IV. *The Development of the Vertebral Column in Mammals, as illustrated by its Development in* Mus musculus.

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[Plates 11–14.]

Contents.

Introduction.

The research work to be described was carried out upon the suggestion of Prof. E. W. MacBride. Some justification for it may be found in the prevailing dual conception of the mode of development of the vertebral column in mammals, since notwithstanding the researches of Remak (1851), Rosenberg (1876), Froriep (1886), Weiss (1901), Bardeen (1905), Barge (1915) and others, opposing views are held as to whether or not the vertebræ of mammals are segmental structures. The work of Froriep maintains that they are; that of Weiss, Bardeen and others that they are not. The aim of the present investigation is to attempt to impart some finality to the argument and to settle the controversy that exists concerning the relation between

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the sclerotomes and the membranous and cartilaginous incipient vertebræ with the intervertebral accessorial structures. An attempt has been made to indicate the nature of the modifications effected in the region of the atlas and axis, and special consideration has been given to the so-called pro-atlas. In addition, the nature and number of the occipital segments have been observed, and the part played by these in the formation of the occipital region of the cranium has received attention.

The investigation has been carried out in the Huxley Research Laboratory of the Imperial College of Science and Technology, South Kensington, under the supervision of Prof. E. W. MacBride, F.R.S., to whom my sincere thanks are tendered for kindly criticism and useful advice afforded during the course of the research. I also desire to express my gratitude to Prof. J. P. Hill, F.R.S. (University College, London), for criticism and technical assistance, to Prof. E. S. Goodrich, F.R.S., who kindly lent me a copy of Jager's paper, and to Prof. Graham Kerr (Glasgow University), for supplying details of some methods of reconstruction. My thanks are also due to Mr. H. R. Hewer (Imperial College), for photographic assistance and for useful criticism.

Materials.

The embryos used in this work were those of *Mus musculus*, the common white mouse. When the parent animals were not actually bred by the author, they were bred under his strict supervision by a skilled breeder. The parents were in all cases perfectly healthy and, as far as could be ascertained, quite normal individuals. Altogether, over 200 embryos forming more than 30 stages were obtained, and from these the ones most suitable for the purpose were selected for the work in hand.

Methods.

The embryos were removed from the fœtal membranes and placed directly into the fixing fluid, excepting those smaller than 3·0 mm., which were preserved *in utero.* Since it was not found possible to determine the ages of the embryos accurately, the "crown-rump" measurements, often employed by mammalian embryologists, have been used.* As the curvature of the embryo constantly changes until a certain stage is reached, this system is open to objection, yet not so much so as is an uncertain determination of age. The litter-number and the type of diet were observed in all cases, although it has not been possible to make allowance for slight variations in size arising out of variations in these factors (*vide* Stotsenberg, J. M., 1915). The fixing fluid used was Bouin's, which has given uniformly excellent results, and the amount of distortion was reduced to a minimum by the method of removing the embryos from the uterus prior to fixing. After dehydration, Hill's method of clearing and imbedding has been used, with considerable satisfaction. Celloidin imbedding was found to be quite unnecessary. Continuous transverse, sagittal and frontal serial sections were cut and graphic reconstructions were made from the transverse series.

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* In the following text, "crown-rump length" is indicated by the abbreviation "C.R.Lth."

Camera-lucida drawings of the embryos were made while these were still in the clearing agent, and proved invaluable in providing a curved base-line for the reconstructions, the dorsal boundary of the nerve cord being used for this purpose. As both camera-lucida drawings and reconstructions were made on squared paper, it was a simple matter to convert the scale of the former into that of the latter. The reconstructions were not considered to have fulfilled their purpose until they had been carefully compared with sagittal and frontal sections, and a reasonable measure of conformity was obtained. Sections of early stages before complete tissue differentiation were stained in DELAFIELD's hæmatoxylin, while later stages lent themselves very well to MALLORY's triple stain. These out of a number of selected stains gave the best results. For osseous stages a decalcifying mixture composed of 70 per cent. alcohol with 3 per cent. nitric acid was used, this acid being found more satisfactory than hydrochloric. The Text-figures 1, 5, 6 and 7 are adapted graphic reconstructions, which serve the best purpose in the form of the stereogram. The plate figures are from micro-photographs prepared at the Imperial College of Science.

