced defocus. In this chapter, we will evaluate the feasibility of a refractive surgery myopia model in chicks. In addition, we will evaluate the optical outcomes of the refractive surgery model in chick, by measuring the effective change in corneal curvature, refraction and optical aberrations. A refractive surgery model in adult chickens had been previously used to test the effect of refractive surgery on corneal transparency (Merayo-Lloves et al. 2001).

Several studies, primarily in human patients, have shown that while laser refractive surgery is in general successful at correcting defocus and astigmatism, high order aberrations are generally induced (Moreno-Barriuso et al. 2001) and these affect the quality of the retinal image (Marcos 2001). If, as found in human patients, corneal refractive surgery induces significant amounts of high order aberrations, a refractive surgery model could be used as a model of permanently imposed abnormally high aberrations. Retinal image degradation caused by high order aberrations may be particularly relevant in the chick eye, which (unlike other species (see Chapter 5) (García de la Cera et al. 2006) shows naturally very low amounts of high order aberrations (see Chapter 3) (García de la Cera et al. 2006) allowing the study of potential interactions between aberrations and myopia development. If corneal power is altered (by flattening the anterior cornea) and high order aberrations are induced in the laser treatment, but axial elongation still occurs to compensate for the imposed defocus, we will conclude that the presence of aberrations does not interfere with normal emmetropization.

In this chapter we present post-operative measurements of refraction, axial length, corneal radius of curvature and monochromatic aberrations in chick eyes treated with myopic corneal refractive surgery and their contralateral, untreated eyes during. The aims of the study were to investigate:

1) The potential of a chick myopia model using corneal refractive surgery to impose hyperopic defocus;

2) The changes in corneal curvature, refractive error and ocular aberrations produced by refractive surgery;

3) effects of increased aberrations in the emmetropization process;

If corneal surgery produces a hyperopic defocus in chicks, this technique could be an alternative way for developing chick models of myopia. Also, while the chick has been used as an experimental model of refractive surgery, the optical changes induced by the treatment (other than corneal transparency) have never been evaluated. Finally, the experiments presented in the previous chapter showed that increased aberrations resulted as a consequence on induced myopia. The experiments of the present chapter will aim at testing the hypothesis in the reversed direction, i.e. whether artificially induced aberrations may result in myopia development.

4.3. Methods

4.3.1. Subjects and experimental protocols

Ten White-Leghorn chicks were monocularly treated (OD) with myopic corneal refractive surgery with excimer laser (PRK) one-day post hatching (Day 0), while the left eye was not treated and was used as control. All experimental protocols followed the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research and had been approved by the Institutional Review Boards. Chicks were labeled for identification with color wires attached around their feet. Chicks were reared under fluorescent lighting (12h/12h light/dark cycle conditions) in a cage inside a controlled heated room (24-28 ºC). They were allowed to eat and drink *ad libitum*. Adequate measures were taken to minimize pain or discomfort. Axial length was measured in all chicks on their first day after hatching (Day 0) and prior to surgery. Post-operative measurements were done on both eyes on days 9, 12, 14 and 16. Measurements were not done immediately following surgery since corneal re-epithelization and wound healing processes, as well as increased tear secretion, would have prevented from obtaining reliable results. Measurements consisted of Hartmann-Shack aberrometry and keratometry in five chicks and ultrasound biometry in all chicks. Measurements were done with the animals awake and under natural viewing conditions.

4.3.2. Refractive surgery

Refractive surgery was performed using an excimer laser SVS Apex PlusTM (Summit Technology) (see Figure 4.1). Chicks underwent refractive surgery under total and topical anesthesia (0.02 ml Ketamine, 0.1 g/ml). Prior to laser treatment, the corneal epithelium was removed mechanically, and then laser treatment was applied on Bowman´s layer (178 pulses). Finally the cornea was irrigated with buffered saline solution (BSS). The nominal myopia correction programmed into the laser system was -9.9 D, with an optical zone of 3.5 mm and a nominal corneal tissue depth ablation of 45 microns. Pachymetry measurements on 8 newborn chick eyes (used in trial surgeries) showed a pre-operative corneal thickness of 190 ± 6 microns. Computer simulations using theoretical ablation profiles (based on Munnerlyn or the parabolic approximation of the Munnerlyn equation (Cano et al. 2004)) predicted similar refractive outcomes using chick corneal dimensions than human corneal dimensions. All surgeries were uneventful and all chicks had recovered (i.e. they opened the eyes normally and exhibited no signs of photosensitivy) 8 days after surgery.

Figure 4.1. Photograph of the excimer laser (left) and a chick refractive surgery (right) used to perform surgery on chicks.

