ISSN:1991-8178

Australian Journal of Basic and Applied Sciences

Journal home page: www.ajbasweb.com

Morpho-quantitative and Microstructural description of the pineal gland of Pigs

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ARTICLE INFO

Article history:
Received 6 March 2015
Accepted 25 April 2015
Published 9 May 2015

Keywords:
Pinealocytes, gliocytes, mast cells, Sus crofa domesticus

ABSTRACT

Background: The pineal gland is an active endocrine organ that has a great importance for animals. This organ is involved in the circadian control and other bodily functions, such as the reproductive cycle, thermoregulation, cardiovascular regulation,, pressure and regulating the immune system. **Objective:** Assess the pineal gland of male six-month-old pigs, establishing a relationship between macroscopic aspects such as organ size - length and width, with microscopic aspects, both qualitatively and quantitatively, for standardization of pineal gland constituents. **Results:** Gland average length and width were 6.98±0.35mm and 3.52 ± 0.18mm, respectively. Average number of pinealocytes and gliocytes were 19.83 ± 0.4% and 1.89 ± 0.11%, respectively. Mast cells, reached 11.39 ± 0.92%. Connective tissue average was 66.32 ± 0.68%. **Conclusion:** It was checked pinealocytes over the entire gland parenchyma; connective tissue revealed to surround the gland forming a capsule-like, which sent out projections and formed incomplete lobes and septa; there was no formation of calcareous concretions in any sampled animal; gliocytes were mainly observed along blood vessels. It is noteworthy mention that there was no well-defined pattern concerning gland length and width for pigs.

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To Cite This Article: Júlia M. da Silva, André R. C. Barreto-Vianna, Anna C. dos Santos, Kaique Nogueira, Eduardo M.M. Lima, Morpho-quantitative and Microstructural description of the pineal gland of Pigs. ***Aust. J. Basic & Appl. Sci.,*** *9(11): 366-370, 2015*

INTRODUCTION

Pineal gland originates from diencephalon and is located on the roof of the third ventricle, between the two cerebral hemispheres. It is an active endocrine organ and connected to the retina by retinol-hypothalamic projections via the suprachiasmatic nucleus (COMMENTZ & HELMKE, 1995). The gland secrets melatonin, an indole amine; this hormone is involved in the circadian control and other bodily functions, such as the reproductive cycle, thermoregulation (TILDEN & HUDSON, 1993), cardiovascular regulation, especially blood pressure and regulating the immune system. The importance of melatonin varies according to the ontogenetic development, i.e. according to species this hormone is produced in larger or smaller amount, playing a greater role on the circadian cycle (LANGER *et al*., 1997).

Reproductive activity is directly related to environmental conditions, such as food availability and adequate temperatures, so being fostered when there are favorable conditions for offspring birth (GOLDMAN, 2001). In short-day species, such as goats and sheep, the increase in melatonin production by pineal gland stimulates secretion of gonadotropin-releasing hormone (GnRH). Yet long-day animals, as mares, that increase will inhibit GnRH secretion by the hypothalamus. Thus, differences in day length are recognized and converted into signals capable of turning on or off the sexual activity on a species-specific manner (SRINIVASAN *et al*., 2009).

Since this gland has great importance for animals, especially mammals, this research aimed to assess the pineal gland of male six-month-old pigs. Establishing a relationship between macroscopic aspects such as organ size - length and width, with microscopic aspects, both qualitatively and quantitatively, for standardization of pineal gland constituents.

MATERIAL AND METHODS

For this study, it was assessed 12 pineal glands of male six-month-old Camborough 22 pigs from Sabuguy slaughterhouse in Brasília-DF, Brazil. Immediately after slaughter, encephalon was removed with meninges rejection, and then the gland was isolated and removed by sagittal section, followed by its measurement. The glands were kept in 10% formalin aqueous solution for 48 hours and, after this, they were subjected to conventional

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histological technique, and subsequently fixed in paraffin. Next, histological cuttings were made by handheld microtome (Leica RM2125RT; Leica Microsystems) at 4µm thickness and stained with hematoxylin-eosin, picrosirius red and tolonium chloride for quantification of gliocytes, pinealocytes, connective tissue and mast cells, respectively.

