

Embryonic Development of Spinal Cord in White Choeked Bulbul (*Pycnonotus leucotis*)

Asmaa Basheer Abed¹, Aws Basheer Abed²

¹Lecturer, Department of Biology, College of Education for Pure Science (Ibn Al-Haitham), University of Baghdad, Baghdad, Iraq, ²Senior House Officer in General Surgery CABS, FICMS, Baghdad Teaching Hospital, Baghdad, Iraq

Abstract

The study concerned the embryonic development of spinal cord of white choeked bulbul *Pycnonotus leucotis*. The spinal cord at the age 40 hour's incubation from the neural tube and its tissue wall is composed of two layers, the ependymal layer and mantle layer. The spinal cord differentiation is completed at the age 7 day's incubation, its, were we notice that the central canal is clear and cavity lined with neurons and we note the arrangement of gray matter in the form of the letter H and its inside and it is characterized by dorsal and ventral horns and the gray matter substance and note the formation of dorsal medium spetum and ventral medium fissure.

Keys Words: Spinal cord, Embryonic development, Birds.

Introduction

The white cheeked bulbul *Pyconontus leucotis*, family Pycnonotudiae, order Passerifromes, class Aves and phylum Chordata. The neural tube arises from the neural plate during neurulation, the neural plate results from induction of primitive ectoderm during neural induction⁽¹⁾.

Neural system development is one of earliest systems to begin, and the last to be completed after birth, this development generates the most complex structure within the embryo and the long time period of development means in utero insult during pregnancy may have consequences the development of the nervous system^(2, 3).

The neural tube gives rise to brain and spinal cord, the neural tube is formed from the ectoderm overlying the notochord and ectoderm extends from prochordal plate to the primitive knot, the neural tube is soon divisible in to cranial enlarged part that forms the brain, and a caudal tubular part that forms the spinal cord⁽⁴⁾. The brain and spinal cord develop as unequal swellings at different regions of the neural tube. Caudal cylindrical portion of the neural tube forms the spinal cord. Cavity

of the neural tube is almost like a vertical slit as seen in the cross section⁽⁵⁾.

The spinal cord is one part of the central nervous system, that connects to the myelencephalon and extends within the vertebral canal⁽⁶⁾. The spinal cord is symmetrical on both sides and there are a pair of incisions the dorsal medium sptum and the ventral medium fissure. It is composed of two internal area called the gray material, which appears in the form of layer consisting of frontal and posterior horns and consisting of nerve ganglia cells as well as the fibers contained^(7, 8).

The external area represents the white material and consist of bundles of nerve fibers, which are sensory and motor nerve fibers^(9, 10, 11). The spinal cord in its center contains a small space called the central canal, which is filled with cerebrospinal fluid and extends along the spinal cord as it reaches the front end of the fourth ventricle^(1, 12, 13).

Nervous tissue consists of cells, fibers and blood vessels, two different categories of cells are found in nervous tissue neurons and neuroglial cells. The meninges en copsulates brain and spinal cord, and the meninges are formed in birds as in the other vertebrates

of the dura matter, arachnoid and pia matter^(10, 14)

Material and Methods

Embryos bird *Pycononotus leucotis* were obtained from incubated eggs collected from local market. For light microscope study, the specimens imprisoned in 10% formalin to be fixed, then the specimens washed with tap water, and dehydrated in ascending solutions of ethyl alcohol, the specimens were cleared in xylene, then they were infiltrated and embedded in paraffin wax. Thin serial sections (5 µm) were cut and rotary microtome, and stained with hematoxylin and eosin. These sections were examined and measurement by using ocular and stage micrometer and photographed using an light microscope with camera.

Result

The spinal cord of the embryo age (40) hours incubation shows on oval shape in the cross section, and the thickness of its roof and its height was (13) micrometer, while the thickness of its later wall was (38) micrometer. The wall consists of ependymal layer, which it made up of pseudostratified columnar epithelium surrounding the neurocoel, which represents the spinal cord cavity as it is narrowed at the central part and the followed by the layer of mantle layer, which it contains of neuroblasts that it is active division (Figure 1).

