

Morphological and Histological study of the pineal gland in Aratinga auricapillus (gold capped conure).

Shermean Abdulla Abd-ALrahman

College of Education For Pure Science (Ibn AL-Haitham).

Abstract

The pineal gland of male gold capped conure was investigated to find out the description of pineal parts design and cellular components at the light microscopic level by using Haematoxyline – Eosin and periodic acid schift reagent stain methods .The results of morphological aspects revealed that the pineal body (distal part) was broad conical shaped with apex pointing anteriorly and the pineal stalk (proximal part) was narrow long slender and middle structure , which arises from the diencephalon . The result of microscopic examinations indicated that the pineal parenchyma presented a tubule-follicular structural organization. The rudimentary receptor pinealocytes formed a layer of elongated cells with basal nuclei limiting the follicular lumen ,and had sensory cilia projected into the lumen and granular vesicles .These cells were designed as two forms A and B .Three distinct regions of regular stratified organelles distribution was the characteristic feature of the rudimentary receptor pinealocytes form A , while the cells form B were shorter and lacked that polarity in their distribution of internal structures. The secretory pinealocytes located in the external follicular layers were small , round and oval located in the external follicular layers were small , round and oval shaped cells, with numerous dense core vesicles and with irregular shaped nuclei . There were a populations of non pinealocytes , small vessels, capillaries and nerve fibers found in the vascular surfaces.

Introduction

The pineal gland (also called the epiphysis cerebri , conarium and third eye) is a small and very important photoneuroendocrine organ located in the brain [1,2,3]. The avian pineal gland represents a transitional type between a directly photoreceptors organ of non-mammalian species and prominent secretory organ in mammals[4,5,6]. The pineal glands in birds reveals a varying degree of morphological and structural organization [7,8,9,10, 11,12].The avian pineal glands contain a circadian clock system [13,14,15], it acts like the suprachiasmatic nucleus found in the hypothalamus of mammals [16]. The rhythmic production of melatonin is under control of endogenous oscillators located within the pineal itself [17,18].Melatonin is an indoleamine hormone secretes during night,it is considered as chronological pacemaker hormone

[19].The pineal gland regulates the activity of many physiological processes including reproduction[20] , immune function [21,22], metabolism [19] and facilitate migration[23].The present study is based on the pineal gland of male Aratinga auricapillus (gold capped conure) which belong to the class Aves , Order Psittaciformes , Family Psittacidae and Subfamily Arinae. Gold capped conures are zygodactyls having the four toes on each foot placed two at the front and two back. While the curved beak shapes with the upper mandible having slight mobility in the joint with the skull .The male have white areas around the eyes[24].

Our aim is to identify the morphology , histology and cytoarchitectures of different pineal cells distribution in male diurnal bird species , since the avian pineal gland represent an excellent model for studying the bases and regulation of circadian rhythms , is relatively autonomous in birds as stated by [25].

Material and Methods

Ten healthy adult male gold capped conures were used in this study, obtained from the spinning market during winter season .The birds were decapitated and the pineal glands were obtained by careful dissection. The pineal glands were removed from the meninges and calvarium and then fixed in 10% buffered formalin, washed, dehydrated through deascending grades of alcohol, cleared and embedded in paraffin wax .

Five –Six microns thick sections were cut by using rotary microtome . The paraffin wax was removed by immersing the slides in xylene . The slides were passed through descending grades of alcohol , distilled water and the sections were stained with Haematoxyline – Eosin and special stain periodic acid shift reagent stain methods , cleared with xylene and mounted with D.P.X. as per usual methods [26].

Results

Morphological description of the pineal gland

The pineal gland in gold capped conure was intracranial midline in position , lodged in a narrow triangular space between the right , left cerebral hemisphere and the cerebellum Fig.(2). It was a reddish gray organ surrounded by a capsule which attached dorsally to the closely overlying meninges sheaths of the calvarium by stout filaments Fig.(3). The pineal gland was composed of two parts : 1- a broad conical shaped body (distal part) which found more dorsal towards the roof of calvarium with apex pointing anteriorly , 2- a narrow long slender middle stalk (proximal part) which arises from the diencephalon and connected to the roof of the third ventricle.