Embryo of Mus musculus, 2·0 *mm. C.R.Lth.*

The notochord is a very slender rod of cells extending throughout the length of the embryo. Anteriorly it is oval in cross-section, the transverse axes measuring 0·035 mm. and 0·025 mm. respectively. The long axis is horizontal and 4 or 5 nuclei are seen in a section 0·008 mm. thick. Towards the much-curved posterior extremity of the embryo it is much more flattened, forming a plate of cells closely apposed to the ventral surface of the nerve cord, there being no mesenchymatous cells between the two structures. Here the axes measure 0·080 mm. and 0·025 mm. respectively, and 7 or 8 nuclei are seen in the same section. Throughout the length of the notochord, the cytoplasm of the notochordal cells, projecting beyond the nuclei, forms a very rudimentary sheath, the peripheral part of which is indicated as a thin dark line.

The rudiments of the dorsal spinal ganglia are formed as loosely arranged clusters of cells midway between the dorsal and ventral boundaries of the nerve cord, which is somewhat compressed laterally, so that its canal is almost obliterated, except dorsally and ventrally, and so that the ganglia rudiments lie in the concavity of its lateral margin (Plate 11, fig. 1, *sp.g.*).

The masses of mesoderm flanking the spinal cord are formed into the primitive somites (= protovertebræ), while mesenchymatous cells with spindle-shaped nuclei and long cytoplasmic processes surround the notochord (except posteriorly) and the principal blood vessels (Plate 11, fig. 1). The posterior parts of the primitive somites have already become partially differentiated into dermato-myotomes and sclerotomes. The cells of the inner wall of this part of the somite are rapidly multiplying and the whole of this wall has become converted into a thick cluster of cells, held together only by the most mesially situated ones, which still form a continuous strand. Already some of the sclerotogenous cells have been liberated from the postero-dorsal part of the primitive somite and have come to lie among the mesenchymatous cells in the region between the primitive somite and the nerve cord (Plate 11, fig. 1, *scl.*).

In front of the point of fusion of the lateral aortæ, 5 segments are seen to be in a more rudimentary condition than the typical segment described above. The 5th or most posterior one possesses a pair of sclerotomes and dermato-myotomes which are almost typically developed and almost as extensive dorso-ventrally as those of more posterior segments, but it is characterised by its much smaller dorsal ganglionic cell-cluster. The 4 anterior segments have smaller and less differentiated somites and none of these segments possess rudimentary ganglia.

Throughout the whole length of the embryo intersegmental arteries occur between successive differentiating segments, including the rudimentary anterior ones, and similarly in front of the most anterior of these. The arteries extend dorsally to the horizontal level of the bases of the dorsal ganglia rudiments where these are developed, and in the anterior region of the embryo they are equally well developed and of similar extent. Intersegmental veins have not yet appeared.

Between the developing otic capsule and the most anterior rudimentary somite is a region, slight in antero-postero extent, where the developing central nervous system is flanked by mesoderm in which there is no sign of segmentation, there being no dermato-myotomes or sclerotomes present.

Embryo of Mus musculus, 3·0 *mm. C.R.Lth.*

The notochord is not a uniform rod throughout its length. At the transverse level of the otic capsule, and therefore near its anterior extremity, it is drawn out into a slender strand of cells, only two nuclei thick and about 0·020 mm. in diameter. In the middle region of the embryo it is larger, showing 6 or 7 nuclei in section, and is oval in outline but slightly flattened dorsally, the axes measuring 0·040 mm. and 0·029 mm. respectively. Towards the posterior end of the embryo it is truly oval in outline, the axes measuring 0·040 mm. and 0·020 mm. The notochord is also situated at a distance from the central nervous system anteriorly, being nearer about the middle of its length and closely apposed to the nerve cord posteriorly. Throughout the whole length of the notochord, the cytoplasm of the notochordal cells is clearly differentiated into two zones, an inner granular layer which stains faintly, and a thin outer layer which remains unstained and clear, this latter being the first indication of the definitive sheath rudiment.

