

Research Article – Histology and cell biology

Third eyelid in the small Indian mongoose (*Herpestes javanicus*): a morphological and histological study

Younes Kamali, Zabihollah Khaksar, Soghra Gholami*

Department of Basic Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran

Submitted February 2, 2015; accepted revised May 8, 2015

Abstract

The Indian mongoose (*Herpestes javanicus*) is native to parts of Asia, Iran. The purpose of this study was to describe the gross anatomy of the cartilage and histology of the superficial gland of the third eyelid of two adult mongooses. The animals, in terminal stages of disease and near death due to aging or unknown reasons, were referred from Park Zoo (Shiraz, Iran) to our center. By using a modified maceration technique, the morphological characteristics of the cartilage were examined. For histological examinations of the superficial gland of the third eyelid, the samples were stained with haematoxylin and eosin. Also, to detect the elastic fibers in the cartilage sections were stained with orcein and Weigert's resorcin-fuchsin. The cartilage consisted of an ovoid appendix and a mild reverse sigmoid crossbar. Elastic fibers were scattered throughout the cartilage but were more concentrated in the center. The superficial gland of the third eyelid was compound tubuloacinar with serous acini.

Key words

Third eyelid, Histology, Morphology, Mongoose

Introduction

The third eyelid is a semilunar fold of the conjunctiva in the medial angle of the eye. It is supported by a T-shaped piece of cartilage. The crossbar (arms) supports the free margin of the lid and the appendix (shaft portion) extends to the nasal canthus and is wrapped by the superficial portion of the gland of the third eyelid. There is no deeper part of the nictitans gland in dogs and cats. The superficial part is a seromucus gland in dogs and a serous gland in cats (Cooper, 2010). On the bulbar surface of the third eyelid there is lymphoid tissue which encompasses ductule openings of the nictitans gland.

The cartilage of nictitating membrane is curved to conform to the convexity of the eyeball. The cartilage form varies in domestic animals as follows: elongated triangular in horses, leaf- or scoop-like in ruminants, anchor-like in pigs and crescent-like with a straight appendix in dogs and cats (Heine, 1909).

Various publications have reported the different aspects of the cartilage. According to the results of some authors, it consists of elastic cartilage in horses, pigs and cats and of hyaline cartilage in dogs and ruminants (Smollich and Michel, 1992). A

* Corresponding author. E-mail: gholami@shirazu.ac.ir

mixed character with some elastic portions was also described in the cartilages of cats and pigs (Czuberka, 1923).

The small Indian mongoose, *Herpestes javanicus* (Herpestidae), is native of Pakistan and northern India to southern China and the Malay Peninsula, as well as Hainan and Java and Iran and Iraq (Corbet and Hill, 1992). To the authors' knowledge, there is no morphological study regarding the third eyelid of the mongoose in veterinary literature. Therefore, the purpose of this study was to describe the gross anatomy of the cartilage and the histology of the superficial gland of the third eyelid in this species.

Material and methods

Two mongooses, in terminal stages of disease and near death due to aging or unknown reasons, were referred from Park Zoo (Shiraz, Iran) to the veterinary college of Shiraz University, Iran, and were used for this research. For relief, the animals were euthanized with ketamine 10% and xylazine 2%. The eyelids were incised parallel to their free edges. After removing them with the eyeball from the orbit, the third eyelids were cut off from their connections. For morphological studies of the cartilage of the third eyelid, a modified maceration technique according to Amselgruber and Kogel (1991) was used. The maceration was performed with 2% KOH up to a maximum of 18 hours at 40°C in incubator. After that the third eyelid was washed 3 times in TBS-buffer (pH 6.8) and was consequently conserved in 0.15% formalin. All the samples were investigated with a stereo-microscope equipped with a camera (Carl Zeiss, Jena, Germany). For histological examinations, the third eyelids were cut in half lengthwise and were fixed in 4% buffered formaldehyde solution, dehydrated through ascending grades of alcohol (70%, 80%, 90% and absolute alcohol), cleared in xylene and embedded in paraffin wax. Each sample was sectioned along the long axis in 5 µm thick slices. The sections were stained with haematoxylin and eosin. Also, to detect the elastic fibers in the cartilage, sections were stained with orcein and Weigert's resorcin-fuchsin. The stained sections were evaluated at the light microscope (Olympus BX51 equipped with Olysia Software, Tokyo, Japan) at magnification 5×, 10×, 40× and 100×.