In the fetus (48) hours incubation the spinal cord was distinguished its oval shape, consisting of a neuronal matrix and it composed of several rows cells. The cells of ependymal layer are active split and spherical shape. The ependymal layer either the cells that followed and the result of the division of cells layer nerve lining aggregates to from the precesser mantle layer of neuroblasts, and the thickness of roof was (15) micrometer, the rate of thickness of the hall had reached (19) micrometer, while the average thickness of the lateral wall was (64) micrometer. The cavity of spinal cord appeared narrow in the central region anal expanded in the dorsal and abdominal regions.

The section of the fetus (72) hours incubation, the spinal cord is oval shape and it was composed of the ependymal layer, which it consist of the epithelial tissue followed by neuroblast layer, which extends its protrusions to from the marginal layer, and differentiation of the mantle layer to gray matter and differentiation of

the marginal layer to the white matter, the spinal cord contains coelom called newocoel. The rate thickness of roof of spinal cord was (18) micrometer, the rate of thickness of the floor was (25) micrometer, and the rate of thickness of the later wall was (80) micrometer (Figure 2).

The spinal cord in the (96) hours incubation appears to be oval of the shape with a gray matter that represents the inner part of the wall, and the white matter that represents the outer part of its wall, and it is noted the beginning of differentiation of gray matter into the dorsal horns, and ventral horns, the rate of thickness of roof was (30) micrometer, the rate of thickness of the wall of floor was (45) micrometer, and rate of thickness of the later wall was (140) micrometer, and the spinal cord contain elongated cavity (Figure 3).

The fetus (5) days' incubation, the rate of thickness of roof was (59) micrometer, the rate of thickness of floor was (69) micrometer, and the lateral wall was (149) micrometer, and its contains elongated cavity. As for the structure of the tissue showed the gray matter which was synthesized by spherical neurons while the white matter composed of the nerve fibers, as well as the appearance of dorsal horns and ventral horns (Figure 4).

The spinal cord in the (6) days' incubation the rat of thickness of roof was (65) micrometer, the rat of thickness of floor was (75) micrometer and the rate of thickness of the lateral wall was (152) micrometer. As for the structure of the tissue has distinguished the wall of its roof to the roof plate, whose cells are active split in the form spherical neurons and nuclei central location.

As for its lateral wall, ependymal was shown to be a split nucleus with a central site of dark color followed by several rows of cells with central nuclei, these cells represent the mantel layer, which forms a gray matter of spinal cord surrounded by the exterior, the marginal layer represents white matter. The histological structure of the wall floor has differentiation to the floor plate (Figure 5).

At age (7) days' incubation increase in the side walls, where the average thickness of rood was (163) micrometer, as well as the rate of thickness of floor was (70) micrometer, while the average thickness of the lateral wall of the floor of (79) micrometer. The spinal

cord differentiates at this age into areas, the white matter outside and the gray matter inside, which appears in the shape of the letter x, this region differentiates to the dorsal and ventral horns, and it is notice at this age dorsal medium septum and ventral medium fissure (Figure 6).



Fig 1: T.S. through embryo at 40 hour's incubation showing spinal cord, notice: Ependymal layer (EP), Mantle layer (ML), Notochord (NO) (H & E, 100x).