Histological structure of the pineal gland

The pineal gland in gold capped conure was encapsulated by thin layers of connective tissues that closely associated with the meningeal layers . The

capsule formed incompelete septa separated the pineal parenchyma components . The pineal parenchyma presented tubule- follicular structural organization, and was formed by large oval follicles with wide lumen and small spherical follicles with reduced lumen Fig.(4)

The nerve fibers formed a basket like structures around the pineal parenchyma . The follicular wall was formed by layers of cells resembled the pseudostratified epithelium , the follicular lumen was limited by a layer of two forms A and B of the rudimentary receptor pinealocytes ,they appeared as a slim elongated cells with nuclei located in their basal parts Fig.(5). The external layers of the follicular wall were formed by layers of small round and oval secretory pinealocytes .The rudimentary receptor pinealocytes form A were elongated cells bordering the luminal sides of the follicles , extended through the whole follicular wall and situated directly on the basement membrane reached to it by the cytoplasmic processes . These cells were characterized by the regular and stratified distribution of organelles located in the form of three distinct regions, they were a basal region , narrow neck region and outer segment region .Aggregation of large

amount of granular vesicles positive to periodic acid schift reagent stain were seen at the vestigial apical prolongation of the outer segment Fig.(6). The cytoplasm of the narrow neck region connecting the basal cell region with the outer segment that extended into the follicular border

A large amount of dense core vesicles located beside , underneath the basal nucleus and in the basal cytoplasmic processes of the basal region. The rudimentary receptor pinealocytes form B were shorter than the cells of form A .These cells were connected with the follicle border and showed polarity in their internal structure, also they were lacked the regular and stratified distribution of organelles Fig.(6) . The typical features of the apical parts of the rudimentary receptor pinealocytes form A and B were the sensory cilia that projected into the follicular lumen Fig.(5,6).

The secretory pinealocytes were characterized by the absence of the outer segments , the cytoplasmic granules and the presence of numerous dense core vesicles . The cell boundaries were indistinct , the nuclei had irregular shapes which due to the invaginations of the nuclear membrane Fig.(6).The pineal also contained non pinealocytes cells such as the interstitial supporting cells (astrocyte glial cells) .

These cells had small elongated nuclei which stained more darker than that of pinealocytes ,they were found in the external parts of the follicular wall Fig. (5) .The interstitial spaces around the follicles (i.e. the vascular surfaces) were contained connective tissues , many small vessels , capillaries, nerve fibers and pleomorphic neurons , the latter synapses with the pinealocytes that found inside the follicles. Singly and small clusters of lymphocytes were found Fig.(7).

Morphological and Histological study of the pineal gland in Aratinga auricapillus (gold capped conure). Shermean Abdulla Abd-ALrahman

The proximal parts of the pineal gland were characterized by the presence of many non myelinated nerve fibers along the posterior wall of the stalk and a number of nerve cells located in the form of a layer subjacent to the pinealocitic lining around the basal portion of the stalk.

Discussion

The gold capped conure possessed a well developed pineal gland similar to that described in other birds [6 ,9,12,27], and unlike to that of some owl species [13] and some temperate owl species [20], they had degenerated pineal gland .

The morphological observation of this study revealed that the pineal body (distal part) had conical shape, in comparison to that of other birds, It was triangular in common gull [6] , enlarged club in tinamid birds [8] , club top shaped in turkey [10] and wide in domestic goose [11] . The pineal stalk (proximal part) arises from the diencephalon and connected by a lumen to the roof of the third ventricle , this finding was inaccordance with the statement that found in common gull [6] and passeriform birds[7] . The pineal parenchyma presented tubule- follicular structural organization . Similar finding were found in tinamid birds [8] , Perdicula asiatica [10] , turkey [10,12] and domestic goose [11] . The tubule follicular type described as intermediate state between the saccular type (i.e. a system of open tubes) that found in passeriform birds , two species of crows[7] and the lobular type (i.e. masses of parenchymal cells arranged in closed clusters) that found in adult domestic fowl[11] .