Results

The crossbar of the cartilage shows a mild reverse sigmoid (S) shape with 2 approximately equal branches. The ends taper off to a point and a feeble bump is seen along the crossbar. The appendix with the average length of 1/12 mm is ovoid and enveloped by fat tissue (Fig. 1).

The elastic fibers are distributed throughout the cartilage. However, the density of the fibers in the center of the cartilage seems higher than at the periphery and beneath the perichondrium. These fibers are arranged in different directions (Figs. 2-4). The cartilage of the nictitating membrane is, therefore, elastic in nature.

The superficial gland of the third eyelid is a compound tubulo-acinar gland composed of secretory units (acini) and excretory ducts (Fig. 5). The cells forming the acini are tall pyramidal in shape, whereas the different ducts are lined by simple

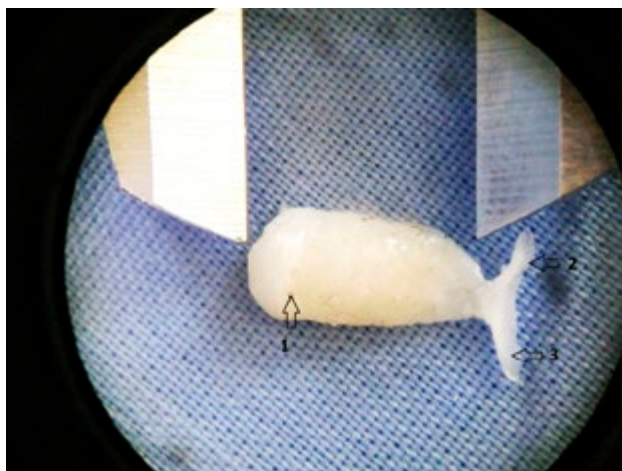


Figure 1 – Outline of the cartilage of the third eyelid in the mongoose. The appendix (1) is ovoid in shape and enveloped by fat tissue. The crossbar (2 and 3) of the cartilage shows a mild reverse sigmoid (S) shape with 2 approximately equal branches.

cuboidal, short columnar and bistratified columnar epithelium with large lumina. The acinar cells have deep basophilic, granular and vacuolated cytoplasm with round to ovoid nuclei located basally or centrally, as characteristic of serous cells.

Discussion

There are significant differences among domestic animals for the shape and histological type of the cartilage of the third eyelid. By many authors the cartilage is known as T-shaped or anchor-like (Dobberstain and Hoffmann, 1964; Franz, 1967; Sisson and Grossman's, 1975; Miessner, 1982; Constantinescu and Moore, 1998). In a study by Schlegel et al. (2001) it was found, however, that despite similarities there are important differences among the appendices of the cartilage of domestic animals. Very noticeable differences were also observed in the crossbar. In dogs the appendix is cone shaped at the basal end and extends to form a triangular plate. The crossbar is crescent-like in shape and has a marked bulge. The cartilage of cats consists of an appendix which is enlarged at the proximal end compared to that of dogs. The crossbar resembles a reverse S with ends tapering off to a point. The dorsal branch of the crossbar is double the size of the ventral one. In contrast, the cartilage of pigs and cows has a typical anchor-like shape whereas the cartilage of small ruminants starts with a thin rod which extends in a slightly curved form ending in an oval plate. The crossbar is crescent-like in these animals. In horses the base of the cartilage is surrounded by massive fat tissue and the crossbar has a characteristic hook-like form (Sclegel et al., 2001).

In the mongoose the crossbar resembles that of cats. But, contrary to cats, the dorsal and ventral branches are of approximately the same size.

