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Research Article

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Investigation of Age-Related Changes in Corneal Curvature in Chicken Embryos (Gallus Gallus Domesticus l.) During Embryonic Development

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Abstract

The study is devoted to the study of age differences in the radius of curvature of the anterior and posterior surfaces of the cornea of the left and right eyeballs of chicken embryos on the 10th-20th day of incubation.

The left eyeball is characterized by a predominantly large radius of curvature of the anterior surface of the cornea. The superiority was revealed on the following days of incubation: 11th (2.84%), 12th (1.82%), 13th (2.98%, p0.05), 14th (3.30%, p0.05), 15th (0.21%), 16th (1.23%), 17th (0.40%) and 18th (0.79%).

The superiority of the left eyeball in the radius of curvature of the posterior surface of the cornea was noted on the 10th (64.00%), 12th (20.45%), 13th (18.97%), 15th (8.51%), 16th (11.82%), 18th (2.80%) incubation day.

Studies demonstrate a dynamic change in the curvature of the cornea, reflecting both temporary asymmetries and subsequent stabilization, which may be part of normal embryonic processes.

Keywords: Chicken Embryo, Eyeballs, Cornea and Measurement

1. Introduction

The development of the cornea in chick embryos is an important and complex morphogenetic process. The formation of the cornea begins in the outer ectoderm under the influence of inductive signals from the optic cup [3]. Experimental studies have shown that the area of the forming cornea is directly proportional to the area of its contact with the optic cup, which emphasizes the importance of the interaction of these structures at the beginning of embryonic development [1].

The developmental features of the cornea are the subject of numerous scientific studies. Recently, the main attention of scientists has been focused on the study of genetic and humoral mechanisms that control the development of the cornea [5]. The present study is aimed at analyzing changes in the radius of curvature of the anterior and posterior surfaces of the cornea at different stages of embryonic development in chicken embryos [2].

During the embryonic development of chickens, the cornea undergoes significant changes in the curvature of the anterior and posterior surfaces [4]. In the early stages, around the 4th to 5th day of incubation, the cornea retains a more spherical shape. This sphericity is due to the active growth and development of the eyeball, which requires a certain configuration for the correct formation of visual structures (cornea, lens, and retina) [8].

By the 15th to 17th day of incubation, the cornea gradually flattens. During this period, the differentiation of eye tissues is completed and the cornea adapts to the future functions of the visual system that it will correspond to after hatching. This adaptation includes changes in the optical properties of the eye to ensure optimal vision of the chick [7].

These morphological changes are necessary to ensure the correct functioning of the visual system after hatching. The gradual transformation of the cornea, from a more spherical shape to a flattened one, allows the chick to adapt to external conditions and ensures effective focusing of light rays on the retina. Thus, the study of corneal development helps to understand the fundamental processes involved in the formation of the visual system in birds [6,9].

2. Materials and methods

The study was conducted at the scientific laboratory of the Velikiye Luki State Agricultural Academy of the Pskov Region. The object of the study was the embryos of chickens (Gallus gallus domesticus L.) of the Lohmann Brown cross at various stages of embryonic development.

A total of 30 chicken eggs weighing 51-59 g were selected for incubation. All eggs were carefully selected according to the criteria of shell integrity and marbling degree in order to standardize the source material and ensure its suitability for incubation. The corneas of the embryos' eyeballs, extracted from the 10th to the 20th day of incubation, were subjected to the study.

To access the cornea, microsurgical instruments were used to retract the eyelid to visualize the cornea. The corneal surface (epithelium) was carefully removed using a special scraper [10]. The scleral area, at the transition between the cornea and sclera, was cut using microsurgical scissors. After that, the cornea was circumscribed using tweezers to avoid damaging important structures. The extracted cornea was immersed in 10% formalin solution for further analysis [11].

Corneal morphometric measurements were performed using specialized Screen Meter software, which allows analyzing parameters in micrometers. Corneal diameter measurements were made both horizontally and vertically on the anterior and posterior surfaces. These procedures helped to ensure the accuracy of the data needed to analyze changes in corneal curvature at different stages of embryonic development.

3. Results and Discussion

The radius of curvature of the cornea changes with age. In chicken embryos, it is more spherical and flattens by the 15th-17th day of incubation, which is accompanied by a change in the diameter and area of the cornea. On the 18th day of incubation, the cornea becomes more compact, acquiring its final shape and transparency, which is important for preparing for hatching.

The sphericity and smoothness of the anterior surface of the cornea are key factors that ensure its transparency. Violation of sphericity can lead to the development of astigmatism, which significantly reduces the quality of vision. This is especially noticeable in keratoconus. In cases where scar tissue of the cornea is formed, but its curvature is preserved, visual acuity suffers to a lesser extent.