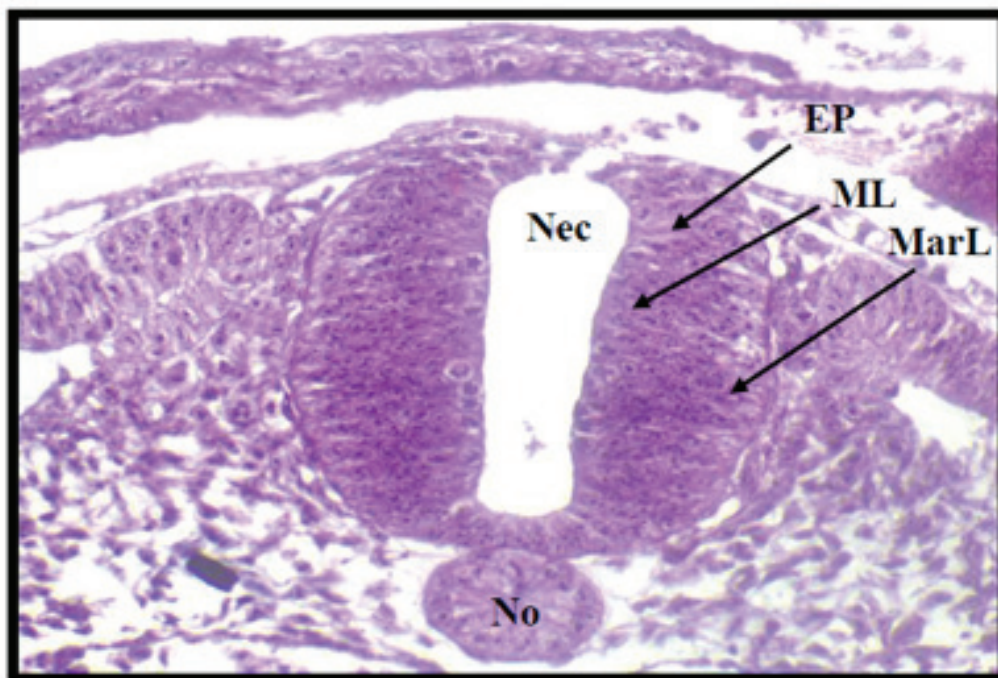


Fig 2: T.S. through embryo at 72 hour's incubation showing spinal cord, notice: Neurocoel (Nec), Ependymal layer (EP), Mantle layer (ML), Marginal layer (MarL), Notochord (NO) (H & E, 40x).

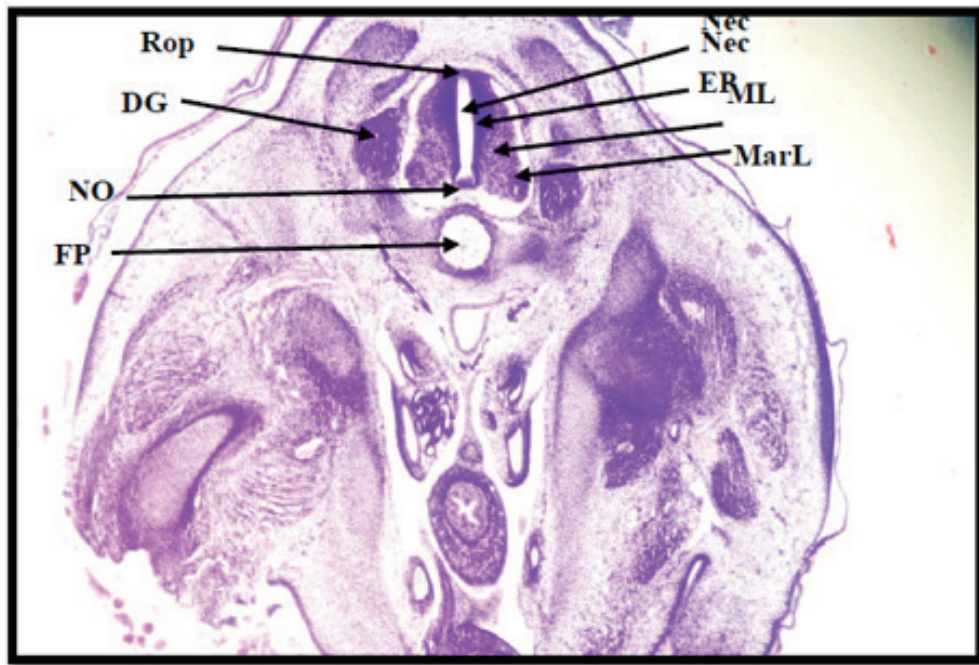


Fig 3: T.S. through embryo at 96 hours incubation showing spinal cord, notice: Neurocoel (Nec), Ependymal layer (EP), Mantle layer (ML), Marginal layer (MarL), Roof plate (Rop), Floor plate (FP), Notochord (NO), Dorsal root ganglion (DG) (H & E, 10x).

