



ISSN (E): 2277-7695

ISSN (P): 2349-8242

NAAS Rating: 5.23

TPI 2023; 12(5): 4098-4100

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www.thepharmajournal.com

Received: 05-02-2023

Accepted: 18-04-2023

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Histomorphometrical study on cornea of buffalo (*Bubalus bubalis*)

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Abstract

Ten pairs of eyeballs from buffalo were collected to study and record the histomorphometry of the cornea. The anterior epithelium, corneal stroma, descemet's membrane, and endothelium were found to be arranged serially from the external to the internal surface of the cornea. The mean value of the total thickness of the cornea at the periphery was $798.607 \pm 69.17 \mu\text{m}$. The major part of the cornea (>80%) was formed by the stroma. The anterior epithelium was stratified squamous epithelium, and the number of layers ranged from 7 to 12. The endothelium was formed by a single row of flattened cells with prominent elongated nuclei behind the Descemet's membrane.

Keywords: Cornea, histomorphometry, buffalo, epithelium

Introduction

The performance of animals is greatly influenced by their proper vision. The cornea, being one of the refractive media of the eye, plays an important role in vision or image formation. The whole cornea is always exposed to the external environment when the eye is open; therefore, it is frequently subjected to injury that would cause discomfort to the animal. This work was carried out to study the cornea of buffalo (surti) to provide baseline data as well as to help the ophthalmist or surgeon have a better understanding of the cornea.

Materials and Method

The research specimens of eyeballs were collected from a local slaughterhouse in Anand. Davidson's fixative was used for fixation, and the histological sections were stained with haematoxylin and eosin stain as per the method of Singh and Sulochana (1996) [1] and Masson's trichrome stain as per the method of Luna (1968) [2]. The histomorphometrical measurements, taken from the periphery of the cornea, were recorded with the help of a graduated eyepiece. The statistical data were analysed as per the method of Snedecor and Cochran (1994) [3].

Results and Discussion

The cornea was found to be composed of four layers (Fig. 1). The observation was in agreement with Martin and Anderson (1981) [4] in domestic animals and Ramkrishna *et al.* (1997) [5] in Indian water buffalo. The different micrometrical observations of the cornea are shown in Table 1.

The microscopic layers of the cornea, from the external to the internal, were:

Anterior Epithelium: The anterior epithelium was of stratified squamous non-keratinized type, and the nuclei of the cells were of blackish blue colour (H&E staining). The basal columnar cells were lying on the basement membrane, and the cells towards the outer surface were flattened or squamous (Fig. 2). The anterior epithelium was the second thickest layer, and the mean thickness recorded was $99.92 \pm 5.32 \mu\text{m}$. The present finding was in agreement with Khaled (2003) [6] in bovine ($98.00 \pm 1.50 \mu$). The mean number of rows of epithelial cells recorded was 9.42 ± 0.11 , which was in agreement with Banubakode (1992) [7] in cattle (9.28 ± 0.17) but found to be higher as compared to Barhaiya *et al.* (2015) [8] in goats (5 to 9 rows of epithelial cells).

Corneal stroma: The major part (>80%) of the cornea is constituted by this layer. The stroma was characterised by the presence of regularly arranged sheets of collagen fibres and fibroblasts (Fig. 1). The mean thickness recorded of the stroma in the present study was $667.32 \pm 67.27 \mu\text{m}$. Khaled (2003) [6] reported a lower mean value of the corneal stroma ($580 \pm 40 \mu\text{m}$) in bovines.

Descemet's membrane: This membrane was in between the corneal stroma and the endothelium. This homogenous, eosinophilic membrane was composed of faintly stained, loosely arranged collagen fibres. (Fig.1). The mean thickness of Descemet's membrane was found to be $18.79 \pm 2.36 \mu\text{m}$. Khaled (2003) [6] in bovines ($30 \pm 1.0 \mu\text{m}$) reported the higher value; however, Bloom and Fawcett (1962) [9] in human beings (5 to 10 μm) reported the lower value than the present study.

Endothelium: The last and most posterior layer of the cornea was formed by a single row of flattened cells with prominent elongated nuclei, the endothelium. It was lying behind Descemet's membrane, parallel to the corneal surface (Fig. 1). The mean thickness of endothelium recorded was $5.08 \pm 0.37 \mu\text{m}$ in the present study. The finding was in agreement with Prince *et al.* (1960) [10] in cattle (6 μm) and Diesem (1977) [11] in bovines (6 μm).