Astigmatism in chickens, as in other animals, is associated with an irregular shape of the cornea or lens, which leads to distortion of the image on the retina. In chickens, this may manifest itself as uneven refraction of light, affecting their vision. However, in general, chicken vision is well adapted to their lifestyle, and astigmatism is not a common problem. Analysis of the radius of curvature of the anterior surface of the cornea in chicken embryos was carried out from the 10th to the 20th day of incubation. Table 1 and Figures 1 and 2 present data on changes in the radius of curvature of the anterior and posterior surfaces of the cornea of the eyeballs of chickens at various stages of embryonic development. On the 10th day of incubation, the radius of curvature of the cornea in the right eyeballs is greater than that in the left eyeballs, amounting to 3.80%. This indicates a more significant curvature of the cornea in the right eyeballs.

The situation reverses on the 11th day of incubation, when the radius of curvature of the cornea in the left eyeballs exceeds the radius of curvature of the cornea in the right eyeballs by 2.84%. On the 12th day of incubation, the superiority of the radius of curvature of the cornea in the left eyeballs remains, although the percentage decreases to 1.82%.

The difference increases again, and the radius of curvature of the cornea in the left eyeballs significantly exceeds the curvature of the cornea in the right eyeballs on the 13th day of incubation (2.98%, $p<0.05$), which may be an indicator of abrupt growth. On the 14th day of incubation, the superiority of the radius of curvature of the cornea in the left eyeballs is again reliable and amounts to 3.30% ($p<0.05$).

The difference in the radius of curvature of the cornea from the 15th to the 18th day of incubation between the left and right eyeballs with the superiority of the left becomes less noticeable, from 0.21% to 1.23%, which indicates stabilization and minimization of asymmetries.

However, a slight excess of the radius of curvature of the cornea is observed from the 19th to the 20th day of incubation in the right eyeballs, 0.19% and 0.18%, but these differences remain insignificant and have virtually no effect on the overall symmetry of corneal development.

Development Day	Radius of curvature of the anterior surface of the cornea, µm		Radius of curvature of the posterior surface of the cornea, µm	
	Left (L)	Right (R)	Left (L)	Right
Early fetal stage (7-12 days)				
10	3,16±0,10	3,28±0,13	0,41±0,07	0,25±0,03
11	3,62±0,13	3,52±0,06	0,40±0,09	0,44±0,04

12	3,91±0,20	3,84±0,08	0,53±0,03	0,44±0,04
Mid-fetal stage (13-17 days)				
13	4,49±0,02*	4,36±0,03	0,69±0,04	0,58±0,06
14	4,69±0,03*	4,54±0,04	0,80±0,03	0,84±0,05
15	4,79±0,01	4,78±0,06	1,02±0,07	0,94±0,06
16	4,93±0,05	4,87±0,03	1,23±0,04	1,10±0,04
17	4,96±0,03	4,94±0,06	1,41±0,05	1,42±0,02
Late fetal stage (18-20 days)				
18	5,10±0,05	5,06±0,03	1,47±0,02	1,43±0,02
19	5,21±0,08	5,22±0,06	1,53±0,03	1,44±0,06
20	5,59±0,15	5,60±0,07	1,51±0,01	1,58±0,04
*p<0,05				

Table 1: Age Differences in the Radius of Curvature of The Anterior and Posterior Surfaces of The Cornea of The Eyeballs of Chickens at Different Days and Stages of Embryonic Development, µm

The presented data on the radius of curvature of the anterior corneal surface demonstrate periodically occurring asymmetries in development, which in most cases are balanced by natural growth mechanisms. Changes in the parameters indicate a complex dynamics of morphogenesis, during which adaptation and optimization of the visual structures of embryos at different stages of incubation occur.

On the 10th day of incubation, the radius of curvature of the posterior corneal surface in the left eyeballs exceeds the radius of curvature of the right eyeballs by a significant 64.00%, but the differences are not reliable. This significant difference may be caused by differences in local cellular metabolism and initial tissue differentiation. Different rates of development may be due to the fact that in one eyeball, special genetic programs are activated that cause cells to divide faster and form eye structures such as the cornea, lens, and retina faster than in the other eyeball.

However, on the 11th day of incubation, a slight, 10.00%, superiority of the indicator in the right eyeballs compared to the left ones was revealed. This may mean that processes are activated that begin to function in order to correct the initial differences in the development of the cornea of the eyeballs, to ensure their more uniform and balanced development.