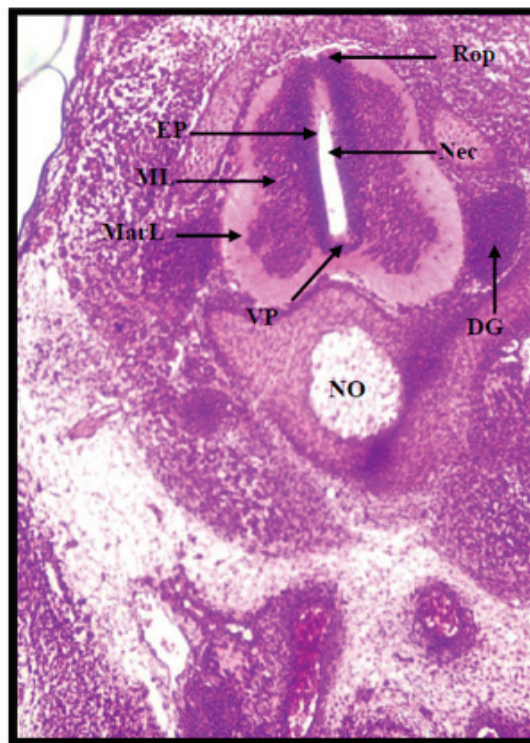


Fig 4: T.S. through embryo at 5 day's incubation showing spinal cord, notice: Neurocoel (Nec), Ependymal layer (EP), Mantle layer (ML), Marginal layer (MarL), Notochord (NO), Roof plate (Rop), Floor plate (FP), Dorsal root ganglion (DG) (H & E, 10x).

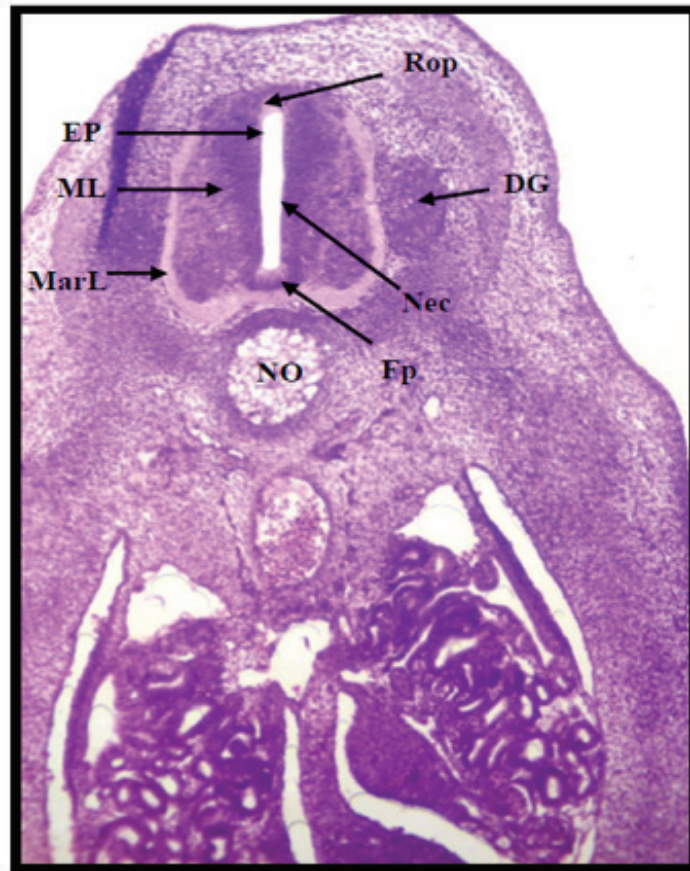


Fig 5: T.S. through embryo at 6 day's incubation showing spinal cord, notice: Neurocoel (Nec), Ependymal layer (EP), Mantle layer (ML), Marginal layer (MarL), Roof plate (Rop), Floor plate (Fp), Notochord (NO), Dorsal root ganglion (DG).