4.3.3. Hartmann-Shack aberrometry and refraction

Aberrations were measured using the custom-built Hartmann-Shack (HS) aberrometer described in section 2.1.1. Measurements of the refractive state with streak retinoscopy were attempted in treated eyes, but the bad quality of the reflections (showing scissor-type images) prevented us from obtaining reliable results. The HS aberrometry data were repetitive and consistent, and the spherical error was obtained from 2^{nd} order polynomials. The animal were placed on an elevated platform in front of the system, which was mounted on an x-y translational stage, allowing correct centering and focusing of the pupil. The eye pupil was continuously monitored and aligned to the optical axis of the instrument. The animal fixated the illumination spot during a few seconds, allowing obtaining 5-10 Hartmann-Shack images per eye. Typically, HS image frames contained 17-21 spots in the pupillary zone. Pupil diameters were 2.7 ± 0.3 mm on average. The best H-S images were selected for processing and computing the centroids of the retinal spots, following a procedure described in detail in methods chapter. Estimating the wave aberrations were done using modal fitting (up to 5th order Zernike expansion) of the ray deviations. We obtained defocus, astigmatism, and RMS high order aberrations from Zernike coefficients for the maximum pupil size, and also scaled down to 2-mm pupils for comparative purposes. Point spread functions and modulations transfer functions were computed from the wave aberrations assuming a pupil with homogeneous transmittance.

4.3.4. Keratometry

Measurements of the corneal radius of curvature were obtained using a custom-built infrared (IR) photokeratometer, implemented specifically for this study and described in section 2.2.3. The chick was held in front of the camera, at a distance of 27 mm from the LED ring and 71 mm from the CCD. Sequences of images were captured when the pupil appeared in focus and the image of LED-ring was well aligned with the pupil center. Images were processed according to the description of section 2.2.3. Differences in corneal curvature across 4 meridians (45, 90, 135, and 180 deg) allowed estimation of corneal astigmatism

4.3.5. Ultrasound biometry

Axial length was obtained by an adapted ultrasound biometer (Allergan Humphrey Mod. 826), described in 2.2.2. The axial length for each eye was specified as the average of at least five measurements.

4.3.6. Statistical analysis

Agreement of repeated measurements was tested using confidence intervals (CI), with confidence levels of 95%. Statistical differences between control and treated eyes were tested using paired-t test, with significance levels of $p<0.05$. Significance of linear correlations was tested using Pearson´s coefficient of correlation, with significance levels of $p<0.05$.

4.4. Results

4.4.1. Refractive error

Refractive error and astigmatism were obtained from the defocus Zernike term for 2 mm pupil diameter. Figure 4.2 shows the average spherical refractive errors and astigmatism in treated and control eyes on 4 different days, starting 8 days after surgery. Both eyes were close to emmetropia, and although spherical refractive error tended to decrease slightly with age (-0.03 D/day and in control eye and -0.07 D/day) these changes were not statistically significant. There were no statistically significant differences in refractive error (paired t-test) between the treated and control eyes in any of the days. Individually, we only found significant differences in chick #1, day 14 ($p=0.0014$), chick #3, day 16 ($p=0.0176$) and chick #4, day 12 ($p=0.0243$). The changes and amounts in refractive state were consistent with previous data in the literature, and surprisingly, these were not modified by refractive surgery. Measurements tended to be slightly noisier in the treated eyes than in the control eyes, with average standard deviations for repeated measurements of 0.99 D and 0.57 D, respectively. The 95% confidence interval (CI) for repeated measurements was ±0.97 D and ±0.57 D respectively. Inter-subject variability was also larger in treated eyes than in the control eyes (0.98 D in treated eye and 0.66 D in control eyes), and 95% CI were ±1.26 and ±0.62 respectively. Astigmatism was almost constant throughout the measurement period. Treated eyes showed higher values of astigmatism (2.6 ± 0.5 D) on average than the control eyes (1.0 ± 0.2 D), and these differences were statistically significant in all days ($p=0.0013$).