Sections were examined under an optical microscope (Olympus BX51, Olympus Optical LTD, Tokyo, Japan) coupled to an image capture and analysis program (ProgRes® Capture Pro 2.5); then, it was obtained photomicrographs at 20 different and random fields. Then, pinealocyte, gliocyte and mast cell rates, calcareous concretion presence and connective tissue aspect were analyzed using a STEPanizer® program through a test-system with 100 points. The data underwent Kolmogorov-Smirnov normality test and by means of descriptive analysis, means + standard deviation values were obtained. Moreover, it was applied Pearson's correlation test, aiming to determine relationships between macro and microscopic dimensions.

RESULTS AND DISCUSSIONS

The pineal gland length and width were 6.98 ± 0.35 mm and 3.52 ± 0.18 mm, respectively. Table 1 sows the stereological analysis results for pinealocytes, gliocytes, mast cells and connective tissue. From microscopic analysis, it was found that no calcareous concretions were found in any of the pineal glands. Otherwise, it was shown that the pia mater connective tissue (Figure 3) covered the gland and send out into parenchyma projections that form incomplete septa of this tissue. Gliocytes elongations established a frame for other cells, being more pronounced closer to blood vessels. It was applied Person's linear correlation test from both macro and microscopic analyses (Figures 1 and 2), seeking for a possible relationship between gland size data and structural parameters observed in this study. Therefore, it was possible to state positive and negative weak correlations. Even correlations of the gland length (6.98 ± 0.35 mm) with average number of pinealocytes (r = -0.04), gliocytes (r = 0.03), mast cells (r = -0.20) and connective tissue (r = 0.58). Moreover, correlations of width (3.52 ± 0.18 mm) with average number of pinealocytes (r = 0.48), gliocytes (r = -0.29), mast cells (r = 0.26) and connective tissue (r = 0.07) all over the entire gland.

Table 1: Absolute values obtained by quantification of structural components of pineal gland of swines. Std.: standard.

STRUCTURES	MEAN ± STD. DEVIATION
Pinealocytes	19,83 ± 0,4%
Gliocytes	1,89 ± 0,11%
Mast cells	11,39 ± 0,92%
Connective tissue	66,32 ± 0,68%

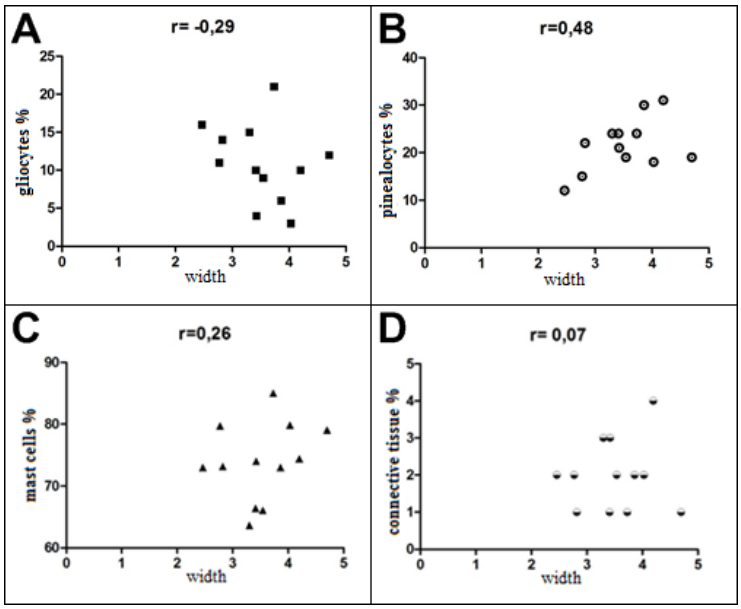

Fig. 1: Representative graphs obtained by Pearson correlation between the macroscopy width of the pineal gland and quantification of gliocytes, pinealocytes, mast cells and connective tissue.