The embryonic tissues are generally much looser than in the previous stage, as if the embryo had increased in size by a more complete separation of its component cells (Plate 11, fig. 2). The primitive somites at and posterior to the level of the point of fusion of the dorsal aortæ and the ductus Cuvieri, have all reached the same stage of differentiation. The entire wall of each somite, from the region immediately below the sharply defined apex of the dermato-myotome to the horizontal level of the ventral boundary of the nerve cord, has broken down. In this way, a fairly dense mass of sclerotogenous tissue, having the shape of a wedge, has come to lie on each side of the notochord, the point of the wedge reaching ventrally as far as the level of the lower boundary of the nerve cord (Plate 11, fig. 2, *scl.*). Mesenchymatous tissue, histologically and genetically different from the sclerotogenous tissue, completely surrounds the notochord (Plate 11, fig. 2, *p.mch.*, *n.ch.*).

The 5 anterior rudimentary segments in front of the point of fusion of the lateral aortæ can still be recognised. The most posterior one has retained its ganglionic cluster, which is very small, and it has a fairly well developed sclerotome, which, however, does not extend so far ventrally as a typical one. Its dermato-myotome is also well developed. Of the 4 anterior segments, none of which possess dorsal ganglia rudiments, the most anterior one is least developed, having a very small dermato-myotome and a very loosely arranged sclerotome (Plate 11, fig. 3, *a.r.d.my.*, *a.r.scl.*). The 2nd, 3rd and 4th segments are similar to the 5th, except that the sclerotogenous tissue is less condensed. Loose mesenchymatous tissue still characterises the region immediately posterior to the otic capsule, which remains devoid of segmentation. Subsequent development proves that the 5 anterior rudimentary segments give rise to the occipital region of the cranium, and to facilitate simplicity in description these segments will be referred to henceforth as the occipital segments.

The intersegmental arteries are large and well developed, extending close to the nerve cord on each side as far as its dorsal boundary. Branches from these arteries now pass mesially below the nerve cord, almost but not quite reaching the sagittal plane. Intersegmental veins have now appeared and are as well developed as the arteries (Plate 11, fig. 3, *i.s.v.*). They extend ventrally from the ventro-mesial part of the dermato-myotome to the cardinal veins, passing in front of and slightly lateral to the intersegmental arteries. It is interesting to find that the veins are developed between the rudimentary sclerotomes of the occipital segments, as were the arteries earlier, and also, similarly, in front of the most anterior segment.

Embryo of Mus musculus, 4·0 *mm. C.R.Lth.*

The notochord is little changed from the previous stage, except that it now possesses a series of strictly segmental waves, the ventral troughs of which lie within the segment, while the dorsal crests occur in the intersegmental region. The waves are less pronounced in the anterior cervical region and are altogether absent from the occipital region. The notochordal axes in the middle part of the embryo now measure 0·045 mm. and 0·030 mm. respectively, but still only 6 or 7 nuclei are seen in a single transverse section. The clear peripheral zone of cytoplasm is still evident and the outer boundary is indicated as a thin, dark blue-stained line, the rudiment of the definitive sheath. Mesenchymatous cells are becoming closely apposed to this, their flattened and attenuated nuclei forming an outer secondary sheath.

The inner wall of the primitive somite, which was completely broken down in the previous stage to form the sclerotome, is now replaced by cells from the tip of the dermato-myotome, which cells form the muscle-plate of the segment. This structure, which extends ventrally almost to the horizontal level of the notochord, is separated from the sclerotome throughout its entire length by a narrow but well-defined parasagittal cleft. Frontal sections show that the sclerotomes are now divided each by a narrow, transverse, mid-segmental fissure into approximately equal anterior and posterior halves. This fissure, which will be referred to subsequently as the intrasclerotomic fissure, is best marked at this stage in the lateral part of the sclerotome, where it passes into the parasagittal cleft separating the sclerotome and the muscle-plate. It extends almost to the tip of the myotome dorsally and almost to the horizontal level of the notochord ventrally. It does not extend to the sagittal plane mesially, traversing little more than half the region between the myotome and the notochord.