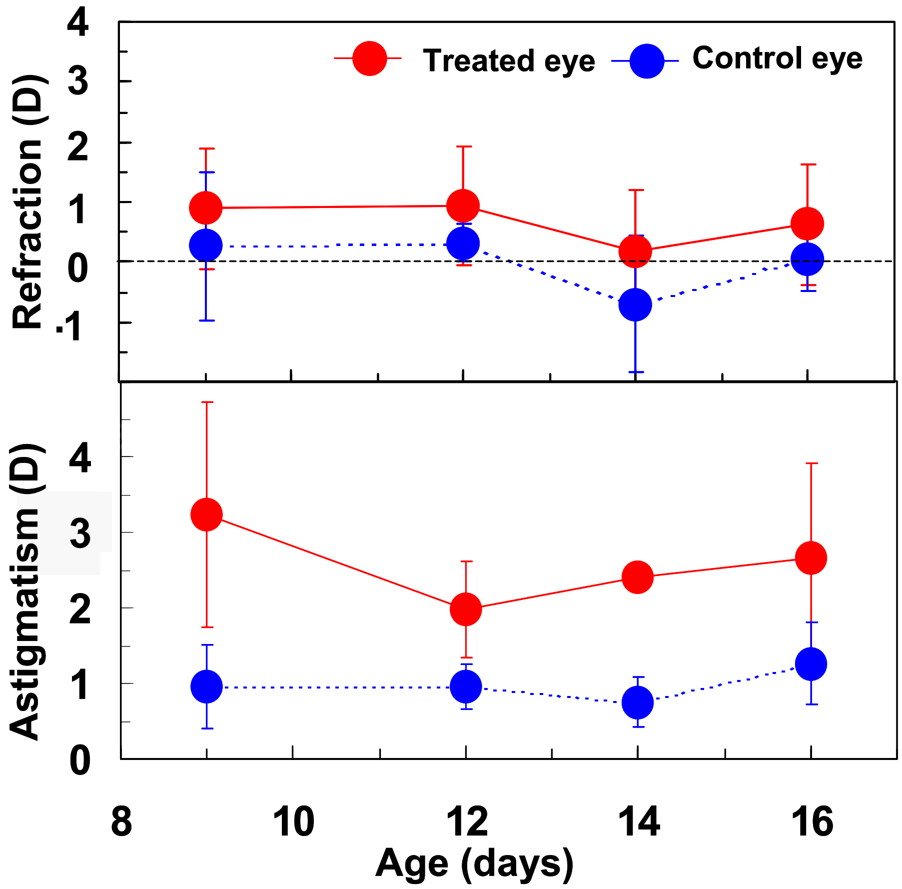

Figure 4.2: Spherical error and astigmatism obtained from defocus and astigmatism Zernike terms during the experiment period (from 8 to 15) days post-operatively. Red circles correspond to eyes treated with refractive surgery, and blue circles to untreated contralateral eyes. Error bars represent ± standard deviations.

4.4.2. Optical aberrations

Figure 4.3 shows examples of wave aberrations for 3rd and higher order in the treated and control eye of the same chick, on day 16 and their corresponding PSFs for 2-mm pupils. The higher number of contour lines in the wave aberration map and larger PSF in the treated eye were indicative of larger optical degradation.