Total thickness of the cornea

The mean total thickness of the cornea recorded in the present study was $798.98 \pm 69.17 \mu\text{m}$ and ranged from 474.49 to 1215.90 μm . The finding was in agreement with Prince *et al.* (1960) [10] in cattle (750 to 850 μm). However, Diesem (1977) [11] in bovines (1500 to 2000 μm) and Banubakode (1992) [7] in cattle ($933.72 \pm 15.35 \mu\text{m}$) reported the higher mean value of cornea, whereas, Camber *et al.* (1987) [12] in pig (722 μm) and Barhaiya *et al.* (2015) [8] in goat ($716.39 \pm 13.84 \mu\text{m}$) reported the lower value for the same.

The present histomorphometrical study showed that domestic animals showed variations in the thickness of different layers of the cornea. These variations may be due to differences in breed, species, etc.

The present study showed similar general histological structures of the cornea in different species. However, the layer of the cornea, i.e., "Bowman's membrane," reported by Smythe (1956) [13] and Prince *et al.* (1960) [10] in cattle, Ross and Edward (1985) [14] in human beings, Martin and Anderson (1981) [4] in cattle, Banubakode (1992) [7] in cattle, Khaled (2003) [6] in bovine, and Barhaiya *et al.* (2015) [8] in goat eyeball, was indistinct in the present study. Gelatt (2007) [15] mentioned that the Bowman's layer was absent in the cornea of most animals.

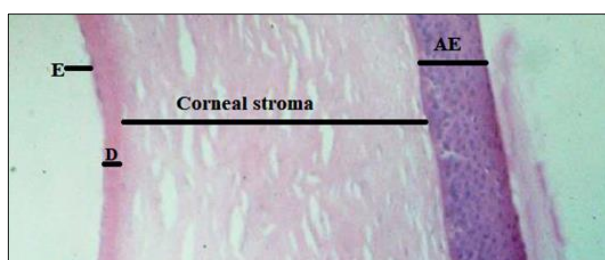


Fig 1: Cornea of surti buffalo (AE- Anterior epithelium, D- Descemet's membrane and E-Posterior endothelium) (150X magnification, H&E Stain)

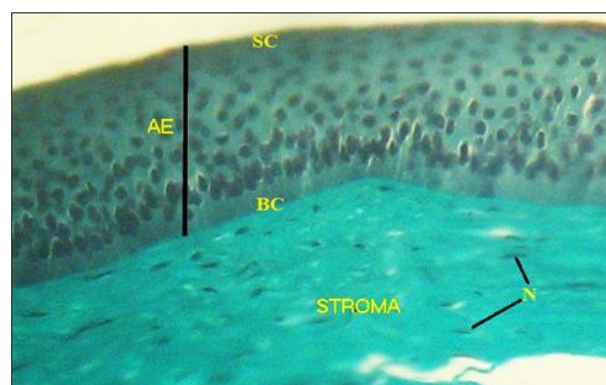


Fig 2: Cornea of surti buffalo (AE- anterior epithelium, BC- Basal columnar cells, SC- Superficial squamous cells, N- Nuclei of fibroblasts and green colour stained collagenous bundles of stroma) (300X magnification, Masson's trichrome stain)

Table 1: Statistical analysis of micrometry of cornea

| Sl. no. | Parameters | Range | Mean \pm SE |
|---------|--|-------------------|--------------------|
| 1 | Number of epithelial cell layers | 7 to 12 | 9.42 \pm 0.11 |
| 2 | Thickness of epithelium (μm) | 67.59 to 132.00 | 99.92 \pm 5.32 |
| 3 | Thickness of stroma (μm) | 324.24 to 1080.80 | 667.32 \pm 67.27 |
| 4 | Thickness of Descemet's membrane (μm) | 9.90 to 27.02 | 18.79 \pm 2.36 |
| 5 | Thickness of endothelium (μm) | 3.30 to 6.60 | 5.08 \pm 0.37 |
| 6 | Total thickness (μm) | 474.49 to 1215.90 | 798.98 \pm 69.17 |

Conclusion

The histological section of the cornea was composed of four layers: anterior epithelium, corneal stroma, Descemet's membrane, and endothelial layer. The Bowman's membrane of the cornea was not evident in the present study. The mean total thickness of the cornea recorded in the present study was $798.98 \pm 69.17 \mu\text{m}$ at the periphery.

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