Research Article

AL- ANBAR JOURNAL OF VETERINARY SCIENCES

Vol. 14 Issue:1, (2021) ISSN: P-1999:6527 E-2707:0603

Morphological and Histological Study of the Pineal Gland in local breed Goats (*Capra hircus*)

Hebat Allah M. Hameed* and khayreia K. Habeeb

Department of Anatomy &histology. College of Veterinary Medicine, University of Baghdad, Iraq.

Corresponding author; hebamajed870@gmail.com . Mobile:07822277830

Doi: https://doi.org/10.37940/AJVS.2021.14.1.7

Received: 2/11/2020 Revised:2/4/2021 Accepted:11/4/2021

This article is licensed under a CC BY (Creative Commons Attribution 4.0) http://creativecommons.org/licenses/by/4.0/.

Abstract

The current study was carried out to investigate both morphological and histochemical features of pineal gland in goat. Eight samples of pineal glands were used. The morphological study was involved the position, relation, weight, length and width of the glands. The tissue sections were prepared for paraffin embedding technique and stained with Hematoxyline & Eosin, Masson trichrom, Alizarin and PAS stains. The morphological result of pineal showed slightly irregular round-spherical shaped, grayish white in color. It located in the same mid-depression between the thalami body and two colliculi. The weight of gland was 0.478±0.02 mg and measured 2.10±0.06mm in length and 2.52±0.15mm in width. Histologically, the pineal gland has enveloped by a smooth thin layer of loose connective tissue capsule showed less trabiculae that carried the blood vessels into gland and the trabiculae have composed of fine collagen fibers. The glandular parenchyma showed marked lobular pattern of division with marked lighter central region which showed less cellular population. The glandular lobule consisted of three types of cells; Type I pinealocytes or dark pinealocytes, the second type was the less populated type II pinealocytes or light pinealocytes and the third type was the glial cell. The Alizarin stain was revealed multiple of variable size calcium patches within collagen fibers. The cytoplasmic contents of both pinealocytes types I & II showed no glycogen granules.

Keywards: Pineal Gland, Pinealocytes, Goat, Glial Cells, Histochemical.

دراسة شكليانية نسيجية للغدة الصنوبرية في الماعز المحلي البالغ

الخلاصة

أجريت الدراسة الحالية لبحث الخصائص المورفولوجية والكيميائية النسيجية للغدة الصنوبرية في الماعز. تم استخدام ثمانية عينات من الغدد الصنوبرية. اشتملت الدراسة المورفولوجية على موضع الغدد وعلاقتها ووزنها وطولها وقطرها. تم تجهيز أقسام الأنسجة لتقنية تضمين البارافين وصبغتها ببقع الهيماتوكسيلين والأيوزين وماسون ترايكروم والأليزارين و PAS. أظهرت النتيجة المورفولوجية للصنوبرية شكل كروي دائري غير منتظم قليلاً ، ولون سنجابي. يقع في نفس منتصف الاكتئاب بين الجسم المهاد واثنين من القولون. كان متوسط وزن الغدة 0.478 ± 0.02 مجم وقطرها 2.10 ± 0.06 مم 2.5 عرض 2 ± 0.15 مم. من الناحية النسيجية ، تم تغليف الغدة الصنوبرية بطبقة رقيقة ملساء من كبسولة الأنسجة الضامة الرخوة والتي أظهرت عددًا أقل من الترابيق الذي حمل الأوعية الدموية إلى الغدة ، كما أن الترابيق يتكون من ألياف كولاجين دقيقة. أظهرت الحمة الغدية نمط انقسام مفصص ملحوظ مع منطقة مركزية أفتح بشكل ملحوظ والتي أظهرت عددًا أقل من الخلايا. تتكون الفصيص الغدي من ثلاثة أنواع من الخلايا ؛ النوع الأول من الخلايا الصنوبرية أو الخلايا الصنوبرية المظلمة ، والنوع الثاني هو النوع الثاني الأقل كثافة بالسكان من الخلايا الصنوبرية أو الخلايا الصنوبرية الخفيفة وكان النوع الثالث من الخلايا هي الخلايا الساندة. كان هناك العديد من بقع الكالسيوم متغيرة الحجم داخل ألياف الكولاجين. المحتويات السيتوبلازمية لكلا النوعين الأول والثاني من الخلايا الصنوبرية لم تظهر أي حبيبات جليكوجين.

