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Post-hatching evolution of the pineal gland of the chicken

J. Boya and J. Calvo

Department of Histology and General Embryology Faculty of Medicine, University Complutense, Madrid, Spain

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Abstract. The authors studied the evolution of the pineal gland of the chicken *(Gallus gallus)* from hatching until 34 months of age. They describe the progressive decrease of the follicular cavities and the appearance of solid-looking cavities. The stroma increases with age, dividing the pineal parenchyma into territories of small caliber.

Introduction

After hatching, the pineal gland of the chicken is an organ in the form of a drumstick located in the triangular space between the cerebral hemispheres and the cerebellum. It is surrounded by a conjunctive capsule joined to the dura mater [*Quay,* 1965; *Pearson,* 1972; *Hodges,* 1974], from which thin partitions originate which penetrate toward the interior and separate the lobules and the glandular follicles [*Kappers and Schade,* 1965; *Beattie and Glenny,* 1966; *Wight and McKenzie,* 1971; *Hodges,* 1974].

The pineal stroma contains various cell types, blood vessels and amyelinic nerve fibers. A typical component of the pineal stroma of the chicken is the lymphoid tissue. It was described by *Romieu and Jullien* [1942] and later confirmed by *Spiroff* [1958] and *Quay* [1965].

The pineal nerve fibers originate in the superior cervical ganglion [*Stamer,* 1961; *Kappers and Schade,* 1965; *Lauber,* 1968]. After penetrating into the gland, they are distributed into the conjunctive spaces from which location they may penetrate the parenchyma [*Stamer,* 1961; *Quay and Renzoni,* 1963; *Kappers and Schade,* 1965; *Quay,* 1965]. *Wight and McKenzie* [1970] demonstrated the presence of fibers with green fluorescence using the FIF method. These authors also described the existence of cholinergic fibers in the pineal gland of the chicken.

According to *Studnička's* [1905] classification, the pineal gland of the chicken was considered to pertain to the solid lobular type [*Gladstone and Wakeley,* 1940; *Spiroff,* 1958; *Quay,* 1965; *Wight and McKenzie,* 1971] or to a type between the lobular and follicular forms [*Romieu and Jullien,* 1942; *Oksche,* 1965].

The wall of the pineal follicles presents two cell types: ependymocytes and hypendymocytes. They were first described by *Funkquist* [1912] and later accepted by the majority of the authors [*Spiroff,* 1958; *Beattie and Glenny,* 1966; *Wight and McKenzie,* 1970; *Pearson,* 1972; *Hodges,* 1974]. The ependymocytes are columnar cells radially positioned around the follicular lumen. The hypendymocytes are located between the ependymocytes and the basal membrane, they are small in size and have a polygonal contour.

In 1971, *Wight and McKenzie* completed the only histochemical study done on the pineal gland of the chicken. These authors noted an intense activity in the acid phosphatase, lipase, ATPase, ß-glucuronidase, and succinic dehydrogenase.

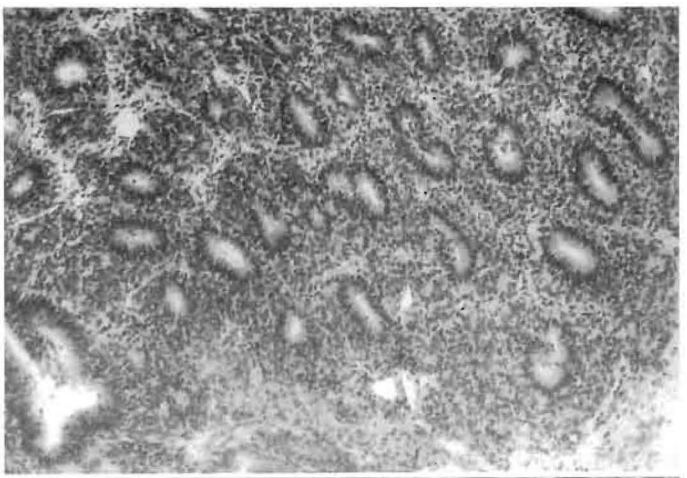

Fig. 1. 5-day-old chicken. Note the cellular density of the pineal gland and the small caliber of the follicular lumens. HE.