By the 12th day, a superiority of the corneal curvature in the left eyeballs is again observed, now by 20.45%, which may indicate a phase of accelerated cell growth or changes in water-salt metabolism, leading to an increase in the curvature of the cornea. On the 13th day, this advantage slightly decreases, to 18.97%, which may be a sign of gradual equalization of hormonal or nutritional influences. On the 14th day of incubation, the corneal curvature again shows a superiority in favor of the right eyeballs, where the indicator exceeds that of the left eyeballs by 5.00%. Such changes could be due to adjustments in the volume and composition of the intraocular fluid or changes in the activity of specific growth factors.

On the 15th and 16th days of incubation, the radius of corneal curvature in the left eyeballs again turns out to be greater than in the right, first with an excess of 8.51%, then by 11.82% on the 16th day.

On the 17th day of incubation, the radius of corneal curvature in the right eyeballs again demonstrates a slight superiority in development (0.71%), which may reflect an intermediate phase of stabilization and focusing of resources on preparation for hatching. From the 18th to the 19th day of incubation, another superiority of the radius of curvature of the cornea in the left eyeballs is recorded, first by 2.80%, and then by 6.25%. This indicates active processes of final alignment and formation of the structure of the eyeballs to functional maturity.

However, on the 20th day of incubation, it was found that the radius of curvature of the cornea in the right eyeballs was again greater than the radius of curvature of the cornea in the left eyeballs by 4.64%. This change may indicate cyclical processes in the development and regulation of interocular asymmetry.

Analysis of the radius of curvature of the posterior corneal surface in chicken embryos from the 10th to the 20th day of incubation shows complex changes that reflect different stages of development and adaptation of the visual structures. During this time, it is observed that the radius of curvature of the posterior corneal surface becomes greater in the left and right eyeballs. This alternation may indicate that the left and right eyeballs develop at different rates or that they adapt differently during growth during the embryonic period.

In general, the data analysis shows that the radius of curvature of both the anterior and posterior surfaces of the cornea demonstrates an advantage in the left eyeballs, including on the 13th-14th day for the anterior surface of the cornea reliably. This may indicate more active growth processes or specific physiological mechanisms that contribute to an increase in the radius of curvature of the cornea in the left eyeballs. The study of age-related differences in the

radius of curvature of the anterior and posterior surfaces of the cornea in the left and right eyeballs of chicken embryos made it possible to identify several key phases characterized by temporary asymmetries and their subsequent alignment (Fig. 1, 2).

On the 10th day of incubation, the first significant change in the radius of curvature of the anterior surface of the cornea is observed (Fig. 1). In the right eyeballs, the curvature of the anterior surface of the cornea is higher than in the left eyeballs. On the 11th-14th day of incubation, a change in dynamics occurs – the radius of curvature of the anterior surface of the cornea in the left eyeballs begins to exceed similar indicators of the right eyeballs.

By the 15th day of incubation, the radius of curvature of the anterior surface of the cornea in both the left and right eyeballs is leveled. By the 16th day, an increase in the indicator of the left eyeballs is again recorded. At the final stages of incubation, the opposite change is observed - on the 19th-20th day of incubation, the radius of curvature of the anterior surface of the cornea of the right eyeballs is higher than that of the left eyeballs.

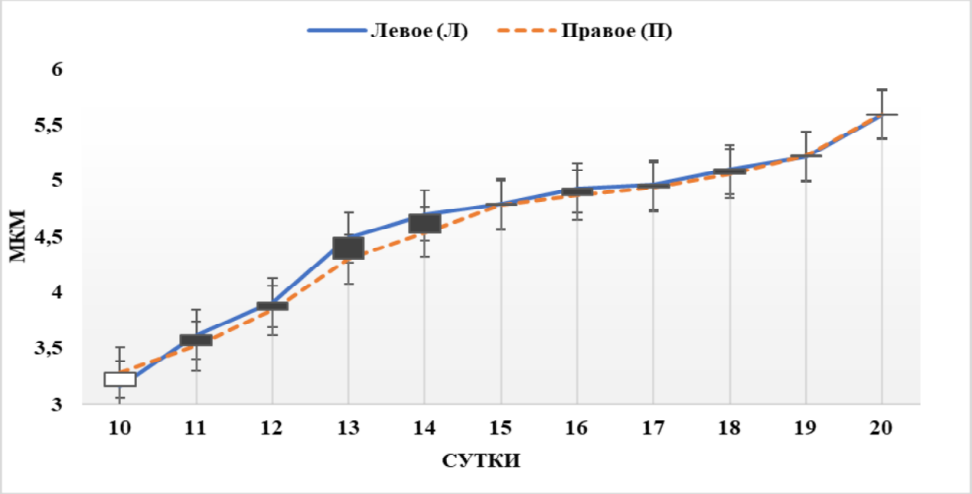

Figure 1: Radius of Curvature of The Anterior Surface of The Cornea, µm

The information presented in the figure demonstrates the characteristic stages of development and dynamic changes in the radius of curvature of the cornea, revealing both temporary asymmetries and subsequent stabilization of these parameters. Such changes can be part of normal embryonic processes reflecting adaptations and unique developmental conditions.