Introduction

A pinecone shaped gland belonging to the endocrine system is the pineal gland. It is situated on the midline of the cerebrum, connected to the third ventricle's posterior end of the roof. (1&2). It is found to be situated below the cerebral hemispheres laterally. It tends to appear in front of the cerebellum as a slight grayish hump. The gland varies in size among species; it is around 1 cm in length for camels and humans. (3,4 and 5). Whereas it is much smaller in dogs, it is not longer than 1 mm. (6), The pineal gland produces several important hormones including melatonin. Melatonin influences sexual development and sleep-wake cycles. The pineal gland is a small endocrine gland in the brain of most vertebrates shaped like a pinecone, the pineal gland located in the midbrain and it's caudal part is almost in contact with the third ventricle (7). This gland in mammals is a neuroendocrine organ that exhibits a circadian rhythmicity in its secretion of hormone melatonin (8, 9, 10, 11, 12, 13 and 14). The gland is composed of pinealocytes, astrocytes and glial cells (15). Pineal gland acts as neuroendocrine glands which secrete melatonin and other enzymes that help to regulate the animal's circadin rhythem.

Materials and methods

Eight samples of pineal glands of healthy adult goat (1.5-2 years) were used for this study. The animals' heads were immediately removed of the carcass after slaughter and brains were dissected for achieving the following pineal glands position and relation. Then the glands were removed out to record the morphological parameters; length, width by vernia caliber, weight by digital electronic balance and color. For histochemical study the samples of pineal glands were immediately washed up with normal slain and fixed with neutral buffer formalin 10% (16). The specimens were processed as the paraffin embedding technique. The tissues were sectioned at 5µm by rotatory microtome and the tissue sections were stained with Hematoxyline & Eosin, Masson trichrom to demonstrate the type of connective tissue, Alizarin stain for detection the calcium deposition and PAS stain for detection the presence of intra cytoplasmic glycogen content. The morphological images were examined by stereoscope and histological images were captured by digital camera 5 Mega pixel, Future Win Joe China.

Result and Discussion

Morphologically, goats pineal had slightly irregular round-spherical shaped, grayish white in color and were located in the mid-depression between the thalami body and two colliculi. The thalamic body "Massa" represented the anterior border of the pineal gland while the superior coliculus and inferior coliculus were represented the caudal border of the pineal gland. Dorsally the pineal gland was covered by the splenuim of the corpus callosum (fig.1 & 2). This result was differing from other animals like buffalo which have pea-shape according to (18) weather in horses &camels it was fusiform in shape (19 and 20) in the human pineal gland have a pine-shaped according to (21).According to Rasha *et al.,* (2016) and Vollrath (1981) the tonsillar-like aspect differs from the general bat-shape found in rodents. Branco (1997) showed that doges pineal gland shaping from conic to tongue-like (concerning the human tongue). With regarded to the location our study showed that the pineal gland in the deep mid-depression between the thalami body and two colliculi and this generally agree with all studies on mammals (18, 19 and 24).). In local breed goat the mean weight of gland was 0.478±0.02 mg and measured 2.10±0.06 mm in length and 2.52±0.15mm in width, the pineal gland weight of goat was smaller than what noticed by other researchers whom study on Buffalo, camel and horse and more heavily than another animals like fox, dog and

rodents (25 and 26). The size of pineal gland in local breed goat was closely related and was between (2.52-2.73mm) this result was the first study for illustration of morphology of pineal gland ,there's no studies on large scale except very little papers restricted morphometric data of a few species of animals most studies where found on human (27).and large ruminant(28).Also recent studies was very few concerning morphology of pineal gland (29), so the comparison between morphology in general have a very difficult distinct and this is due to the large differences between sizes of animal

Histologically, the pineal gland in goat was enveloped by thin layer of loose connective tissue capsule of pia matter which revealed fewer trabeculae. The insertion of trabeculae into glandular parenchyma was showed fewer of glandular surface indentations with marked nerve stalk. The inserted of trabeculae were carried the blood vessels into gland and have composed of fine collagen fibers (fig. 3 & 4). This result was differing from another animals like buffalo the gland which is covered with dense capsule that infiltrates the entire parenchyma with their trabeculae according to (25), but in donkey the gland is surrounded by well vascularized connective tissue capsule and the greater part of blood vessels are seen in the trabeculae of the of the connective tissue (24).In foals the gland connective tissue is denser compared to that adult horses (19), but in camel the pineal gland is surrounded by thick fibrous connective tissue capsule according (20).