The posterior sclerotome half consists of densely aggregated cells with oval nuclei, which are radially disposed with respect to the notochord. The condensed tissue so formed, in which the nuclei are so crowded as almost to touch, extends ventrally almost as far as the notochord (Plate 11, fig. 4, *k.scl.t.*). The anterior sclerotome half consists of loosely arranged cells, except in the region between the dorsal spinal ganglia and the corresponding ventral roots (*vide infra*), where the nuclei are quite as closely packed as in the posterior sclerotome half. Cells are also condensed on the anterior border of the intrasclerotomic fissure (Plate 11, fig. 4, *c.scl.t.*). In order to render further description more clear, the anterior sclerotome half will be referred to as the cranial sclerotomite, the posterior half as the caudal sclerotomite.

The ventral spinal nerves, the first appearance of which marks this stage, each consist of a stout unbranched ramus extending ventrally and laterally from its point of origin in the ventro-lateral part of the spinal cord, towards the ventralmost part of the corresponding dermato-myotome, which it serves. Being formed, like the corresponding spinal ganglion, in the anterior part of the segment, it passes through the tissue of the cranial sclerotomite, but tending to pass towards the anterior boundary of its segment, it is found slightly posterior to the intersegmental blood vessels, which lie in the intersclerotomic fissure marking this boundary.

The most anterior occipital segment does not develop ventral spinal nerves, but instead loses its dermato-myotome and becomes vestigial. The four remaining occipital segments all develop spinal-nerves, which are separate and distinct and occur in the anterior parts of their segments (Plate 11, fig. 5, ii, iii, iv and v). As before, only the most posterior occipital segment possesses dorsal ganglia, although all four remaining ones have sclerotomes. Only in the last sclerotome is a division into anterior and posterior halves possible, and even in this there is no sign of the intrasclerotomic fissure. The two anterior sclerotomes consist of fairly loosely arranged cells throughout, while even looser tissue occupies the region immediately behind the otic capsule, which region now corresponds to the 1st occipital sclerotome vestige plus the tissue unsegmented *ab initio.*

Since the previous stage there has been a slight change in the external form of the embryo, a change which has already effected an alteration in the topographical relationship between the occipital segments and which, becoming more pronounced in subsequent stages, is destined still further to affect this relationship, as well as that between the occipital segments and those of the future cervical region. The cervical or neck flexure has appeared, and although at present it is but slightly developed, yet the anterior part of the hind-brain is appreciably bent ventralwards, so that the otic capsule now lies below the horizontal plane passing through the notochord in the future cervical region (Text-fig. 4 (*b*)). The flexure has already brought about a slight crushing of the ventral parts of the occipital segments and has effected a closer approximation of the ventral occipito-spinal nerves (Plate 11, fig. 5, ii, iii, iv and v). These nerves are distributed in the myotomes situated dorsal to the cervical sinus, which is being developed at this stage. The maximum curvature of the cervical flexure at this stage is in the region of the penultimate occipital segment.

Embryo of Mus musculus, 4·8 *mm. C.R.Lth.*

The notochord is slightly more circular in transverse outline, but apart from this is unchanged from the previous stage. The cells of the dermatome display the arrangement observed previously, but those of the myotome have multiplied rapidly and this structure now possesses a characteristic inward bulge just above the level of the notochord (Plate 11, fig. 6, *my.*). The sclerotomes have become more completely differentiated and extend considerably further ventrally. Two formations are now evident in the caudal sclerotomite which, although genetically related are nevertheless quite different. They divide the sclerotomite into dorsal and ventral halves. The nuclei of the cells of the dorsal half are fairly deeply stained, closely aggregated and elongated in form, with their long axes arranged radially with respect to the notochord, while those of the ventral half are lightly stained, less crowded and indicate no definite arrangement. The line of demarcation between these two formations passes from the inward bulge of the myotome to the notochord (Plate 11, fig. 6). The dorsal formation, being situated above the notochord and immediately in front of the intersegmental blood vessels and the ventral roots of the spinal nerves, is the membranous forerunner of the Basidorsal (GADOW), while the ventral formation lying below the notochord and occupying a similar position with respect to the blood vessels and nerves must correspond to the incipient Basiventral.*

Cells pass inwards from the basiventral primordia and, fusing subchordally, give rise to a strand of condensed tissue below the notochord. This latter structure is bounded only by mesenchymatous cells on its dorsal and lateral aspects (Plate 11, fig. 6, *n.ch.*).