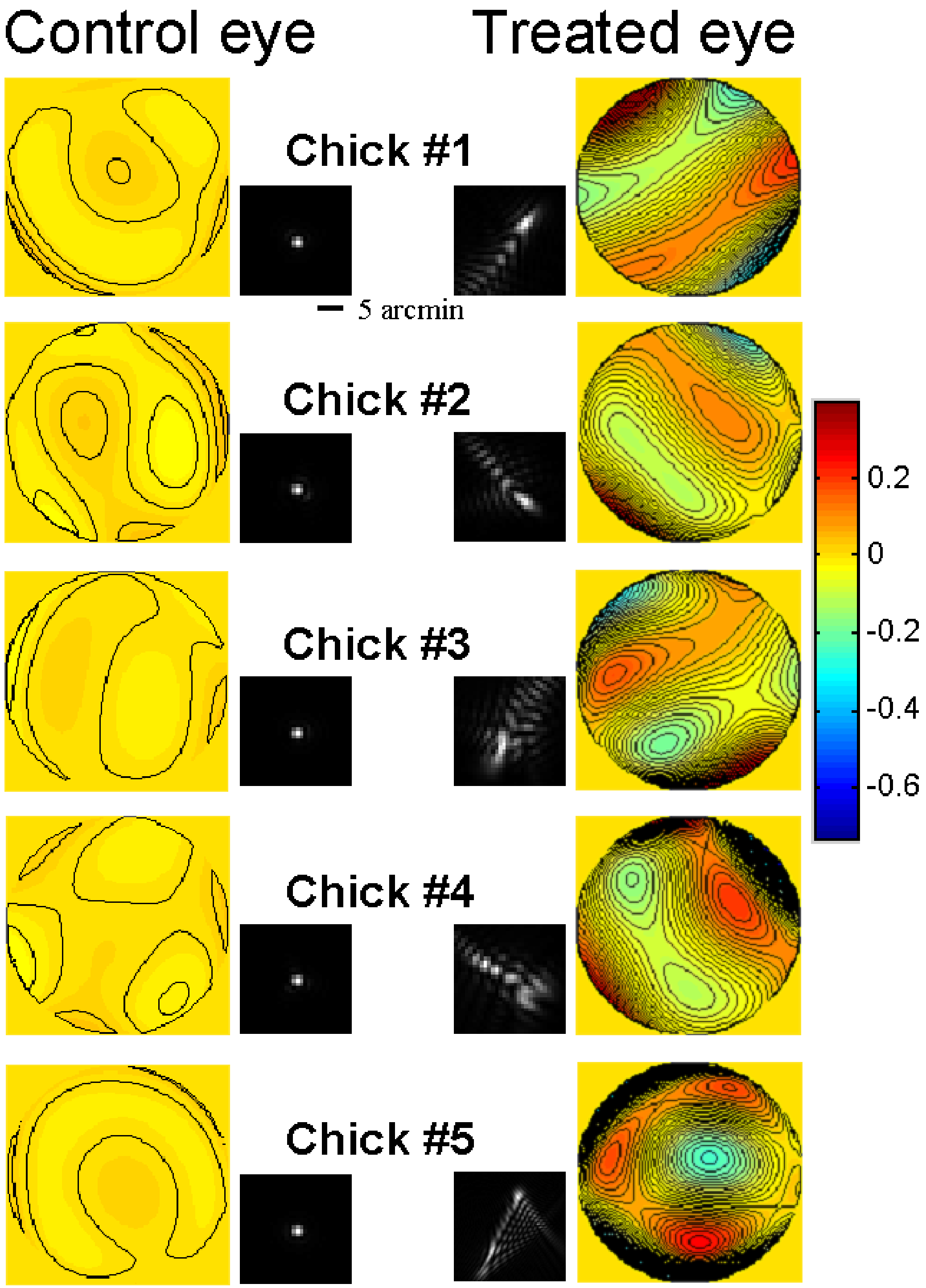

Figure 4.3: Examples of wave aberration maps from all chicks (treated and untreated eye) for 3rd and higher order Zernike coefficients at day 16 and their corresponding PSF for 3rd and higher order aberrations. Data are for 2-mm pupil diameters. Map contour lines are plotted in 0.01 µm steps.

Figure 4.4 shows average 3rd and higher order (A), 3rd order aberrations (B) and spherical aberrations (Z_{40} term) (C) in treated and control eyes on 4 different days, starting 8 days after surgery. Third and higher order root-mean-square wave front error (RMS) was higher in the surgical eyes (4.3 times larger,