The parenchyma of the goat's pineal gland was showed marked lobular pattern of division with marked lighter central region which showed less cellular population (fig. 5). The glandular lobule of goat pineal gland was more obvious and there were marked demarcation among the lobule that related for less of cellular population of pinealocytes (fig.6). This result was similar that recorded by (20) in female camel cells of pineal glands are ovoid to round shape and appeared light to dark brown in color. The current result was showed three types of cells in the pineal gland; Type I-pinealocytes (Dark pinealocytes) which were irregularly shaped with peripheral processes, type II-pinealocytes (Light pinealocytes) which were had large round nuclei the third type of cell was the neuroglia cells or interstitial cells (glial cells) which were possessed round, oval or cone-shaped nucleus with round or oval strongly basophilic nuclei, or elongated nuclei closely associated with the blood capillaries and a few cells with large nuclei (fig.7). This result was similar that recorded by (20) in female camel, also some of these cell showed a higher amount of pigment content in their cytoplasm according to viscacha,(30), and showed 2 types of cells one which have around or oval nucleus contained prominent nucleolus (type I cells or pinealocytes of population I), presented an irregular shaped nucleus (type II cells or pinealocytes of population II). Light dark pinealocytes distinguished with in type I cell. Some of glial cells and pigmented cells are also observed. The pineal parenchyma of the pregnant females presented major amount of dark pinealocytes according to viscacha (31). The pinealocytes had large round nuclei with a remarkable loose chromatin and a weak basophilic cytoplasm, while typical astrocytes intermingled them according to (32). So in goat agree with the idea about the unique behaviors that seen in camels have especially that concerning to some physical properties (20). Also the goat pineal gland cytoarchetecture were similar that seen in bird (turkey pineal gland) which consist of 3 types of cells (33). In the albino rat cells differs from small cells with little cytoplasm and small darkly stained nuclei to large cells with abundant cytoplasm & large pale nuclei with prominent nucleoli according to, (34). The results showed multiple of variable size calcium patches within glandular parenchyma and the magnified section revealed oval concentrated calcium particles between pinealocytes and

Research Article
AL- ANBAR JOURNAL OF VETERINARY SCIENCES
Vol. 14 Issue:1, (2021) ISSN: P-1999:6527 E-2707:0603

within collagen fibers (fig. 8 & 9). The result of PAS stain showed that the cytoplasmic contents of both pinealocytes types I & II showed no glycogen granules (fig.10). Goat's pineal gland had the pineal stalk which has composed of dense nerve tract enclosed by pia matter (fig.11). .

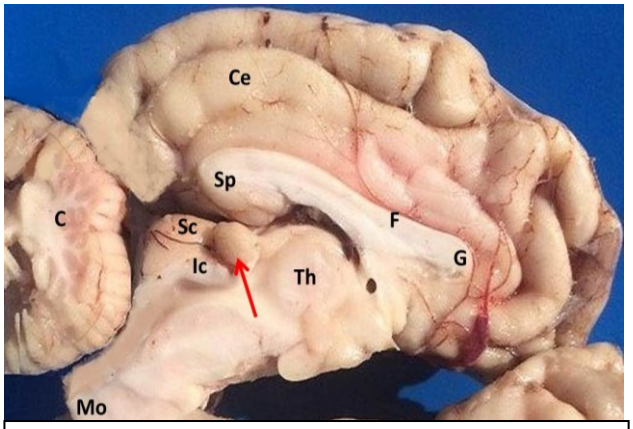

Figure 1; Sagittal section of brain goat shows: cerebellum (C). Cerebrum (Ce). Medulla oblongata (Mo). Thalamic body 'Massa" (Th). Superior coliculus (Sc). Inferior coliculus.