The pineal parenchyma in gold capped conure was formed by large oval follicular with wide lumen and small spherical follicles with reduced lumen . In Rhynchotus and Nothura there are a predominance of small follicles with a reduced lumen , while in Tinamus and Crypturellus , there are a predominance of large follicles with wide lumen[8].

The cytoarchitectural results indicated the two different types of pinealocytes, the interstitial glial cells and the pleomorphic neurons , similar observation reported in Corvus splendens [7] , turkey [10,12] and domestic goose [12] . The rudimentary receptor pinealocytes showed in two forms A and B . This finding was reported only in domestic goose and not in other birds as stated by [11] .The sensory cilia found in the apical part of the rudimentary receptor pinealocytes , represented the photic input pathway to the oscillator as stated by [14] in chicken pineal . The apical prolongations of the outer segments were vestigial,it was due to the absence of the membrane multiplications that found in the photoreceptor outer segments of non mammalian species [4]. The granular vesicles found in this study possessed immunocytochemical serotonin [27] and photo pigments pinopsin [28].

Morphological and Histological study of the pineal gland in Aratinga auricapillus (gold capped conure). Shermean Abdulla Abd-ALrahman

The synapses between long and short processes of the rudimentary receptor pinealocytes and pleomorphic neurons contained synaptic ribbons which appeared small rod like during the day and large , slightly curved at nights [4] , these synaptic ribbons contained neurohormonal endings [2,7] , photopigment like rhodopsin , iodopsin [5,28] , melanopsin and cryptochromes which involved in regulation of clock function and melatonin production [26].

The secretory pinealocytes were characterized by the absence of the outer segments and by the presence of numerous dense core vesicles, these cells were strongly similar to mammalian pinealocytes as stated by [16,23] .

References

- 1- Hickman,CL.p. ; Roberts , L.S. ; Keen, S.L. ; Eisenhour, D.J. ; Larson, A. and Anson, H.(2011). Integrated principles of zoology .5th ed. ,ch. 34 , Mc. Graw- Hill New York .
- 2- Haldar, Ch. and Araki, M. (2002). Morphometric analysis of photoreceptive , neuronal and endocrinal cell differentiation of avian pineal cells : an in vitro immunohistochemical study on the developmental transition from neuronal to photo endocrinal property. Zoology Sci. Vol. 19(7): PP. 781-787 .
- 3-Acer, N.; Ilica, AT. ; Turgut, AT. ; Ozcelik, O. ; Yildirim, B. and Turgut ,M.(2012). Comparison of three methods for the estimation of pineal gland volume using magnetic resonance imaging . Scientific world Journal , Vol. 10 , PP.11- doi 10.001 123412-Epub .
- 4- Vigh, B. and Vigh-Teichmann, I. (1992).Two components of the pineal organ in the mink Mustela vison ; their structural similarity to sub mammalian pineal complexes and calcification. Arch. Histol. Cytol. , Vol. 55 (5) , PP. 477-489.
- 5- Sato, T. (2001). Sensory and endocrine characteristics of the avian pineal organ. Microsc. Res. Tech. ,Vol. 53(1) , PP. 2-11 .
- 6- Przybylska, G.B. ; Lewczuk , B.; Prusik , M. ; Kalicki , M. and Ziolkowska , N. (2012). Morphological studies of the pineal gland in the common gull Larus canus , reveal uncommon features of pinealocytes . The Anatomical Record : Advances In Integrative Anatomy And Evolutionary Biology . PP. 1-3 , doi: 10.1002/ar . 22407, Wiley Periodicals , Inc.
- 7- Chauhan , B.C. and Ambadkar , P. M. (1984). Certain observation on the pineal organ of Indian house crow , Corvus splendens (vieillot) during annual cycles . Yamashina. Inst. Orinth. , Vol. 16, PP. 63-82 .
- 8-Redins, CA. and Machado, AB. (1989). Histologic study of the pineal organ in tinamid birds. Rev. Bras. Biol. Vol. 49(2) , PP. 429-434 .
- 9- Haldar, C. and Bishnupuri , K. S. (2001). Comparative view of pineal gland morphology of nocturnal and diurnal birds of tropical origin . Microsc. Res. Tech. , Vol. 53, PP. 25-32 .
- 10- Przybylska, G. B. ; Lewczuk, B. ; Prusik, M. and Nowicki, M.(2005). Post hatching development of the turkey pineal organ : histological and immunohistochemical studies . Neuro. Endocrinol. Lett. , Vol. 26(4) , PP. 383-392 .