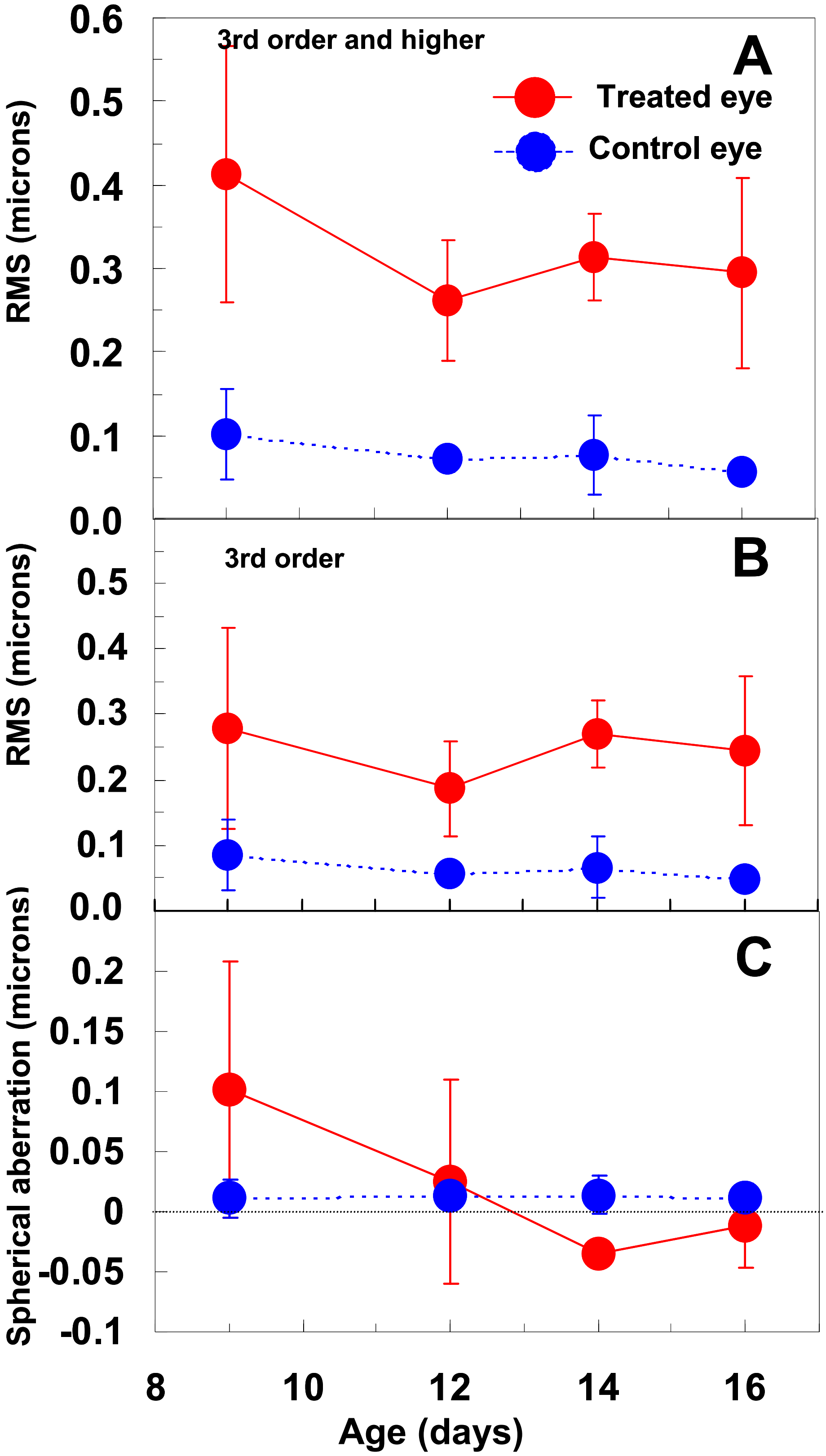

Figure 4.4: A. Mean 3rd and higher order RMS B. 3rd order RMS C. spherical aberration Zernike coefficient (Z_{40}) for days #9,#12,#14 and #16 for treated (red circles) and untreated (blue circles) eyes. Error bars represent ± standard deviations. Data are for 2-mm pupil diameters.

on average) than the control eyes, and the differences were highly statistically significant in all days ($p<0.001$, paired t-test). The increase in RMS was primarily driven by 3rd order aberrations. There were no significant changes in aberrations with time during the studied period. In the control eyes, spherical aberration was not significantly different from zero ($p=0.56$), it presented very little inter subject variability and it remains unchanged across days. In the treated eyes, spherical aberration showed larger inter-subject variability, and tended to decrease with time from positive values to negative values in the studied period, although the differences between treated and control eyes were only significant on day 14. The increase of high order aberrations in the treated eyes resulted in significantly lower modulation transfer functions (MTFs). Figure 4.5 shows MTFs (averaged across eyes) on day 16, for 3rd and higher order in both the treated and control eyes, for 2 mm pupil diameters. Contrast was reduced with surgery at all spatial frequencies. For example, for 4.5 c/deg and 10 c/deg modulation transfer (from 3rd & higher order aberrations) was 1.7 and 2.6 times higher in control than treated eyes.

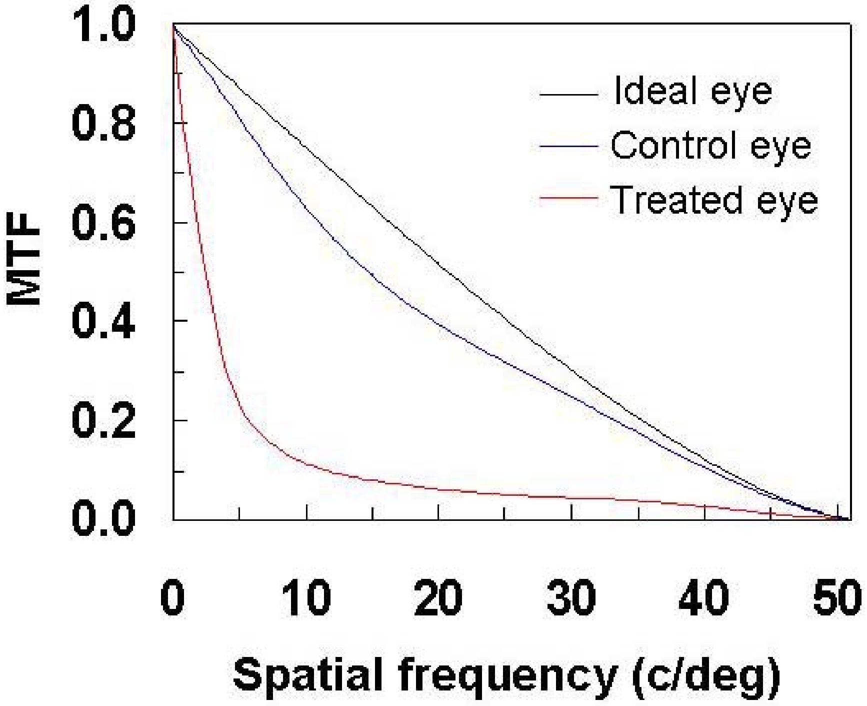

Figure 4.5. Mean MTFs (radial profile) for treated (circles) and untreated (cross) chick eyes, for 3rd and higher order aberrations and 2 mm pupil diameter. For comparison the theoretical MTF of a diffraction-limited eye is also represented (680 nm).