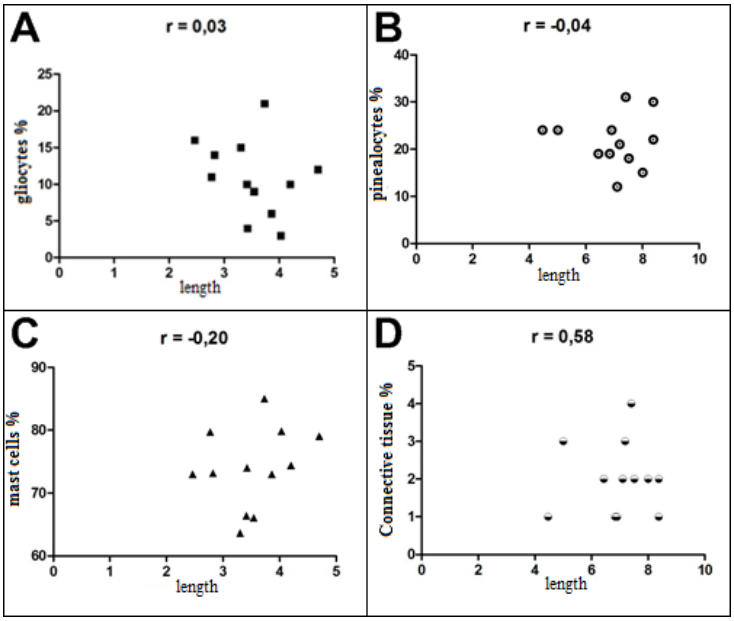

Fig. 2: Representative graphs obtained by Pearson correlation between the macroscopy length of the pineal gland and quantification of gliocytes, pinealocytes, mast cells and connective tissue.

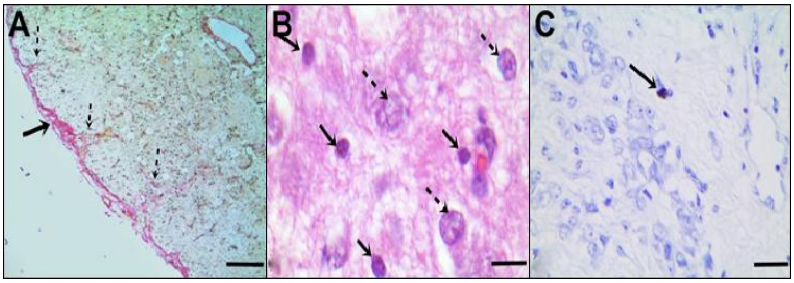

Fig. 3: Optical fotomicrography of swine's parenchyma pineal gland ; A) Connective tissue capsule (arrow) and its septums (dashed arrow) colored by Picrosirius Red, 10 µm. B) Pinealocytes (branched arrow) and gliocytes (arrow) colored by Hematoxiline-eosine, 10 µm. C) Mast cell (seta) colored by Tolonium Chloride, 10 µm.

Pig pineal gland length and width were 6.98 ± 0.35 mm and 0.18 ± 3.52 mm, respectively; otherwise, Lima *et al*. (2011) found length of 0.79 ± 6.98mm and width of 6.40mm ± 1.35 in Santa Inês sheep. Carvalho *et al*. (2009) reported length of 8.8 mm ± 1.0 and width of 7.9 mm ± 0.8 in buffaloes. Yet, according to Marques *et al*. (2010), who evaluated *Procyon cancrivorus*, the gland length and width were 7.5 mm and 4.5 mm, respectively. Given these data, plus the current findings, it was possible to infer that the gland dimensions had not great specificity. Pineal gland size can change according to reproductive period alterations of the species and, in addition to the animal size, or even in accordance with brain dimensions.