Morphological and Histological study of the pineal gland in Aratinga auricapillus (gold capped conure). Shermean Abdulla Abd-ALrahman

- 11- Prusik, M. ; Lewczuk, B. ; Nowicki , M. and Pruzbylska , G. B. (2006) . Histology and ultrastructure of the pineal organ in the domestic goose . Histol. Histopathol. Vol. 10 , PP. 1075-1090 .
- 12- Prusik, M. and Lewczuk, B.(2008). Structure of the avian pineal gland . Medycyna Weterynaryjna , Vol. 64(6) , PP. 764-769 .
- 13- Taniguchi, M. ; Murakami, N. ; Nakamura, H. ; Nasu, T. ; Shinohara, S. and Etoh, T. (1993) . Melatonin release from pineal cells of diurnal and nocturnal birds . Brain Res. , Vol. 620(2), PP.297-300 .
- 14- Okano, T. and Fukada, Y. (2001). Photoreception and circadian clock system of the chicken pineal gland . Microscopy Research and Technique , Vol. 53(1) , PP. 72-80 .
- 15- Natesan, A.; Geetha, L. and Zatz, M. (2002). Rhythm and soul in the avian pineal . Cell and Tissue Research , Vol. 309(1) , PP. 35-45 .
- 16- Fukada, Y. and Okano, T. (2002). Circadian clock system in the pineal gland . Neurobiol , Vol. 25(1) , PP. 19-30 .
- 17- Korf, H.W.(1994) . The pineal organ as a component of the biological clock . phylogenetic and ontogenetic considerations . Ann. N.Y. Acad. Sci. , Vol. 719, PP. 13-42 .
- 18- Csernus, VJ. (2006). The avian pineal gland . Chronobiol. Int. , Vol. 23(1-2), PP. 329-339.
- 19- Bailey, M.J. ;Beremand, Ph. ; Hammer, R. ; Bell, D. ; Thomas, T. and Cassone, V. (2003).Transcriptional profiling of the chick pineal gland, a photoreceptive circadian oscillator and pacemaker. Molecular Endocrinology , Vol. 17, PP.2084-2095.
- 20- Guchait, P. and Haldar, C. (2001). A reproductive phase-dependent effect of dietary L- tryptophan on pineal gland and gonad of a nocturnal bird , Indian spotted owl Athene brama . Acta. Biol. Hung. , Vol. 52(1) , pp. 1-7 .
- 21- Sonta, K.S.(1996). Functional connections between the pineal gland and immune system. Acta. Neurobiol. Exp. , Vol. 56, PP. 341-357.
- 22- Ayana, R.(2010). Fluoride and the pineal gland . National research council university of surrey in England . PP. 1-15.
- 23-Bentley, G.E.(2001). Unraveling the enigma : The role of melatonin in seasonal processes in birds. Microscopy Research and Technique. Vol.53(1), PP. 63-71.
- 24-Gaunt, A. and Woring, L. (1999). The ornithological council , 2nd , Washington D.C. 20006- 1401 , pp. 1-3 .
- 25-Natesan, A. ; Geetha, L. and Zatz, M. (2002). Rhythm and soul in the avian pineal . Cell Tissue Res. , Vol.309(1) , PP. 35-45.
- 26- Sood,R.(2009). Medical laboratory technology : methods and interpretations 6th edi. Ch.27, New Delhi , India, PP. 1436-1450 .
- 27- Fejer, Z. ; Rohlich, P. ; Szel, A. ; David, C. ; Zadori, A. ; Manzano, M.J. and Vigh, B.(2001). Comparative ultra structure and cytochemistry of the avian pineal organ. Microscopy Research and Technique , Vol. 53 , PP. 12-24.
- 28-Yamac, M. ; Araki, M. ; Okano, T. ; Fukada, Y. and Oishi, T.(1999). Differentiation of pinopsin - immunoreactive cells in the developing quail pineal organ: An in vivo and in vitro immunohistochemical study. Cell and Tissue Research , Vol. 296(3) , PP.667-671.