It is noteworthy that on the 10th day of incubation, in contrast to the anterior surface, the posterior surface of the cornea demonstrates the opposite dynamics (Fig. 2). In chicken embryos, the radius of curvature of the posterior surface in the left eyeballs is higher than in the right eyeballs. The advantage of the radius of curvature of the posterior surface in the left eyeballs is maintained from the 12th to the 13th day of incubation. By the 14th day of incubation, a partial alignment of the indicators occurs. The rate of corneal growth stabilizes, and significant differences between the left and right eyeballs are no longer observed.

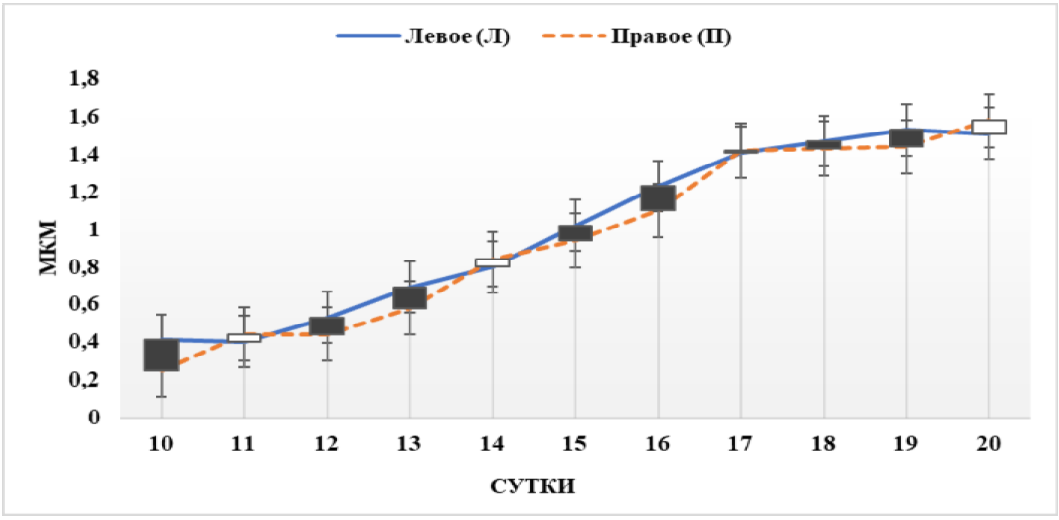

Figure 2: Posterior Corneal Surface Curvature Radius, µm

On the 15th-16th day of incubation, the posterior corneal curvature radius of the left eyeballs again begin to exceed the right eyeballs. By the 17th day of incubation, the differences begin to smooth out, but on the 18th-19th day, a slight superiority of the indicator is again noticeable in the left eyeballs. On the 20th day of incubation, the opposite change occurs: the posterior surface curvature radius in the right eyeballs begins to slightly exceed the indicator in the left eyeballs.

4. Conclusions

In the radius of curvature of the anterior surface of the cornea, the advantage is noted in the right eyeballs on the 10th day. From the 11th to the 14th day of incubation, the radius of curvature of the cornea in the left eyeballs exceeds that in the right eyeballs, gradually reaching almost equal values by the 15th day. Then, from the 16th day, the curvature of the cornea in the left eyeballs increases, and on the 19th-20th day, an insignificant but inverse asymmetry is again recorded with the superiority of the right eyeballs.

In the radius of curvature of the posterior surface of the cornea, on the 10th day, the advantage is in the left eyeballs, the opposite result is on the 11th, 14th and 17th days of incubation. On the 12th-13th, 15th-16th and 18th-19th days, the left eyeballs again show superiority, which smooths out later, and by the 20th day, the right eyeballs already show predominance.

The study showed that changes in the corneal curvature occur gradually and unevenly, reflecting complex adaptation processes. These changes play an important role in ensuring balanced growth and formation of eye structures necessary for the normal functioning of the visual apparatus after hatching. Data analysis shows that the radius of curvature of both the anterior and posterior surfaces of the cornea usually demonstrates an advantage in the left eyeballs, including on the 13th-14th day for the anterior surface of the cornea reliably.

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