Figure 2; Macrograph of goat pineal gland after dissecting and removing from brain

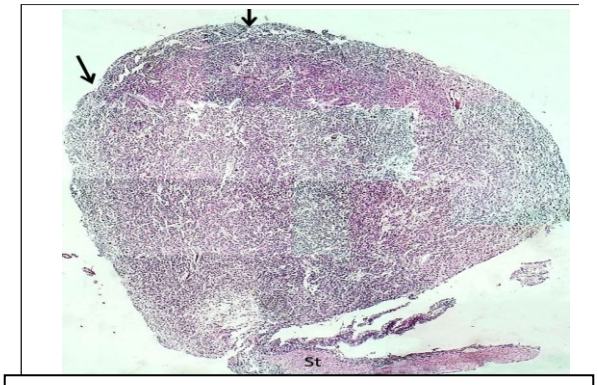

Figure 3: Histological section of the goat pineal gland shows: less of capsule indentations (Black arrows) & pineal gland stalk (St). H&E

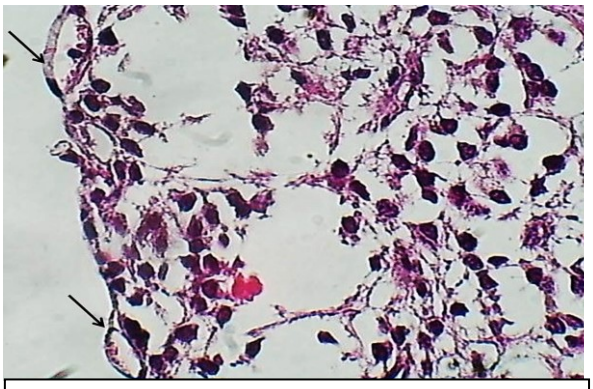

Figure 4; Histological section of the goat pineal gland shows: thin pia matter capsule (Black arrows) . H&E stain.400x

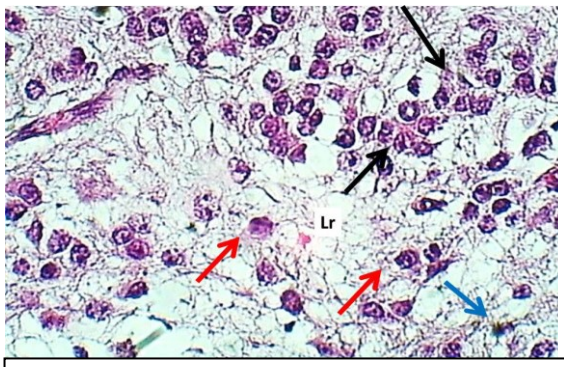

Figure 5; histological section of the goat pineal gland shows: lighter central region (Lr), dark pinealocytes (Black arrows), light pinealocytes (Red arrows) and glial cells (Blue arrow) . H&E stain.400x

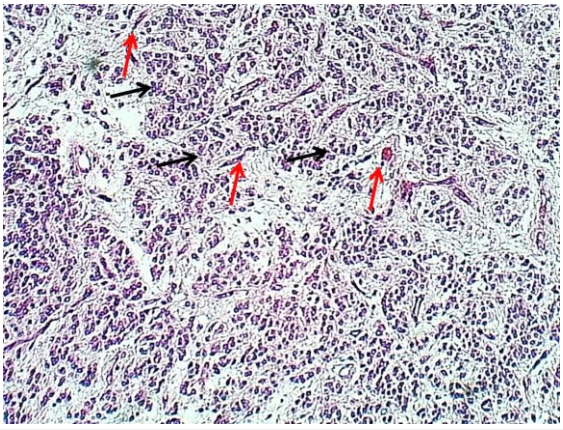

Figure 6 ; histological section of goat pineal gland capsule shows: lobule (Black arrow) & thin trabiculae (Red arrows). H&E. stain. 100x

Research Article
AL- ANBAR JOURNAL OF VETERINARY SCIENCES
Vol. 14 Issue:1, (2021) ISSN: P-1999:6527 E-2707:0603