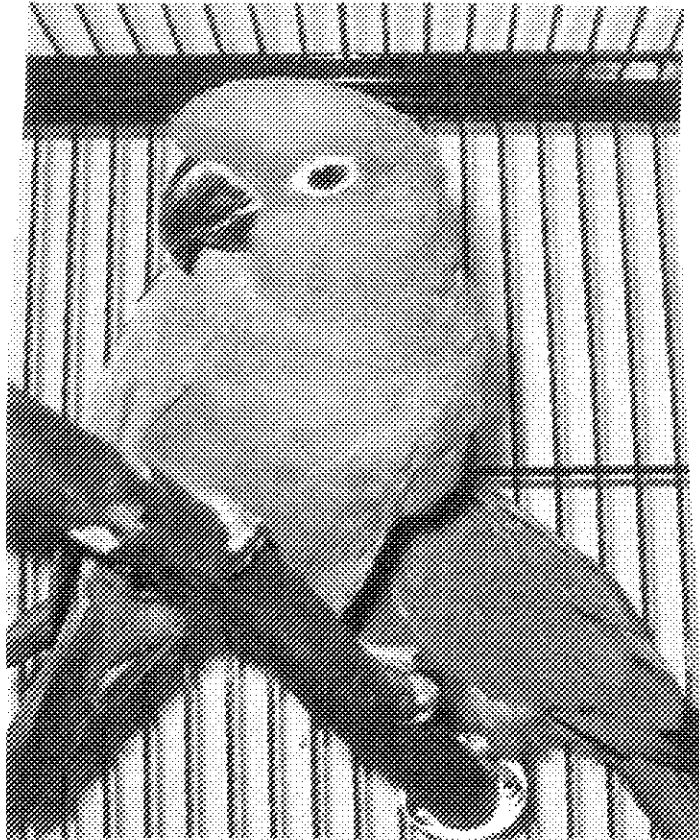
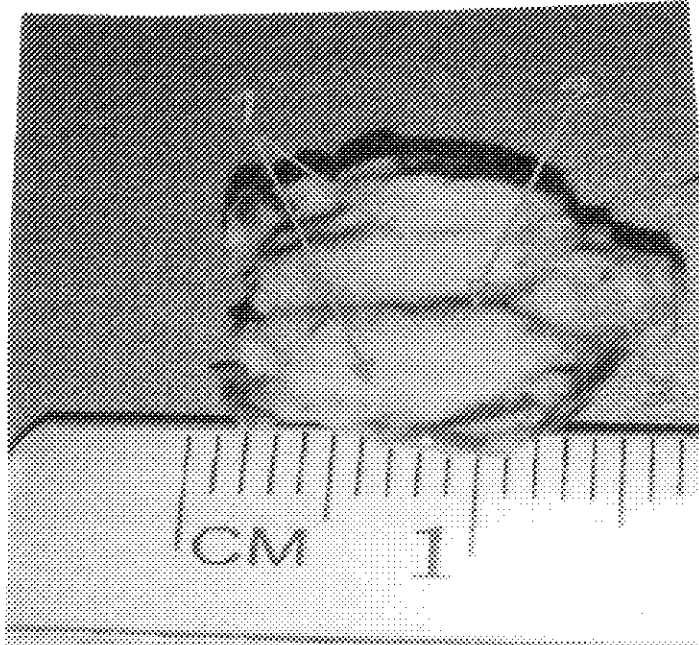
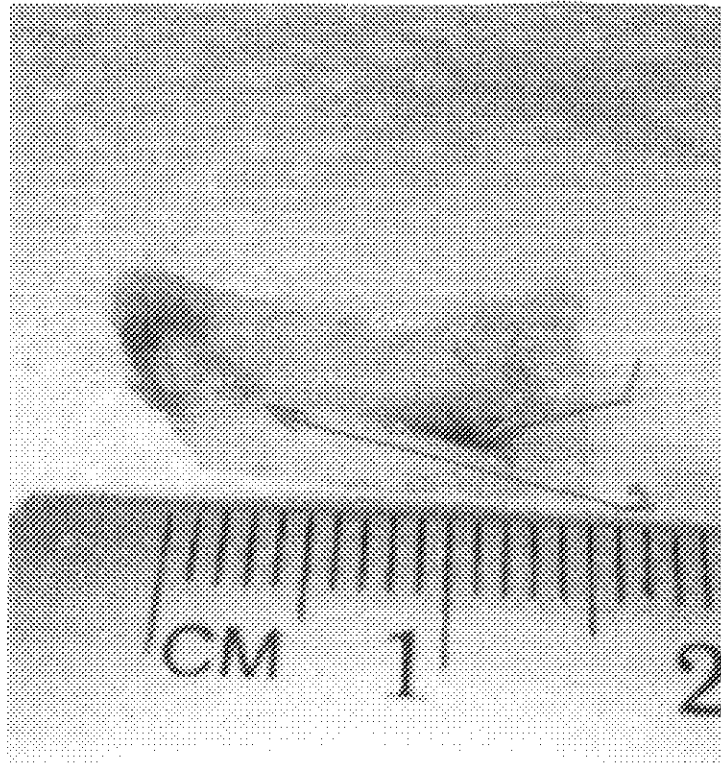