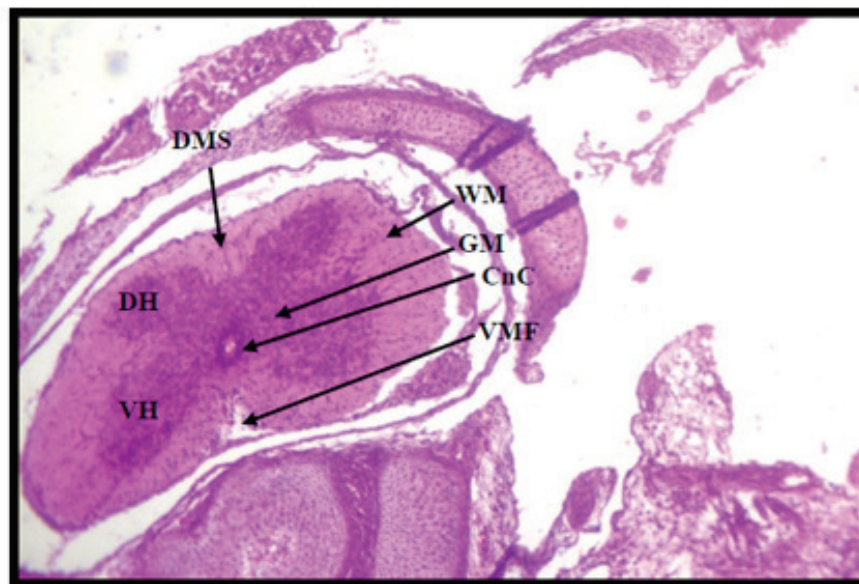


Fig 6: T.S. through embryo at 7 day's incubation showing spinal cord, notice: Central canal (CC), Dorsal horn (DH), Ventral horn (VH), Dorsal medium sptum (DMS), Ventral medium fissure (VMF), White matter (WM), Gray matter (GM) (H & E, 10x).

Discussion

The caudal segment of the neural tube becomes the spinal cord, it gradually tapers in the tail region where terminates as the primitive filum terminale. It is formed by the spinal cord of the neural tube which does not form the brain.

This part of the neural tube which was initially cylindrical thickens differently at different regions to form the adult spinal cord⁽¹⁵⁾.

It swells in girth at the level of the arms and legs. The original cylindrical neural canal elongates dorso-ventrally and gets compressed laterally. The neural tube consists of the thickened columnar epithelium, but changes soon take place in its shape and histological structure. All the cells of neural tube are elongated with their long axes radiating out from the ependyma to periphery. The neural tube possesses two layers, the ependyma, which lines the neural canal and contains a large number of mitotic cells and the marginal layer, and the mantle layer is also recognizable⁽¹⁶⁾.

Neuroblasts are visible in embryo from about (2 days) of incubation in the ventrolateral part of the neural tube, this region is about six cells deep, whereas in the floor plate there is only a single layer of cells. Spinal nerves have developed, and the regions of grey and white matter are recognizable at (3-3.5 days) of incubation.

Dorsal and ventral horn can be seen in the grey matter and glial cells in the white matter at (7 days) of incubation. During the following days the spinal cord becomes larger in transverse section and there is change in shape of lumen from a longitudinal slit to an almost square or round shape.

The wall is thin in the midline dorsally and ventrally forming roof and floor plates respectively, these areas will save path way for crossing nerve fibers. The sulcus limitans divides the mantle layer in the lateral wall into a dorso lateral portion called the alar plate, which contains sensory nuclei and a ventrolateral portion called the basal plate, which contains motor nuclei. This is accompanied by the development of dorsal and ventral fissure, with the outgrowth of spinal nerves size and shape of the spinal cord varies along its length. Its wall in the dorsal part fuses to form a suture, the dorsal suture. Its ventral part remains a canal of spinal cord

and it called central canal. Median line to enclose a mid-ventral groove, the ventral fissure.

A fate map of the spinal cord in the thoracic, lumbar and caudal regions were published⁽¹⁷⁾. The meningeal layers of the spinal cord, and the endoneurium of the peripheral nerves were all formed from mesenchymal cells⁽¹⁸⁾.