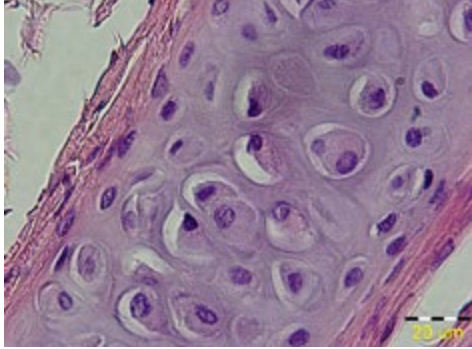


Figure 2 – Cartilage of the third eyelid. Haematoxylin and eosin. Bar = 20 µm.

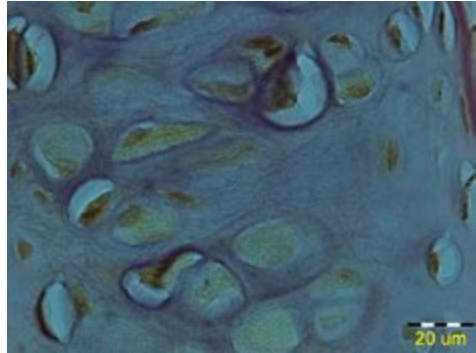


Figure 3 – Light micrograph showing the elastic fibers in the cartilage of the third eyelid of the mongoose. Weigert's resorcin fushin staining. Bar = 20 µm.

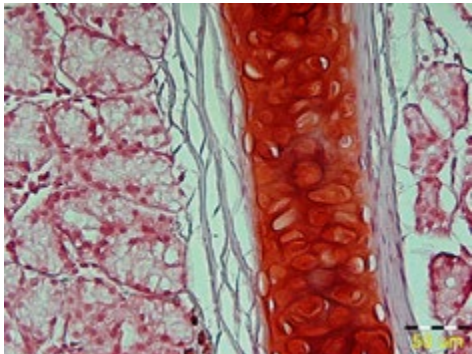


Figure 4 – Light micrograph showing the elastic fibers in the cartilage of the third eyelid of the mongoose. Part of the superficial gland of the third eyelid is shown on both sides of the cartilage. Orcein staining. Bar = 50 µm.

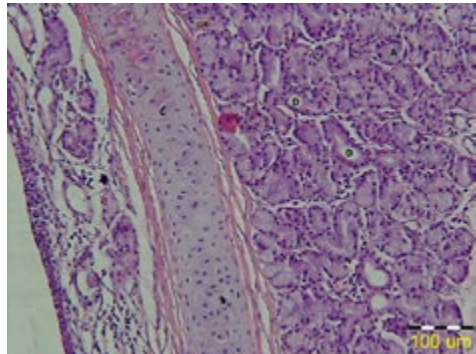


Figure 5 – Longitudinal section of the third eyelid of the mongoose at survey magnification. Note the acinar units (A) and excretory ducts (D) surrounding the cartilage (C). Haematoxylin and eosin. Bar = 100 µm.

In some studies, the presence and distribution of elastic fibers of the cartilage has been described in domestic animals. Among the investigated species, elastic fibers originating from the neighboring connective tissue were detected only in cats and horses. Additionally, the fine elastic fibers concentrated mainly in the central parts of the cartilage (Grau and Walter, 1967; Smollich and Michel, 1992; Schramm et al., 1994; Sclegel et al., 2001). Our study shows that in the mongoose the cartilage is elastic, similar to cats and horses.

The Harderian gland (deep gland of the third eyelid) is present only in some species. In species such as terrestrial carnivores, non-human primates and human beings this gland does not exist. However, it is well developed in most laboratory animals, amphibians, reptiles and birds (Seely, 1987; Sabry and Al-Ghaith, 2000). The lacrimal gland and the superficial gland of the third eyelid are tubulo-acinar in pigs, horses, guinea pigs, rabbits and primates (Kleckowska-Nawarot and Dziegiel, 2007; Kleckowska-Nawarot and Dziegiel, 2008; Ding et al., 2010; Schechtar et al., 2010). As mentioned above, the superficial gland of the third eyelid of the mongoose is a serous gland with a tubulo-acinar structure.

In conclusion, our investigation showed that in histological aspect, the third eyelid of the mongoose is more similar to cats than other domestic animals.