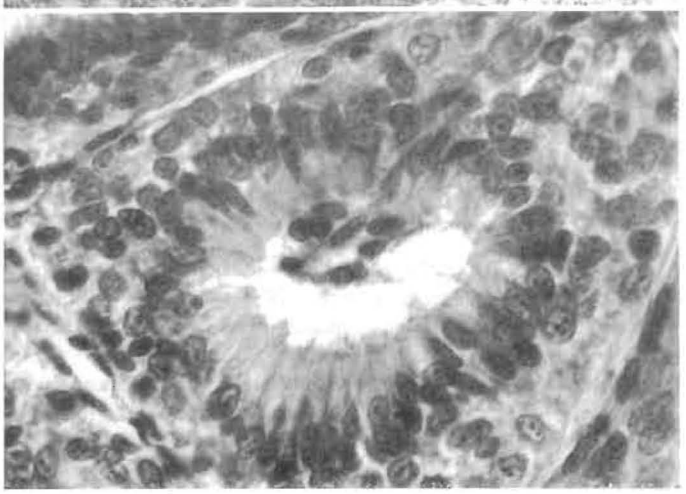

Fig. 2. 10-day-old chicken. A follicle whose lumen is limited by columnar cells that have apical projections. Cells apparently loose in the follicular lumen. HE.

In an earlier work [*Calvo and Boya*, 1977], we studied the embryonic development of the pineal gland of the chicken and in this work we studied the post-hatching evolution of this gland.

Materials and methods

We studied the post-hatching morphological evolution of the pineal glands of chickens that were maintained under normal conditions of illumination and diet. We took samples 1, 2, 5, 7, 10, 15, 20, 25, 30 days and 2, 3, 4, 7, 8, 13, 20 and 34 months after hatching. Except for the first few days, we took the isolated pineal glands, attached to the boney vault, to obtain an easier fixation and to avoid artifacts from trauma.

At each time interval, we sacrificed 6 chickens. The pineal glands were fixed in 10% formaldehyde, acetic-Bouin, and Bouin-Hollande. After inclusion in paraffin and making serial 7-μm cuts, we applied the techniques of HE and PAS staining and Gomori's argentic impregnation for reticular fibres.

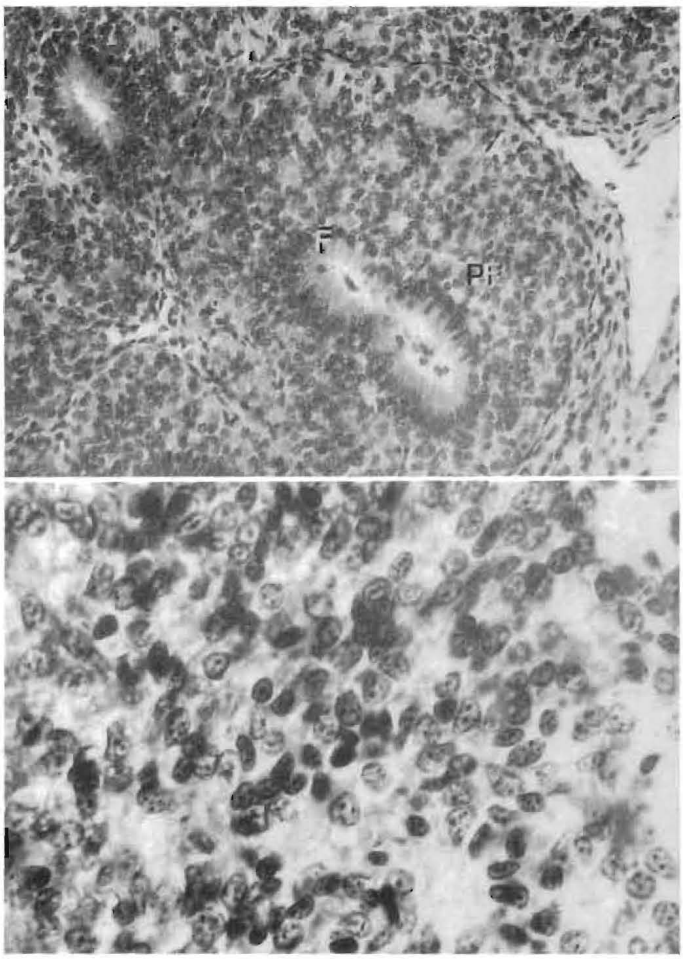

Fig. 3. 5-day-old chicken. A follicle with clear differentiation of the follicular (F) and parafollicular (PF) zones. The follicles seem to be separated by fine connective-vascular partitions. HE.