Conclusion

The spinal cord is differentiated from the neural tube at the age embryo of (40) hour's incubation, and its wall is histologically composed in this age of two layers, the embryonic layer and mantle layer. At the embryo age (48) hour's incubation increases the thickness of the wall of the spinal cord. The marginal layer is differentiated with in the spinal cord wall in embryo age (72) hour's incubation. The spinal cord differentiates into two areas, the gray matter region, which is formed by dorsal horns and ventral horns, and the white matter region in embryo age (96) hour's incubation. The embryo age of (5) day's incubation, the area of the gray matter a histologically-formed cell of spherical neurons. As for the area of white matter it is a histologically-formed of the nerve fibers, as the dorsal and ventral horns observe. The roof and floor plate cells and spinal cord wall cells are dividing active at the embryo age of (6) day's incubation. At the embryo age of (7) day's incubation can be observed, the central canal, the dorsal and ventral horns, the dorsal median suture and ventral median fissure.

Ethical Clearance: The Research Ethical Committee at scientific research by ethical approval of both MOH and MOHSE in Iraq

Conflict of Interest: None

Funding: Self-funding

References

1. Sadler TW. Langman's medical embryology. 12th ed., Lippincott Williams and Wilkins, New York, 2012: 371 pp.
2. Bortier H, Vakaet LCA. Fate mapping the neural plate and the intraembryonic mesoblast in the upper layer of the chicken blastoderm with xenografting and time-lapse videography. In gastrulation (ed. C Stern, P Ingham). Development, 1992: 93-97.

3. Gatesy SM, Dial KP (1996). From frond to fan: *Archaeopteryx* and the evolution of short tailed birds. *Evolution*, 1996; 50: 2037-2048.
4. Leber SM, Sanes JR. Migratory paths of neurons and glia in the embryonic chick spinal cord. *J Neurosci*. 1995; 15: 1236-1248.
5. Gibert SF. *Development biology*. 6th ed. Sinauer Associates. Inc, London. 2000; 9: 749 pp.
6. Carvalho RC, Sousa AL, Oliverira SR, Pinto ACB, Fontenelle JH, Cortopassi SRG. Morphology and topographic anatomy of spinal cord of the red-footed tortoise (*Geochelone carbonaria* Spix, 1824). *Pesq Vet Bras*. 2011; 31: 100-736.
7. Hughes AFW. The growth of embryonic neuritis a study of culture of chick neural tissues. *J. Anat. Lond.*, 1953; 87: 62-150.
8. Dial KP. Evolution of avian locomotion correlates of flight style, locomotors modules nesting biology, body size, development and the origin of flapping flight. *The Auk.*, 2003; 120: 941-952.
9. Kardong KV. *Vertebrates comparative anatomy, function evolution*, 2nd ed., WCB McGraw-Hill, Co., Inc., New York, 1998: 747 pp.
10. McKinley M, Oloughlin VD. *Human anatomy*, McGraw-Hill Com., Inc., New York, 2006: 888 pp.
11. Delalande JM, Thapar N, Burns AJ. Dual labeling of neural crest cells and blood vessels with in chicken. Embryos using chick GFP neural tube graining and carbocynine due Idil injection 2015, e52514.
12. Nauta W, Feirtag G. Ontogeny spinal cord. In *fundamental neuroanatomy* WJH. Nauta and G. Feirtag, New York, 1986: 134-161 pp.
13. Baumel JJ. *Handbook of avian anatomy*. Nuttall orthological club. Cambridge., 1993: 779 pp.
14. Gardner DK, Lane M. Culture of viable blastocysts in defined sequential serum-free media. *Hum Reprod*. 1998; 13(Supp.13): 148-159.
15. Abed AB. Embryonic development and histogenesis of brain and spinal cord in quail *Coturnix coturnix* (Linnaeus, 1758). PhD. Thesis, College of Education for Pure sciences Ibn Al-Haitham, University of Baghdad, 2016: 251 pp.
16. Smith JL, Schoenwolf GC. Notochordal induction of cell wedging in the chick neural plate and its role in neural tube formation. *J Exp Zool*. 1989; 250: 49-52.
17. Catala M, Teillet MA, Derobertis EM, LeDouarin ML. A spinal cord fate map in the avian embryo: while regressing Hansen's nod lays down the notochord and floor plate thus Joining the spinal cord lateral walls. *Development*. 1996; 122: 2599-2610.
18. Selleck MAJ, Brunner-Fraser M. Origins of the avian neural crest: the role of neural plate epidermal interactions. *Development*. 1995, 121: 525-538.