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* GADOW finds that in an embryo of Scyllium the differentiating somite gives rise to dorsal and ventral clusters of sclerotogenous cells, which in turn produce dorsal and ventral "pyramids," the apices of which are directed in each case towards the notochord. The basidorsal is derived from the dorsalmost part of the dorsal pyramid; the basiventral from the ventralmost part of the ventral pyramid. The apex of

The tissue of the cranial sclerotomite consists of loosely arranged cells, except on the anterior border of the intrasclerotomic fissure, where in the dorsal half of the segment there is a slight condensation of cells, and in the region between each dorsal ganglion and the corresponding ventral root, where densely crowded cells constitute a definite formation. This formation, lying in the cranial sclerotomite immediately in front of the membranous basidorsal and immediately behind the intersegmental blood vessels, must correspond to the membranous forerunner of the Interdorsal (*vide* previous footnote). In view of the fact that a formation appears later in the dorsal half of the cranial sclerotomite, but below this structure, it must represent the dorsalmost part of the interdorsal and hence will be referred to in subsequent description as the Dorsal-interdorsal. The intrasclerotomic fissure is even more pronounced than in the previous stage, extending almost throughout the sclerotome.

The ventral part of the 1st cervical sclerotome is even at this stage much more considerably developed than the corresponding part of more posteriorly situated segments. It is composed of a larger number of cells, occupies a greater volume and extends slightly further ventrally. In this segment the dorsal ganglia and the ventral nerve roots are not associated as they are in other segments. Dorsal nerve roots have not yet appeared in any region.

As in the previous stage, the occipital segments are characterised by the four ventral occipito-spinal nerves and by the solitary dorsal ganglion of the most posterior segment, which ganglion is attenuated along the course of the spinal accessory nerve (= Cranial xi) and extends throughout the last two occipital segments. Of the occipital sclerotomes, the most posterior alone consists of a dense posterior half and a loosely arranged anterior half, although there is no sign of an intrasclerotomic fissure in this or any of the occipital segments. The three anterior occipital segments consist of fairly loose tissue in the ventral part, but more dense tissue lateral to the hind brain. The notochord is widely removed from the nervous system in this region. The ventral nerves are the only indication of the primary segmentation, these passing separately between the sclerotomic aggregations. But the cervical flexure has become very pronounced, the hind-brain being inclined to the spinal cord at an angle of about 120° (Text-fig. 4 (*c*)). Accordingly, the subchordal and perichordal parts of the occipital segments are more completely crushed and the ventral nerves are gathered together as they emerge from the sclerotome to form the compound hypoglossal nerve (= Cranial xii).

the dorsal pyramid grows downwards to give rise to a formation lying between successive basiventrals, which is the interventral, while the apex of the ventral pyramid grows upwards to form a structure lying between adjacent basidorsals, which is the interdorsal. GADOW applies these findings to all vertebrates.

It is impossible to conform absolutely to GADOW's nomenclature as his findings do *not* apply to all members of the Vertebrata (*vide* Discussion, Sclerotomes). Indeed in the herring, according to the work of Prof. RAMANUJAM, a dorsal cluster of cells such as GADOW described occurs, but only to give rise to a ray of the dorsal fin and not to a part of the vertebra, as GADOW maintained. In the present work, the terms used by GADOW are applied as defined, the basalia being derived from the caudal sclerotomite, the interbasalia from the cranial one. This gives the closest possible approximation to the nomenclature referred to.

This is distributed in the muscle-plates just anterior to the developing heart and about the transverse level of the cervical sinus.

The region between the otic capsule and the 1st occipito-spinal nerve is occupied by fairly densely aggregated sclerotomic cells, which undoubtedly represent the sclerotome of the 1st occipital segment. In all, therefore, the developing occipital region of the cranium represents the sclerotomal derivatives of 5 rudimentary (or vestigial) segments.