Fig. 4. 10-day-old chicken. Parafollicular zone in which various types of nuclei and small cavities can be seen. HE.

Results

The post-hatching evolution of the pineal gland of the chicken is an evident but slow process. Therefore we prefer to dispense with separate descriptions for each interval studied and give a complete description of the process of evolution.

During the first month after hatching, the morphology of the pineal gland is very similar to that of the older embryos [*Calvo and Boya,* 1977], although there are sufficient differences for the characterization of this period. The volume of the pineal gland gradually increases during the post-hatching life. Its growth is due in part to the division of the pinealocytes. Mitosis is frequently encountered in the pineal gland, especially during the first month after hatching. The second reason, for us the most important, for the increase in

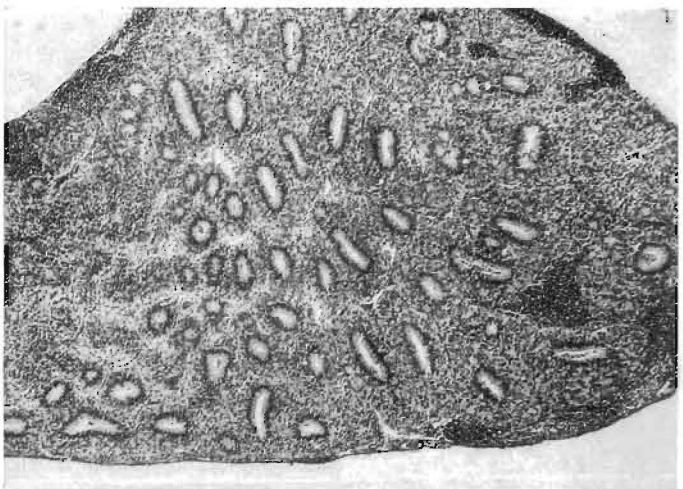

Fig. 5. 15-day-old chicken. Lymphoid nodules, fundamentally, subcapsular in the pineal gland. HE.

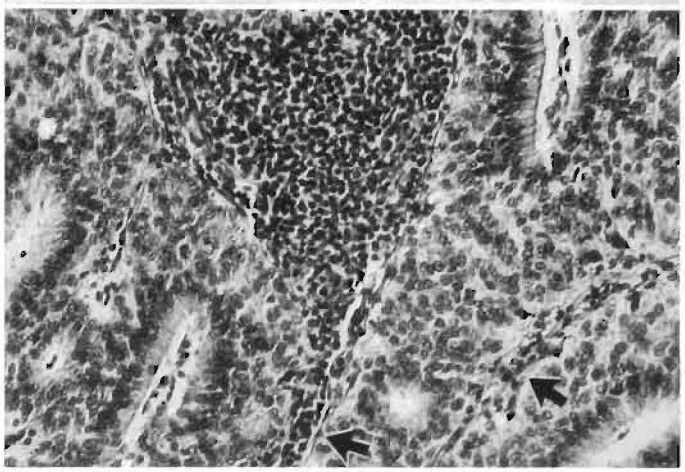

Fig. 6. 15-day-old chicken. Lymphoid nodules between follicles, and lymphoid prolongations on the connective partitions (arrows). HE.

volume of the pineal gland is the hypertrophy of the pinealocytes, as is demonstrated by the gradual decrease of the number of nuclei per unit of surface as the age increases.

Another morphological consideration which serves to differentiate the pineal glands during the first month after hatching from those in the last stages of embryonic development, is the progressive reduction of the caliber of the follicular cavities (fig. 1) to narrow clefts. Observation under high-power magnification permits the verification of a better development of the supranuclear cytoplasm (fig. 2) of the limiting cells of the cavities. There are also, apparently free cells in the follicular lumen, but the study of serial cuts demonstrates that these cells are permanently attached to the follicular wall.