Both pineal gland length and width macroscopic data did not present, in relation to the quantified structures, a well-defined pattern in pigs (Figures 1 and 2). Such outcome suggests other factors that may influence the organ's building, among which it can be cited age, reproductive cycle and even the species.

Connective tissue in pineal glands of swines showed capsular aspect (Figure 3) arranged on the gland surface, being closely related to pia mater. The tissue besides evolving and delimiting, it also send out projection into the gland parenchyma that ascertained incomplete septa and composed two distinct lobes. A large number of blood vessels were seen along the tissue, which was also reported by Carvalho *et al*. (2009) and Marques *et al*. (2010), in buffaloes (*Bubalus bubalis*) and *Procyon cancrivorus*, respectively. In pigs, the glands were involved by pia mater that responded by issuing emissions, which formed complete lobes (LIMA *et al*., 2003). Such arrangement was not observed in Santa Inês sheep, which was featured by a capsule around the gland without septum or lobule formations. In the face of literature findings and reports, it is possible to state the existence of various arrangements, mostly with respect of the given species.

Regarding the number of pinealocytes, swines had 19.83 ± 0.4%; other way, in Santa Inês sheep, it was 86.28 ± 40.87% per field (LIMA *et al*. 2011). Such distinct values revealed a quantitative characterization trend for each animal species. Nevertheless, it differed from goat findings, in which there was a decrease in this number with age, highlighting that this cell type volume increases during the first months of the animal life (NOWICKI & PRZYBYLSKA, 2006).

Calcareous concretions were not present in any of the studied samples, what can be due to animal age of six months. According to Marques *et al*. (2010), the same was related for *Procyon cancrivorus*, as well as for dogs as reported by GOMES (2003). However, these concretions were observed in 28.57% Santa Inês sheep (LIMA *et al*., 2011). Based on the referred literature, it can be concluded that such phenomenon is firstly outlined by age (CIPOLLA NETO, 1996). These concretions in pineal gland may play a dynamic calcium stock (LIMA *et al*., 2003). In 180-day-old pigs, concretions resembling morula-like agglomerations, which somehow justifies the use of swines as experimental model for pineal gland (LIMA *et al*., 2003).

Mast cells were found in all evaluated animals, what has not match the findings of Lima *et al*. (2011), who had verified these cells in 14% of the evaluated Santa Inês sheep. The amount of mast cells in swine was 11.39 ± 0.92%, and it was arranged randomly throughout the pineal gland parenchyma. This outcome has coincided with the report by Barros (2006), who stated that mast cells are globular-shape structures with no visible elongation full of metachromatic grains arranged randomly or in small-sized agglomeration in the gland. On the other way, Mançanares *et al*. (2007) observed mast cells on pineal gland peripheral region. Furthermore, Marques *et al*. (2010) and Carvalho *et al*. (2009) reported these cells in *Procyon cancrivorus* buffaloes, respectively. With respect to gliocytes, it was obtained a number of 1.89 ± 0.11% in pigs (Table 1), which were often found arranged along blood vessels. It could be understood that as a component of the nervous tissue, these cells showed numerous elongations and several intermediate strands, composing about 5% of the gland parenchyma (JUNQUEIRA & CARNEIRO, 2004); playing then an important role in this gland arrangement.

Conclusion:

These findings enhance understanding of pineal gland, especially for pigs of the Camborough 22 lineage, allowing the comparison to other species. It was checked pinealocytes over the entire gland parenchyma. Connective tissue revealed to surround the gland forming a capsule-like, which sent out projections and formed incomplete lobes and septa. There was no formation of calcareous concretions in any sampled animal. Gliocytes were mainly observed along blood vessels. It is noteworthy mention that there was no well-defined pattern concerning gland length and width for pigs.