Figure 4.6 shows average corneal radius of curvature in treated and control eyes on 4 different days. In the control eyes corneal radius of curvature increases slightly and the correlation with time was significant (at a rate of 0.023 mm/day, –0.84 D/day, p=0.02), while longitudinal changes in the treated eye were less systematic and the increase was not statistically significant. There were no statistically significant differences in corneal radius of curvature (paired t-test) between the treated and control eyes in any of the days. The mean values of corneal radius (3.15 ± 0.09 mm, or 120 ± 4 D, in treated eye and 3.10 ± 0.07 mm, or 122 ±3 D, in the control eyes) were consistent with previous data on normal eyes in the literature, and surprisingly, these did not appear to have been modified by refractive surgery 8 days after the procedure. Average standard deviations for repeated measurements were similar in treated eyes than in control eyes (0.10 mm, averaging across days and chicks). The 95% CI for repeated measurements was ±0.08 mm in treated eyes and ±0.04 mm in control eyes, averaging across days and chicks. Intersubject variability was larger in treated eyes (standard deviation: ±0.18 mm, 95% CI= ±0.09 mm, averaging across days) than in control eyes (Standard deviation: ±0.04 mm,

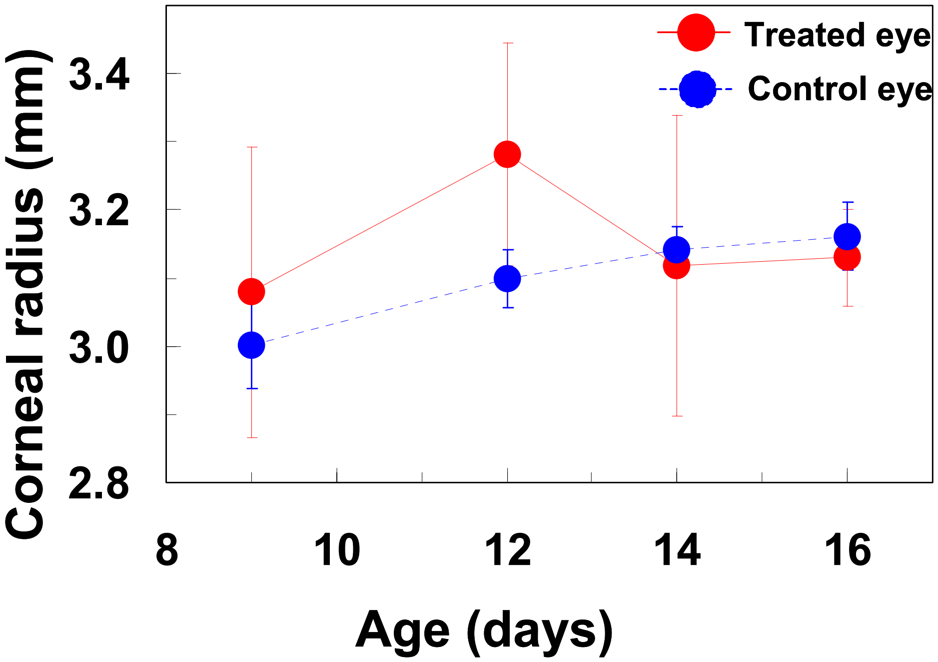

Figure 4.6. Average corneal radius of curvature throughout the experiment period (8 to 15 days post-operatively). Red circles correspond to eyes treated with refractive surgery, and blue circles to untreated contralateral eyes. Error bars represent ± standard deviations.

95% CI= ±0.07 mm). Consistent with the HS measurements of total astigmatism, differences in radii of curvature between the steepest and flattest meridian were higher for the treated eyes (0.28 ± 0.13 mm, averaging across eyes and days) than for control eyes (0.08 ± 0.06 mm), although the differences were not statistically significant.

4.4.4. Axial length

Figure 4.7 shows axial length in treated and control eyes on 4 different days. Axial length increased significantly with age from Day 0 (measured just before treatment, not shown in the graph, 7.39 ± 0.09 mm in the treated eyes and 7.35 ± 0.03 mm in the control eyes) and Day 16 (7.8 ±0.6 mm in the treated eyes and 8.16 ± 0.16 mm in the control eyes). Differences in axial length between treated and control eyes were not statistically significant (paired t-test) in any of the days. The mean values of axial length were consistent with previous data on normal eyes in the literature, and again were not altered by the treatment. Inter-subject variability was slightly higher in treated eyes (0.11 mm, average across eyes and days) than in control eyes (0.08 mm) and 95% CI were ±0.14 mm and ±0.17 mm respectively.