Acknowledgements

The authors would like to express their thanks to Dr. B. Rasouli and Mr. M. Babazadeh Joudi for their technical assistance. This research was financially supported by grants of Shiraz University Research Council.

References

- Amselgruber W.M., Kogel H.C. (1991) Construction of the vascular walls and angio-architecture of terminal blood vessels. In: Kogel H.C. *The Prosthetic Substitution of Blood Vessels. Actual State and Future Development.* Quintessenz-Verlags GmbH. Miinchen. Pp. 19-27.
- Constantinescu G.M., Moore C.P. (1998) Clinical anatomy of the eyelids for small animal practitioners. *WienTierärztl. Mschr.* 85: 229-232.
- Cooper S. (2010) The canine third eyelid. *Companion Animal* 15(6): 52-57.
- Corbet G.B., Hill J.E. (1992) *The Mammals of the Indomalayan region: a Systematic Review.* Oxford University Press, Oxford, UK.
- Czuberka R. (1923) *Ijber den Blinzknorpel einiger Säugetiere.* Dissertation, Tierärztliche Hochschule Wien.
- Ding C., Parsa L., Nandoskar P., Zhao P., Wu K., Wang Y. (2010) Duct system of the rabbit lacrimal gland: structural characteristics and role in lacrimal secretion. *Invest. Ophthalmol. Vis. Sci.* 51: 2960-2967.
- Dobberstein J., Hoffmann G. (1964) *Lehrbuch der vergleichenden Anatomie der Haustiere.* Bd. III. 2. Auflage. S. Hirzel. Leipzig. Pp. 190-191.
- Franz V. (1967) *H6here Sinnesorgane. Vergleichende Anatomie des Wirbeltierauges.* In: Bolk L., Goeppert E., Kallius E., Lubosch W. *Handbuch der vergleichenden Anatomie der Wirbeltiere.* Bd. II. Teil 2. Asher & Co. Amsterdam. Pp. 989-1292.
- Grau H., Walter P. (1967) *Grundriß der Histologie und vergleichenden mikroskopischen Anatomie.* Paul Parey. Berlin, Hamburg. Pp. 148-149.
- Heine E. (1909) *Untersuchungen über das dritte Augenlid der Haustiere.* Dissertation, Veterinärmedizinische Fakultät der Universität Bern.
- Kleckowska-Nawrot J., Dziegiel P. (2007) Morphology of the third eyelid and superficial gland on pig fetuses. *Anat. Histol. Embryol.* 36: 428-432.

- Klećkowska-Nawrot J., Dzięgiel P. (2008) Morphology of lacrimal gland in pig fetuses. *Anat. Histol. Embryol.* 37: 74-77.
- Miessner H. (1892) Die Drüsen des dritten Augenlides beim Schwein. *Z. Tiermed.* 18: 389-403.
- Sabry I., Al-Ghaith L. (2000) The Harderian gland of the Dhublizard *Uromastix microlepis* of the Kuwaiti desert: an ultrastructural approach. *Tissue Cell* 32: 71-78.
- Schechter J.E., Warren D.W., Mircheff A.K. (2010) A lacrimal gland is a lacrimal gland, but rodents and rabbits are not human. *Ocul. Surf.* 8: 111-134.
- Schlegel T., Brehm H., Amsegruber W.M. (2001) The cartilage of the third eyelid: A comparative macroscopical and histological study in domestic animals. *Ann. Anat.* 183: 165-169.
- Schramm U., Unger K., Keeler C. (1994) Functional morphology of the nictitating membrane in the domestic cat. *Ann. Anat.* 176: 101-108.
- Seely J.C. (1987) The Harderian gland. *Lab. Anim.* 16: 33-39.
- Sisson S., Grossman J.D., Getty R. (1975) *The Anatomy of the Domestic Animals*. W. B. Saunders Company. Philadelphia London Toronto. Pp. 228, 1745.
- Smollich A., Michel G. (1992) *Makroskopische Anatomie der Haustiere*. 2. Auflage. Gustav Fischer, Stuttgart Jena, Pp. 473-476.