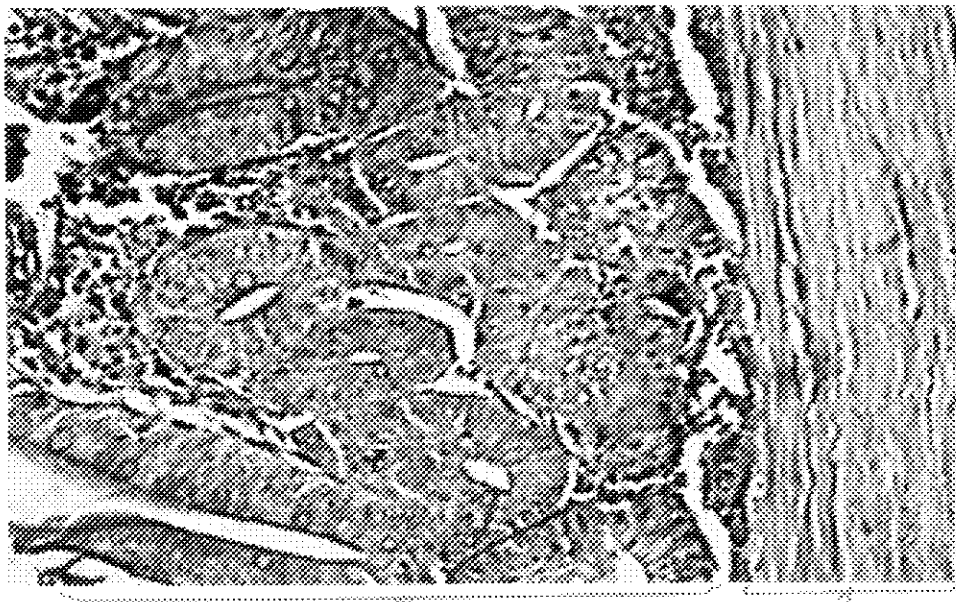
Fig. (1) Aratinga auricapillus (gold capped conure).