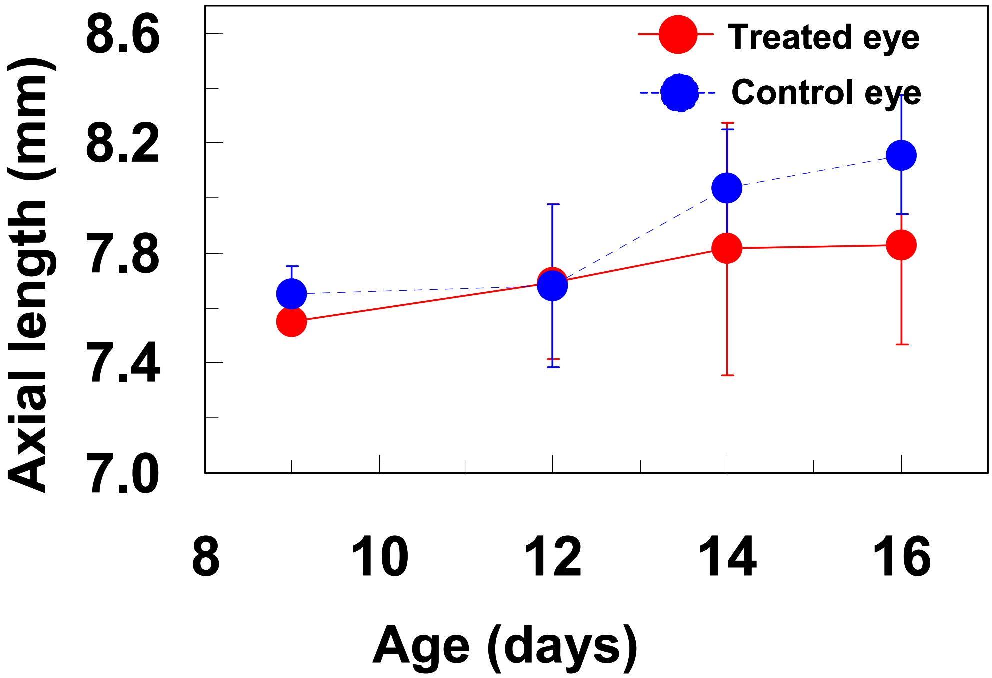

Figure 4.7. Axial length throughout the experiment period (from 8 to 15 days post-operatively). Red circles correspond to eyes treated with refractive surgery, and blue circles to untreated contralateral eyes. Error bars represent ± standard deviations.

4.5. Discussion

We have applied corneal refractive surgery to new born chicks. We did not find that refractive surgery was an efficient way to induce axial elongation: 1) corneal curvature in eyes treated with myopic PRK was not significantly different to control eyes 8 days after treatment; 2) treated eyes exhibited significantly higher amounts of high order aberrations, but the reduction in retinal contrast did not interfere with the emmetropization process.

Chicks have been extensively shown to respond to form deprivation and lens treatments by altering axial ocular growth (Hayes et al. 1986; Wallman and Adams 1987; Troilo and Wallman 1991). In the study presented in Chapter 3 using the same chick strain, from the same hatchery as that used in the present study, and similar time course for treatment and measurements, we found interocular differences between eyes treated with frosted occluders and control contralateral eyes of -17 ± 3 D for refraction and of treatment and axial length of 0.81±0.3 mm by day 13. Numerous studies have shown that functional hyperopia induced by negative lenses induces axial growth that tends to compensate for the induced defocus, at least partially (Schaeffel et al. 1988; Wildsoet and Wallman 1995; Diether and Schaeffel 1997; Priolo et al. 2000; Choh et al. 2006; Schippert and Schaeffel 2006). Some studies found consistently lower amounts of myopia than the power imposed by the negative lens, while others found even larger amounts of myopia that produced by form deprivation when high power lenses were used. For example, Diether and Schaeffel (1997) achieved –3.82 ± 2.48 D using –7.5 D lenses; Schaeffel, Glasser et al. (1988) found similar myopia (-1.5 D) for treatments with either -4 D or -8 D, while Wildsoet and Wallman (1995) achieved –8.6 D after treatment with –15 D lenses and Priolo and Sivak (2000) achieved –12.8± 0.7 D with –10 D lenses in eyes treated one day after hatching. Differences in the effectiveness of the treatment can be affected by the large amplitude of accommodation in chicks (in the experiments performed under natural conditions) and the start day of the treatment (the younger, the more effective).

We attempted to impose hyperopic defocus in chicks (as in negative lens experiments) directly on the cornea, using PRK. Previous studies showed induction of refractive errors in experimental models in infant rhesus monkeys and young rabbits. The hyperopic defocus imposed by treating infant monkey eyes with 3 D myopic PRK, produced consistent hyperopic shifts, corneal flattening and compensatory axial elongation (Zhong et al. 2004). Results from a study in which rabbits (5 and 10 weeks of age) monocularly treated with 5-6 D myopic PRK showed also initially refractive changes which tended to be compensated by increased rate in axial length in the treated eyes (Bryant et al. 1999). In addition to the regression from induced refractive errors in the young group, at the end of the observation period no significant differences were observed in the corneal curvatures between the treated and the control eyes. Surprisingly, hyperopic errors were found in the treated eyes, along with increased axial lengths and similar corneal curvature than in control eyes.