The walls of the pineal cavities continue to demonstrate a clear limitation between the follicular and parafollicular layers (fig. 3). The parafollicular layer has a larger number of

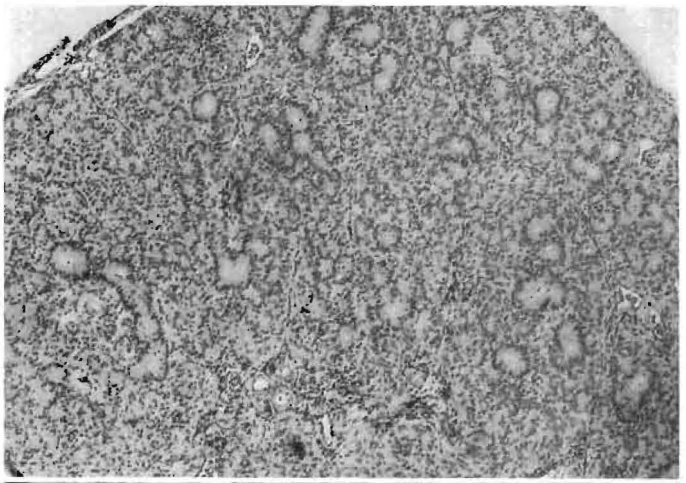

Fig. 7. 4-month-old chicken. Compact aspect of the pineal gland. The follicular lumens are of very small caliber. HE.

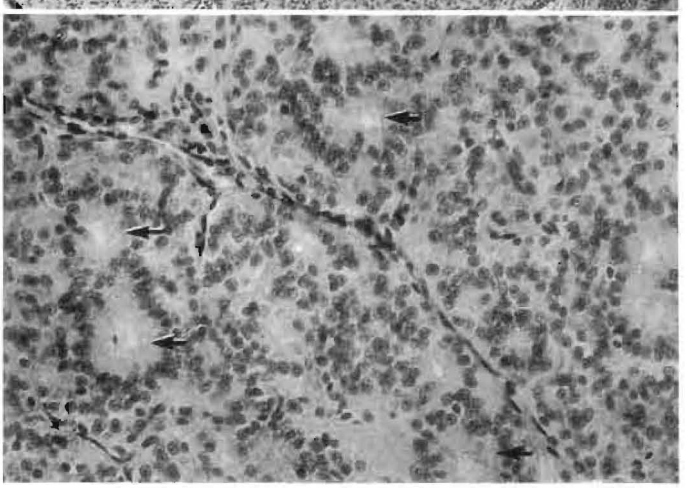

Fig. 8. 4-month-old chicken. Parafollicular area. This zone is principally formed by cellular rosettes (arrows) with a very small lumen. HE.

cellular rosettes and small cavities. The number of dense cells has increased in both layers as compared with those existing in the embryonic states (fig. 4).

A typical characteristic of the pineal gland of the chicken in the first month of life after hatching is the formation of lymphoid tissue, which begins at 10–15 days, and varies according to the lot of the animals (fig. 5). The lymphoid tissue of the pineal gland reaches its maximum development between 15 and 30 days after hatching. After this time, it slowly decreases but does not totally disappear, as the persistence of isolated lymphoid nodules demonstrates in 34-month-old chickens. Morphologically, the lymphoid tissue appears in supracapsular nodules (fig. 5) which tend to prolongate to the interior of the pineal gland, following the conjunctive partitions. We also saw nodules which were more centrally located and small infiltrations of lymphoid cells disseminated in the inter-