REFERENCES

Barros, R.A.C., 2006 Anatomia macroscópica e microscópica da glândula pineal do macaco *Cebus apella*. 2006. São Paulo: Universidade de São Paulo, 2006. 72p. Tese (Pós Graduação em Anatomia dos animais domésticos) Faculdade de Medicina Veterinária e Zootecninia, 2006.

Callegari-Jacques, S.M., 2003. Bioestatística: Princípios e Aplicações, Artmed, Porto Alegre, pp: 86-91.

Carvalho, A.F., C.E. Ambrósio, M.A. Miglino, C.A.F. Mançanares, F.J.H. Blazquez, 2009. Macro-microscopical aspects of the buffalo (Bubalus bubalis Linnaeus, 1758) pineal gland. *Biotemas*, *22*(2): 127-135.

Cipolla, J.N., 1996. Controle neural do metabolismo da glândula pineal. 1996. Tese (Livre docência) – Departamento de Fisiologia e Biofísica, Instituto de Ciências Biomédicas, Universidade de São Paulo, São Paulo.

Commentz, J.C., K. Helmke, 1995. Precocious puberty and decreased melatonin secretion due to a hypothalamic hamartoma. Horm Res., 44(6): 271-275.

Erhart, E.A., 1962. Neuroanatomia. 2ed, Atheneu, São Paulo.

Goldman, B.D., 2001. Mammalian photoperiodic system: Formal properties and neuroendocrine mechanisms of photoperiodic time measurement. *Journal of Biological Rhythms*, 16(4): 283-301.

Gomes, L.A., 2003. Estudo morfológico da glândula pineal no cão. Tese (Mestrado em Anatomia dos Animais Domésticos e Silvestres), Departamento de Cirurgia, Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, São Paulo.

Junqueira, L.C., J. Carneiro, 2004. Histologia básica. Glândulas endócrinas, Guanabara Koogan, Rio de Janeiro, pp: 390-414.

Langer, M., J. Hartman, H. Turkof., F. Waldhauser, 1997. Melatonin in the Human - an overview. *Wien Klin Wochenschr*, 109(18): 707-13.

Lima, E.M.M., M.I. Santana, M.B. Castro, P.M. Ferreira, A.R.C.B. Vianna, 2011. Estudo morfoquantitativo e da microestrutura da glândula pineal em ovinos Santa Inês, *Ars Veterinária*, 27(3): 186-191.

Lima, L.C.M., K.F. Pereira, C.I. Conegero, 2003. Estudo da glândula pineal de suíno por meio de microscopia de luz. *Acta Scientiarum Biological Sciences*, 25(2): 453-458.

Mançanares, C.A.F., I.L.S. Prada, A.F. Carvalho, M.A. Miglino, J.F.P. Martins, C.F. Ambrósio, 2007. Morfologia da glândula pineal em

gambás (*Didelphis* sp.) *Brazilian Journal of Veterinary Research and Animal Science*, 44(3): 222-229.

Marques, L.O., A.F. Carvalho, A.C.F. Mançanares, C.A. Mançanares, 2010. Estudo morfológico da glândula pineal de *Procyon cancrivorus* (Cuvier, 1798) (mão-pelada). *Biotemas*, 23(2): 163-171.

Rocha, R.M.P., M.H.T. Matos, L.F. Lima, M.V.A. Saraiva, A.M.C.V. Alves, A.P.R. Rodrigues, 2011. Melatonina e reprodução animal: implicações na fisiologia ovariana. Acta Veterinária Brasilica, 5(2): 147-157.

Srinivasan, V., W.D. Spence, W.D. Pandi-perumal, SR. Spence, R. Pandi-perumal, K.P. Zakharia, Bhatnagar, A.K. Brzezinski, 2009. Melatonin and human reproduction: Shedding light on the darkness hormone. Gynecological Endocrinology, 25(12): 779–785.

Tilden, A.R., V.H. Hutchinson, 1993. Influence of photoperiod and temperature on serum in the Diamondback Water Snake (*Nerodia rhombifera*). General and Comparative Endocrinology, 92(3): 347-354.