Embryo of Mus musculus, *5·5 mm. C.R. Lth.*

The notochord is almost perfectly round in cross-section, about 0·040 mm. in diameter in the anterior part of the embryo and 0·050 mm. in the middle part, showing 7 or 8 nuclei in transverse section. The peripheral portion of the notochordal cytoplasm has been converted into the definitive sheath, which stains an intense blue colour. The segmental waves are still preserved and the ventral troughs mid-segmental as before. The mesenchymatous investment is now closely adherent to the notochord, forming a conspicuous violet-staining mass outside the sheath.

There is no change in the condition of the intrasclerotomic fissure, which is best marked at the horizontal level of the notochord (Plate 12, fig. 7, i, *scl.f.*). Sclerotomic cells derived from the anterior half of the ventral division of the caudal sclerotomite, just posterior to the intrasclerotomic fissure, have now grown completely round the notochord. A few cells derived from the ventral portion of the cranial sclerotomite on the anterior boundary of this fissure also form a continuous investment round the notochord (Plate 12, fig. 7). That these perichordal cells are actively dividing is evinced by the masses of chromatin displayed in their nuclei. The condensation which is thus being formed mid-segmentally, and which is derived partly from the ventral division of the caudal sclerotomite and partly from the corresponding division of the cranial one, is the primordium of the perichordal disc. It has already partially obliterated the intrasclerotomic fissure.

The most anterior part of the cranial sclerotomite consists perichordally of loosely arranged sclerotomic cells and axial mesenchyme, and the most posterior part of the caudal one has a similar composition (Plate 12, fig. 7). The incipient basidorsals extend dorsally only slightly above the middle of the spinal ganglia, at which level is situated, in the cranial sclerotomite, the membranous forerunner of the dorsal interdorsal.

In the most anterior occipital segment, the ventral nerve has disappeared. Consequently, the sclerotogenous tissue in front of the most anterior nerve now existing is derived from the two most anterior and rudimentary segments, although there is nothing to indicate this in the present stage. This tissue now overhangs the otic capsule. The cervical flexure is slightly more pronounced (Text-fig. 4 (*d*)) and the maximum curvature occurs about an axis passing through the last and the penultimate occipital segments. The 3rd and 4th occipital sclerotomes consist of rather denser tissue than previously, but apart from this are unchanged, as is the 5th. There is little to indicate the primary segmentation of the occipital region, excepting the ventral nerves of the three most posterior segments, the cervical flexure having greatly modified this region, especially perichordally.

Embryo of Mus musculus, 6·5 *mm. C.R.Lth.*

Before commencing to describe this stage, it must be pointed out that some little differentiation has taken place since the previous stage. A gap exists in the series here, although the difference in length between this embryo and the one characterising the previous stage is merely 1·0 mm. This gap can be bridged with reasonable accuracy, but the author regrets he was unable to obtain an embryo of exactly 6·0 mm.