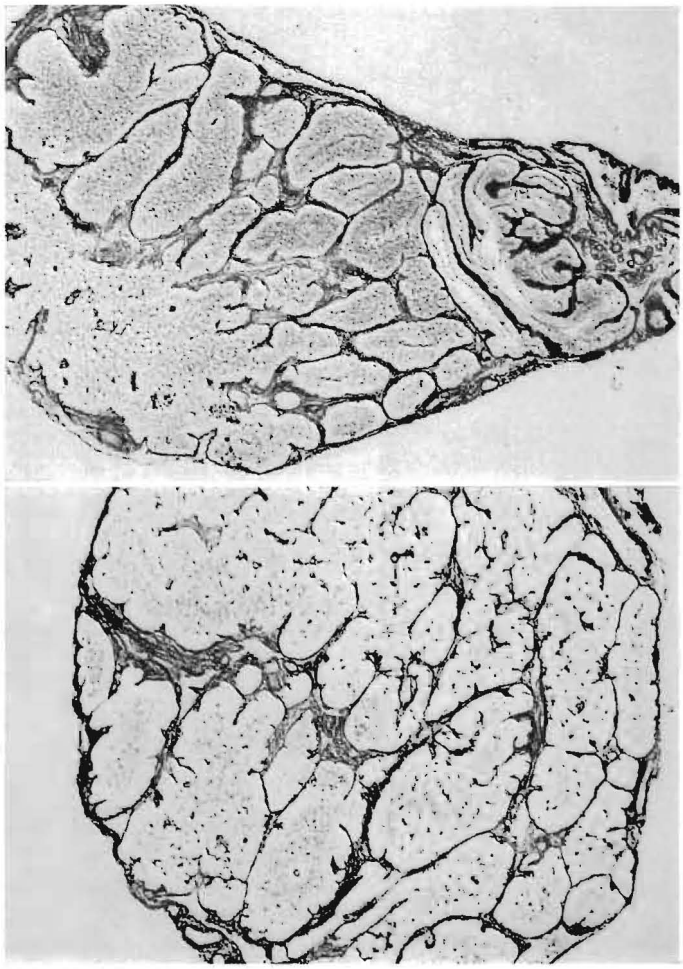

Fig. 9. 15-day-old chicken. Pineal stroma that shows structure of ample lobules, very similar to the one seen in the last stages of embryonic development. Technique of Gomori.

Fig. 10. 4-month-old chicken. The lobules are partially divided by proliferation of the connective-vascular partitions, projecting toward the interior of the lobules. Technique of Gomori.

follicular partitions (fig. 6). No invasion of the parenchyma could be detected with the optical microscope.

The lymphoid nodules contain small lymphoid and large reticular cells, some capillaries and a lax net of reticular fibers. In no instance did we encounter germinative centers.

The morphological evolution of the pineal gland from the first month follows the pattern we described in an earlier paper. The most evident morphological data is the compact appearance the gland assumes with age (fig. 7), principally because of the reduction of the caliber of the follicular lumens. In many cases, the development of the supranuclear cytoplasmic portion and of the apical prolongations of the pinealocytes appears to be the principal factor in the occlusion of the lumen. There is also a progressive fragmentation of the pineal recess and of the large follicular cavities. In effect, after the 2nd–4th

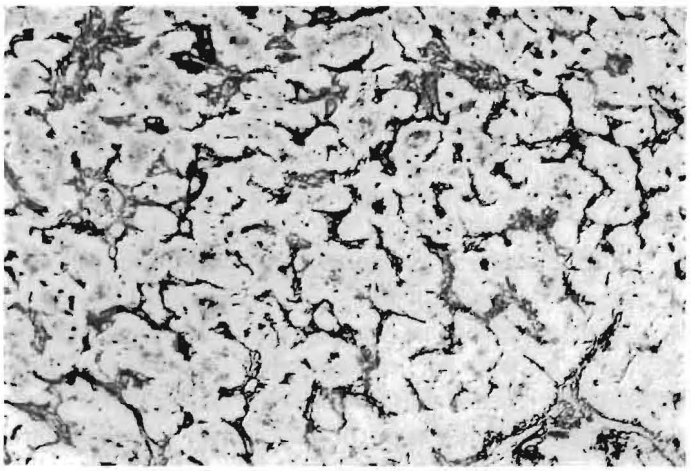

Fig. 11. 34-month-old chicken. Division of the pineal parenchyma into small territories, separated by connective-vascular partitions of larger caliber. Technique of Gomori.

month, the major axis of the follicular cavities is smallest. The pineal recess and the large follicular lumens cannot be identified yet. Aligned follicular lumens are frequently encountered, indicating the aforementioned fragmentation.

These formational changes of the pineal cavities are accompanied by parallel changes in the follicular zones. After 4 months, the follicular layers are still slightly evident at low-power magnification. The nuclei of the follicular cells usually are arranged in a single thread. The parafollicular layer starts and progressively disappears after the first month. At 4 months, the major part of the parafollicular layer is still composed of cellular rosettes or cavities of minimal size which are only visible at high-power magnification (fig. 8). Cytologically, a decrease in the quantity of dense nuclei is noted. The aforementioned hypertrophy of pinealocytes is also very evident.