**Fig. (2) Dorsal view of gold capped conure brain shows:
1. Cerebral hemisphere 2. Pineal gland 3. cerebellum**



**Fig. (3) Pineal gland attached with the calvarium:
1. body (distal part) 2. Stalk (Proximal part)**



**Fig. (4) Longitudinal section in pineal gland illustrates: (PAS) (20 X)
1. capsule 2. Pineal parenchyma**

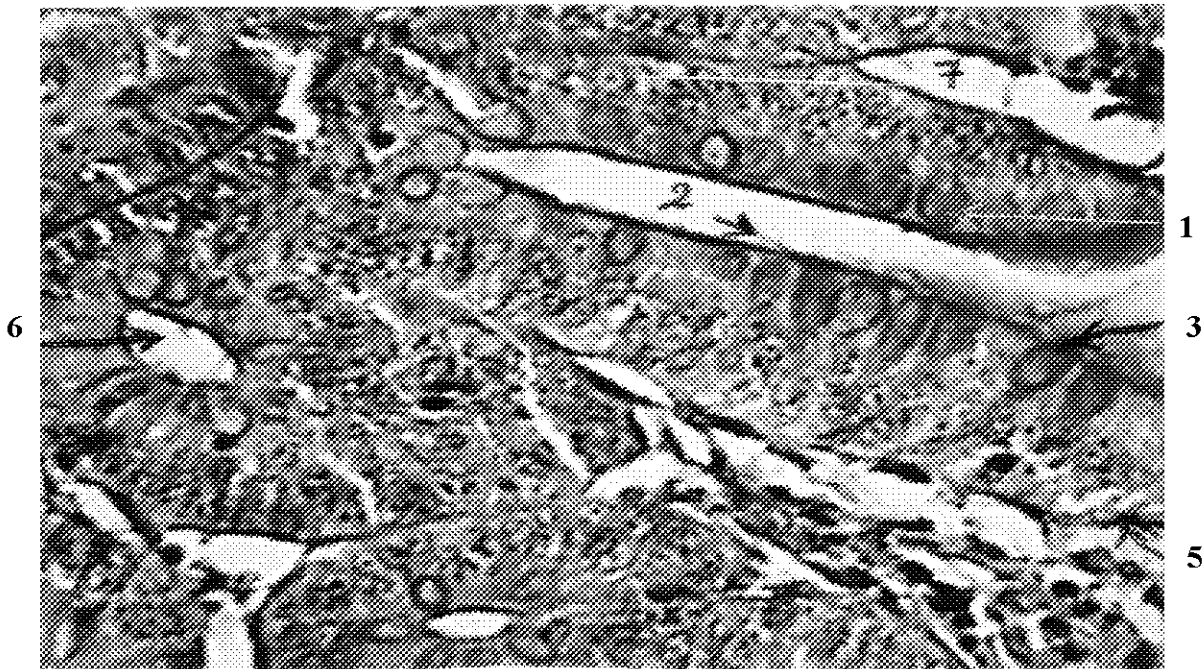


Fig (5) Longitudinal section in pineal parenchyma illustrates: (PAS) (40 X)
 1. granular vesicles 2. sensory cilia 3. rudimentary receptor pinealocyte Form A 4. follicular wall 5. glial cells 6. Lumen 7. secretory pinealocytes