In the present study in chicks, one week after surgery, the refractive treatment with PRK surgery with a nominal negative correction of –9.9 D, did not produce a significant change in corneal curvature. In addition it did not produce increased axial elongations previously obtained as it would have resulted form treatment with a negative lens with the same amount of correction. And it not produce statistically significant anisometropia. Measurements immediately after surgery would have allowed us to assess whether surgery produced the expected corneal curvature and refractive changes which were then cancelled out by regression during the following days. Unfortunately, tear secretion and epithelial changes prevent those measurements to be reliable (even if they were conducted under anesthesia). In this study we did not attempt to measure corneal transparency or scattering following surgery (although transparency measurements in vitro had been performed in this model (del Val et al. 2001)). If haze increased during wound healing, this certainly was not sufficient to induce form deprivation myopia. Refraction, axial length and corneal radius of curvature in the control eyes in this study were similar to previous studies. For example, refraction and axial length of untreated 13-day old chick eyes from the study of the previous chapter on the same chick strain (0.9 ± 0.7 D and 7.9 ± 0.2 mm) (García de la Cera et al. 2006) were similar to those find here despite the differences in the refraction measurement techniques (retinoscopy in the previous study, and Hartmann-Shack here). Published corneal radius of curvature of untreated 2-week old chicks (3.18±0.03 mm) (Li et al. 2000) were similar to these of our study. While some corneal flattening was observed in the treated eyes during the first days of the observation period, the change in corneal power was consistently below the accommodation ability of chicks and in most cases not statistically significantly different from the corneal curvature of the control eyes. If the treatment was effective in reshaping the cornea at all, regression in less than two weeks following surgery may have cancelled the nominally imposed corneal curvature. This effect, also described in a PRK rabbit model, may have occurred more rapidly in chicks for several reasons: 1) the treatment was applied earlier –one day after hatching-, and regression had been associated with earlier treatment (5 versus 10 weeks in the rabbit experiment); 2) chick corneas exhibit higher elasticity than mammalian corneas (Troilo and Wallman 1987; Glasser et al. 1994). It has been proved that under normal physiological conditions, a pressure-mediated mechanism would be able to alter corneal curvature in chicks by about only 3 D (Glasser et al. 1994). However it is likely that the changes in intraocular pressure and decreased corneal thickness following PRK (Schipper et al. 1995) play a major role in increasing corneal curvature and cause regression.

While we have found that, unlike other species, PRK was not effective in changing corneal power of chicks, and therefore as an alternative to spectacle-rearing procedures, high order aberrations were systematically induced by the procedure. As a result, modulation transfer functions in treated eyes were significantly lower than in control eyes. Unlike in human eyes (Marcos et al. 2001; Moreno-Barriuso et al. 2001), spherical aberration did not increase significantly with the procedure (although longitudinal variations were found), perhaps as a result of regression mechanisms similar to defocus. Astigmatism was significantly higher in treated than control eyes (see Figure 4.2). Other asymmetric aberrations such as coma increased significantly, producing increased blur in the retinal images (see Figure 4.2) and consistently decreased contrast (see Figure 4.5) in the treated eyes with respect to control eyes. Bartman and Schaeffel (Bartmann and Schaeffel 1994) found 9 D of induced myopia in chicks wearing diffusers that caused a 4-time decrease in the modulation transfer at 4.5 c/deg. For the same frequency, in this experiment, high order aberrations decreased modulation transfer functions by 2. When astigmatism was considered, the MTF decreased from 0.69 (normal eyes) to 0.21 (treated eyes) for this spatial frequently. Previous studies in chicks had shown that induced astigmatism actually resulted in low but significant hyperopic (and not myopic) refractive error (Mc Lean & Wallman 2003). In infant monkeys it has been shown that induced astigmatism produces both hyperopic and myopic refractive errors (Kee CS, Hung LF et al., 2004). Thus, presence of laser induced astigmatism could prevent myopia development in the treated eye**.** We did not find that the contrast degradation produced by high order aberrations induced neither refractive changes nor significant changes in axial length. This was consistent with recent findings in chicks that had undergone ciliary nerve section (Tian and Wildsoet 2006). The treated chicks showed higher amounts of higher-order aberrations but they did not become myopic. For the same pupil size (2-mm) we found slightly lower HOA aberration values than Tian et al. (Tian and Wildsoet 2006) for the control eyes and of the same order of magnitude for the treated eyes (0.53 D vs. 2-3 D using the equivalent defocus power metric (Cheng et al. 2004)). On the other hand, this was in contrast with studies suggesting that increased aberrations may precede myopia development (Kisilak et al. 2006). Along with differences in magnitude which may set a threshold for image blur below which the emmetropization process was not affected, the nature of the image degradation induced by diffusers (scattering) may be different from that induced by aberrations. We found that the predominant induced aberrations were non-rotationally symmetric. It could be that this type of aberrations (as previously found for astigmatism, Mc Lean and Wallman 2003) may not necessarily trigger myopia development. Future research on the potential involvement of specific high order aberrations (i.e. spherical aberration) in myopia development could be addressed by using phase-plates or customized contact lenses with a better a priori control on the magnitude and type of aberration induced.