The argentic impregnations yield representative image of the evolution of the aforementioned cyto-architectonic pineal model. Until the first month of post-hatching life, the pattern is similar to that observed in the embryos in the last embryonic stages (fig. 9). The pineal follicles are separated by narrow conjunctive laminae and the contour of the follicle is smooth. After 30 days of age, evaginations of the stroma toward the parenchyma which tend to incompletely subdivide into smaller territories start to appear (fig. 10). The penetrations of the stroma continue to increase in number and depth with age. The PAS and HE techniques show that these penetrations correspond to vessels which grow toward the interior of the follicles, accompanied by a narrow conjunctive layer. At the same time, as the chicken advances in age, the pineal stroma becomes denser and richer in collagenous fibers, In the examined pineal glands of older chicken (16 and 34 months; fig. 11), the parenchyma appears to be divided into small territories, incompletely separated by thick conjunctive partitions, with abundant collagenous fibers. The combination with data obtained from preparations stained with hematoxylin-eosin

permits us to affirm that these territories correspond to cellular nests in which a cellular disposition in rosette can be noted, with a practically nonexistent lumen in its center. The parafollicular zone is practically nonexistent.

Discussion

According to our observations, there is a morphological evolution of the pineal gland during post-hatching life, which is demonstrable with the optical microscope. The only earlier article concerning this process is that of *Spiroff* [1958] which describes some of the aspects of this evolution. With the optical microscope, the evolution of the pineal gland of the chicken is principally manifest in its structure. In the first days, the follicular pattern of the older embryos is maintained [*Calvo and Boya,* 1977]. As age increases, there is a progressive reduction of the follicular lumens along with their fragmentation, while the parenchyma assumes an apparently solid appearance. Nevertheless, detailed examination with the optical microscope demonstrates a great number of cavities and cellular rosette formations.

There are parallel changes in the pineal stroma, i.e. progressive fragmentation of the large pineal lobes into small territories by the penetration of interfollicular stroma. In the pineal gland of adult chickens, the parenchyma is arranged in cellular nests separated by thick irregular partitions which are collagenous-rich fibers.

According to our results, the pineal parenchyma of the adult chicken can neither be considered as solid lobular [*Gladstone and Wakeley,* 1940; *Spiroff,* 1958; *Quay,* 1965; *Wight and McKenzie,* 1970; *Pearson,* 1972] nor as an intermediate between the lobular and follicular forms [*Romieu and Jullien,* 1942]. Only the electron microscope will permit an exact characterization of the architectonics of the pineal gland of the adult chicken.

From a philogenetic point of view, the evolution of the structure of the chicken pineal gland is interesting. During the first phases of embryonic development, the pineal gland has cavities with relatively thin walls and ample lumens [*Calvo and Boya,* 1977] and is thus similar to the sacular pineal glands of the more primitive birds (Passeriformes) and other inferior species. On the other hand, in the adult chicken, the parenchyma presents a compact aspect, which is similar to the pineal glands of the mammals. *Wolfe* [1965] has written that in the various species of mammals, there are virtually always cavities.

In our series, the pineal lymphoid tissue suddenly appeared at 10–15 days, and attained its maximum development before 30 days. According to *Romieu and Julien* [1942], the lymphoid tissue appears at 30 days and attains its maximum development at 3–6 months. *Spiroff* [1958] found lymphoid cells in the walls of vessels of the pineal gland at 12 days and in the pineal gland at 18 days. For this author, the maximal development is attained at 3 months. The aspect of the nodules is similar to that described by *Romieu and Jullien* [1942], *Spiroff* [1958] and *Quay* [1965]. In no case did we find a massive invasion of pineal parenchyma, as is seen in other organs of the chicken, e.g. in the pancreas, liver, kidney [*Hodges,* 1974]. The significance of the pineal lymphoid tissue remains unknown [*Hodges,* 1974]. Nevertheless, it has to be considered as a habitual component in the chicken pineal gland after hatching.

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J. Boya, Department of Histology and General Embryology, Faculty of Medicine, University Complutense, Madrid (Spain)