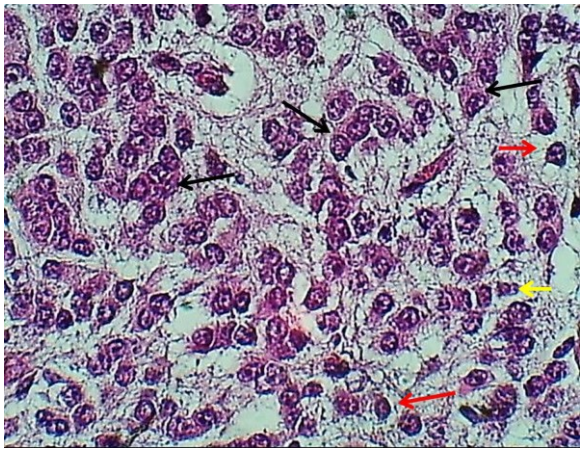

Figure 7; Magnified histological section of goat pineal gland shows: Dark pinealocytes (Black arrows), light pinealocytes (Red arrows) & glial cells (yellow arrow) H& E stain. 400x

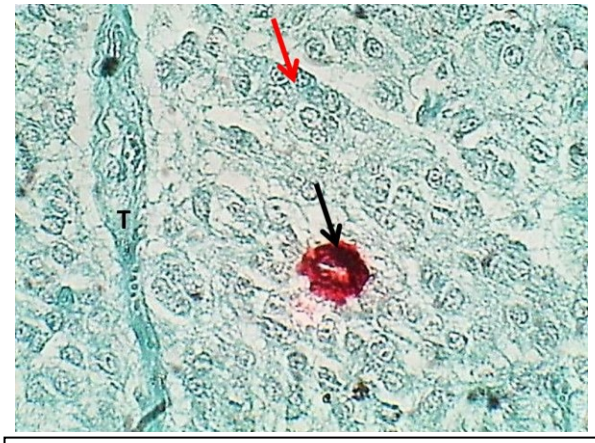

Figure 9; histological section of goat pineal gland lobule shows: multiple small of variable size calcium patches (Black arrow), pinealocytes (red arrow) trabecula (T).Alizarin stain. 400x.

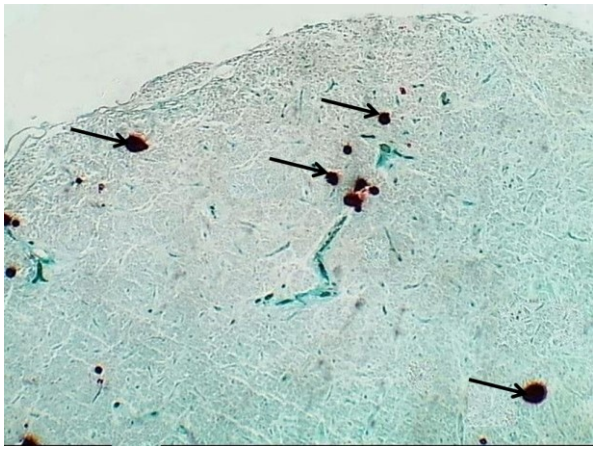

Figure 8; histological section of goat pineal gland shows: multiple small of variable size calcium patches (Arrows).Alizarin stain. 40x.

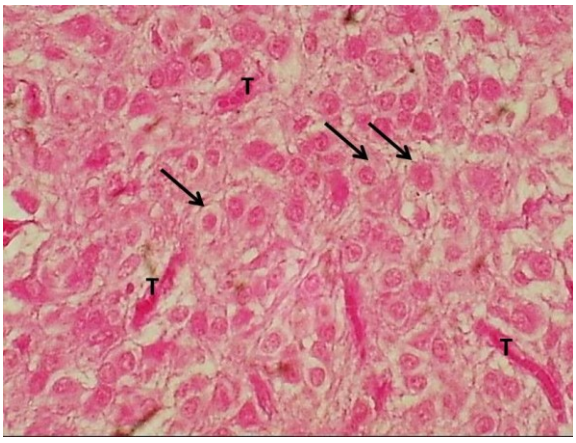

Figure 10; histological section of goat pineal gland lobule shows: No intracytoplasmic glycogen granules within pinealocytes type I&II (Arrows), trabeculae (T). PAS stain. 400x.