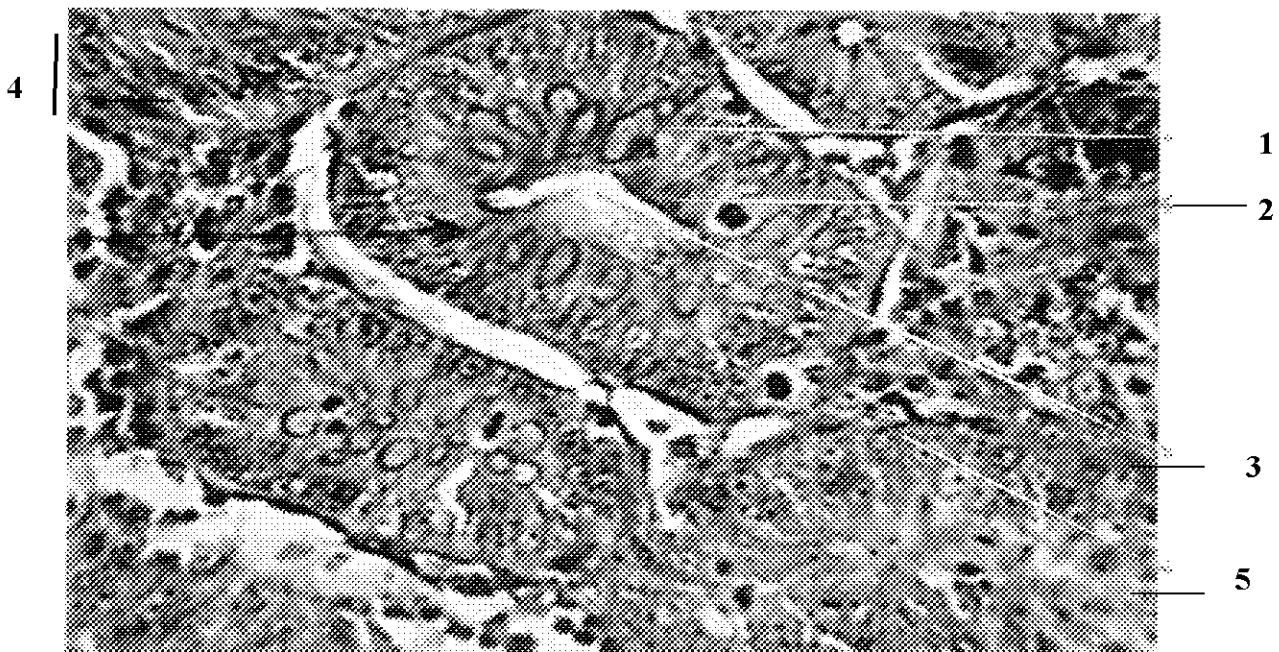


Fig. (6) Longitudinal section in pineal parenchyma illustrates: (PAS) (40 X)
 1. rudimentary receptor pinealocyte Form A 2. rudimentary receptor pinealocyte Form B 3. Lumen 4. secretory pinealocytes 5. glial cells



Fig. (7) Longitudinal section in pineal parenchyma illustrates the interstitial spaces around the follicles (vascular spaces): (PAS) (40 X)

**1.lymphocytes 2. Pleomorphic neurons 3. blood vessels 4. Nerve fibers
5. Septum**

الخلاصة

تناولت دراسة الغدة الصنوبرية لذكر طائر الغندور (gold capped conure) وصف شكل أقسام الغدة الصنوبرية ومكوناتها الخلوية عند فحصها بالمجهر الضوئي و باستعمال تقنية التلوين بالهيماتوكسيلين-ايوسين ، وكاشف شف الدوري، وأظهرت نتائج الدراسة المظهرية الشكل المخروطي الواسع ذو القمة الأمامية للجسم (أي القسم القاصي للغدة الصنوبرية) والشكل المتوسط الاسطواناني الطويل الضيق لتكوين الساق (أي القسم الداني للغدة الصنوبرية والمرتبطة بالدماع البييني) . أظهرت نتائج الفحص بالمجهر الضوئي اتخاذ اللحمية الصنوبرية مظهر الترتيب النببي- الجريبي وظهرت الخلايا الصنوبرية المستلمة الأثرية كطبقة من خلايا متطاولة ذات نواة قاعدية تحيط بجوف الجريبات ، ولها أهداب حسية بارزة إلى الجوف وحوصلات حبيبية . لوحظ وجود نمطين من هذه الخلايا هما أ وب، و يعد انتشار العضيات الطبقي المنتظم هو الصفة المميزة للخلايا الصنوبرية المستلمة الأثرية نمط أ ، في حين افتقدت الخلايا الصنوبرية المستلمة الأثرية نمط ب القصيرة هذا التنظيم وأظهرت صفة القطبية في توزيع محتوياتها الداخلية، واتخذت الخلايا الصنوبرية الغدية موقع ضمن الطبقات الخارجية للجريبات ، وكانت عبارة عن خلايا صغيرة مستديرة وبيضوية ولها حوصلات كثيفة المركز ونواة ذات شكل غير منتظم ، وتواجدت مجاميع الخلايا غير الصنوبرية والأوعية الدموية الصغيرة والشعيرات الدموية والألياف العصبية ضمن السطوح الوعائية.