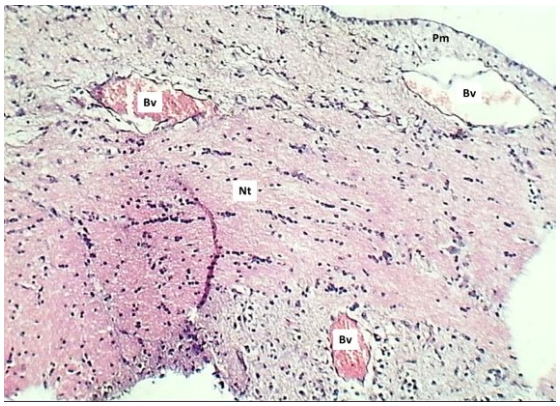

Figure 11 ; histological section of goat pineal gland stalk shows: pia matter (Pm), blood vessels (Bv), nerve tract (Nt). H&E stain. 400x.

Conclusions

The pineal gland of goat was similar to that other special with small ruminant with limited morphometric differences.

References

1. Snell RS. Clinical and functional histology for medical students. Little Brown & Company; 1984.
2. Hamza, LO. Some anatomical and histological observation on the pineal gland in an endogenous moorhen (gallinule chloro). Basrah journal of veterinary research.2007;Oct 6 (2):22-29.
3. Abbas AA, Ewais MS. Histological and histochemical studies of the pineal gland of the one humped camel (Camelus dromedarius). Journal of Egyptian Veterinary Medical Association. 1982;42(2):117-24.
4. Hasegawa A, Ohtsubo K, Mori W. Pineal gland in old age; quantitative and qualitative morphological study of 168 human autopsy cases. Brain research. 1987 Apr 21;409(2):343-9.
5. Galliani I, Frank F, Gobbi P, Giangaspero F, Falcieri E. Histochemical and ultrastructural study of the human pineal gland in the course of aging. Journal of submicroscopic cytology and pathology. 1989 Jul 1;21(3):571-8.
6. Abu-Easa KF. Tissue changes in the pinel, petina and thymus of male dog before and after sexual maturity. Alexandria Journal of Veterenary Science (Egypt). 1997.
7. Cozzi B. Cell types in the pineal gland of the horse: an ultrastructural and immunocytochemical study. The Anatomical Record. 1986 Oct;216(2):165-74.
8. Hales JR, Fawcett AA. Wool production and blood supply to skin and other tissues in sheep. Journal of Animal Science. 1993 Feb 1;71(2):492-8.
9. MIGUEZ JM, MARTIN FJ, LEMA M, ALDEGUNDE M. Changes in serotonin level and turnover in discrete hypothalamic nuclei after pinealectomy and melatonin administration to rats. Neurochemistry international. 1996 Dec 1;29(6):651-8.
10. Guimaraes MA, Lunardi LO, Pellizzon CH, Kempinas WG. Male gonadal denervation by guanethidine at pre-puberty: different doses, different results. The intervention of the pineal deafferentation. Cellular and molecular biology (Noisy-le-Grand, France). 1997 May;43(3):383-91.
11. Khan JR, Dang AK, Singh M. Role of pineal in lactation. *International Journal Of Animal Sciences*.1997; *12*, pp.209-212.
12. Pacchierotti C, Iapichino S, Bossini L, Pieraccini F, Castrogiovanni P. Melatonin in psychiatric disorders: a review on the melatonin involvement in psychiatry. Frontiers in neuroendocrinology. 2001 Jan 1;22(1):18-32.
13. Kus I, Sarsilmaz M, Ozen OA, Turkoglu AO, Pekmez H, Songur A, Kelestimur H. Light and electron microscopic examination of pineal gland in rats exposed to constant light and constant darkness. Neuroendocrinology Letters. 2004 Feb 1;25(1-2):102-8.
14. Reiter RJ. The melatonin rhythm: both a clock and a calendar. Experientia. 1993 Aug 1;49(8):654-64.
15. Boya J, Calvo JL, Rancano D. Structure of the pineal gland in the adult cat. Journal of pineal research. 1995 Mar;18(2):112-8.
16. Bancroft JD, Gamble M, editors. Theory and practice of histological techniques. Elsevier health sciences; 2008.
17. Luna LG. Manual of Histological staining method of Armed forces institute of pathology, third Ed. Graw.1968.
18. Hill Book G. New York. Pp. 4-9, 158 – 169, 34.
19. Abou-Easa K, Tousson E, Abd-El-Gawad M. Involution signs during the postnatal life in the pineal tissue of buffalo and camel. *Nature and Science*.2009; *7*(9):35-44.
20. BOLAT, D., KÜRÜM, A., Bahar, S. and Karahan, S., 2018. Histomorphometric examination of the pineal gland in foals and adult

horses. Veteriner Fakültesi dergisi.2018; 65(2):205-212

21. Rasha R, Beheiry1, Attia AM. Macro and Microscopical Studies On The Pineal Gland Of Camel Wit immunohistochemical localization to Pinealocytes and Glia Cells Markers. Int. J. Adv. Res.2016; 4(8), 1154-1163.

22. Vollrath L. The pineal organ. *Handbuch der mikroskopischen Anatomie des Menschen*;1981;*7*: 288-304.

23. Branco ER, Guimarães A, Miglino MA, Didio LJ, Nurmberger Jr R, de Souza WM. Pesquisa anatômica da glândula pineal em capivaras (Hydrochoerus hydrochoeris). *Brazilian Journal of Veterinary Research and Animal Science*.1997;*34*(4):191-195.

24. Gomes LA, de Santis Prada IL, Farias EC, Zanon JP, Headley SA, Blazquez FX. Estudo morfológico da glândula pineal do cão. Semina: Ciências Agrárias. 2008;29(1):137-50.

25. Ebada S. Morphological and Immunohistochemical Studies on the Pineal Gland of the Donkey (Equus asinus). Journal of veterinary anatomy. 2012 Apr 1;5(1):47-74.

26. de Carvalho AF, Ambrósio CE, Miglino MA, Mançanares CA, Blazquez FJ. Macro-microscopical aspects of the buffalo (Bubalus bubalis Linnaeus, 1758) pineal gland. Biotemas. 2009 Aug 11;22(2):127-35.

27. Legait H, Legait JF. Statistical weight study of the human pineal body, hypophysis and brain. Bulletin de L'association des Anatomistes. 1980 Sep 1;64(186):399-414.

28. Nickel R, Schummer A, Seiferle E, Frewein J, Wilkens H, Wille KH, Siller WG, Stokoe WM. The anatomy of the domestic animals. Volume 1. The locomotor system of the domestic mammals. Verlag Paul Parey; 1986.

29. Lalitha PS, Seshadri VK. Histology And Histochemistry Of Degenerative Changes In The Pineal-Gland Of Adult Indian Buffalos (Bubalus-Bubalis). Indian Veterinary Journal. 1992 May 1;69(5):427.

30. Busolini FI, Rodríguez GB, Filippa VP, Mohamed FH. Pigmented cells in the pineal gland of female viscacha (Lagostomus maximus maximus): a histochemical and ultrastructural study. International journal of endocrinology. 2017 Dec 17;2017.

31. Gil E, Calderón C, Pelzer L, Domínguez S, Fogal T, Scardapane L, Piezzi RS. Morphological and biochemical study of the pineal gland of pregnant and non-pregnant female vizcachas (Lagostomus maximus maximus). Neuroendocrinology Letters. 2005 Oct 1;26(5):575-80.

32. Lazăr R, Solcan C, Creţu C, Lazăr M, Muntean C, Boişteanu PC. Characterization of the relations between morphology and physiological status of the pineal gland in connection with the somatic development level in turkeys reared in Romania. Arquivo Brasileiro de Medicina Veterinária e Zootecnia. 2015 Jun;67(3):763-70.

33. Banks WA, Kastin AJ. Characterization of lectin-mediated brain uptake of HIV-1 GP120. Journal of neuroscience research. 1998 Nov 15;54(4):522-9.

34. Kalinina S, Ilyukha V, Uzenbaeva L. Pineal Gland Morphology in Relation to Age and Season in Three Canidae Species. Journal of Morphological Sciences. 2019 Dec;36(04):247-54.

35. EI-Fadaly A. Cellular Constituents Of The Pineal Parenchyma In The Albino Rat: Effect Of Different Photoperiods. The Egyptian Journal of Anatomy. 2004 Jul 1;27(2):1-29.