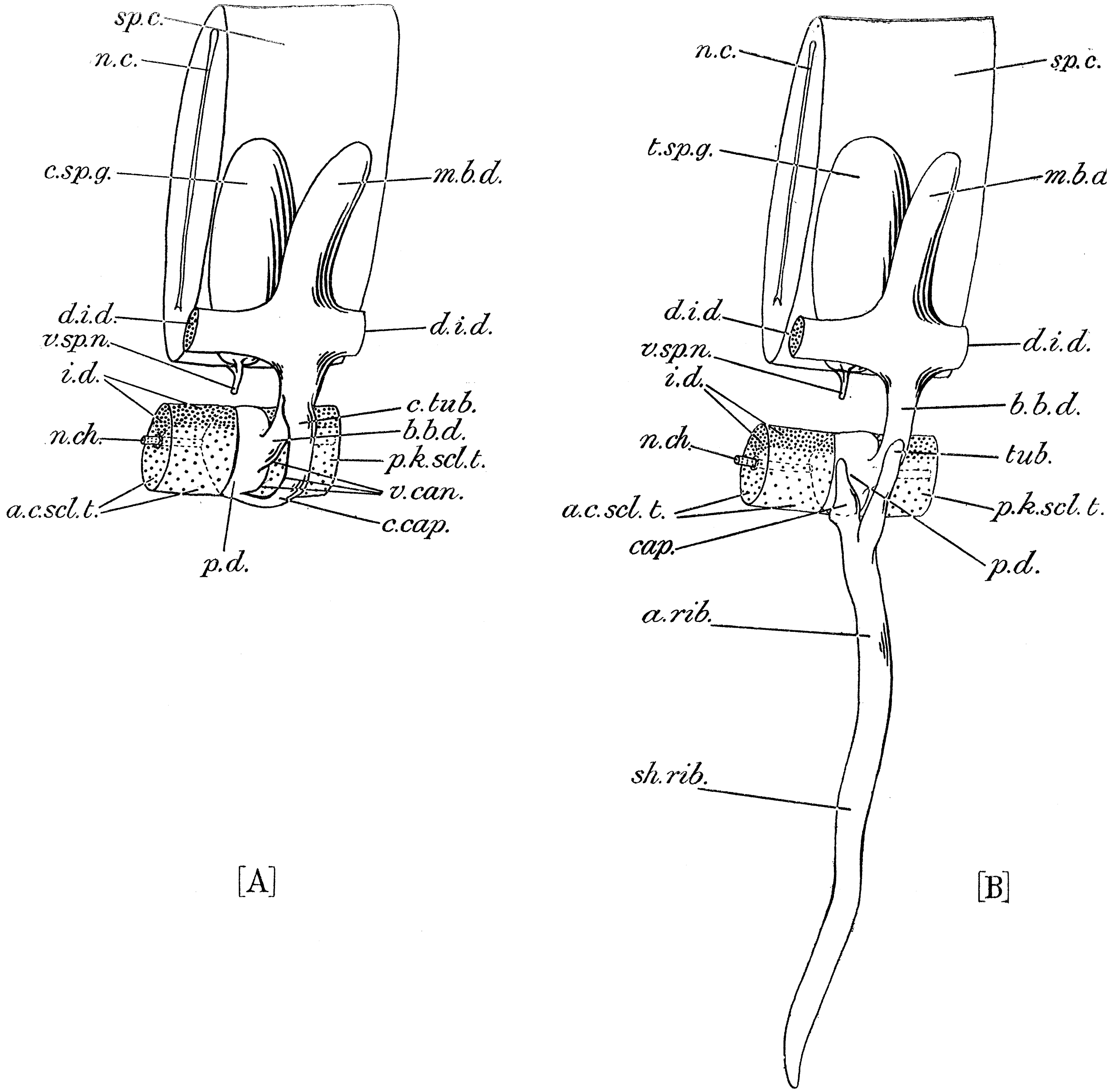
The notochord is slightly larger and shows 6 or 7 nuclei in transverse section. Small vacuoles have appeared in the cytoplasm of the notochordal cells and the sheath is thicker and more intensely stained, except in the occipital region, where it is inconspicuous also on account of its smaller size. A layer of sclerotomic cells now forms a continuous investment round it, completely masking the earlier mesenchymatous layer. The nuclei of these cells are oval or slightly flattened, and quite different from the nuclei of mesenchymatous cells. The tube of cells thus formed round the notochord may be termed the perichordal tube (Plate 12, fig. 8, *p.t.*). The segmental waves of the notochord are still evident posterior to the occipital region, but do not occur within this region.

The occipital, cervical and thoracic regions are quite well defined and the vertebral structures, although still entirely membranous, are in a fairly advanced stage of differentiation. Dorsal nerve roots have been developed and the vertebral arteries, together with the basilar artery, are complete. Intersegmentally, the vertebral artery is large, but within the segments its diameter is smaller, which fact lends support to the view that it is formed by anastomosis of the intersegmental arteries. The intersegmental arteries between the 6th and 7th cervical segments form a connection between the vertebral artery and the aorta. From this point, the vertebral artery runs forward lateral to the notochord, curving over the anterior part of the atlas rudiment to course between the floor of the hind-brain and the occipital vertebral rudiments as the A. basalis.

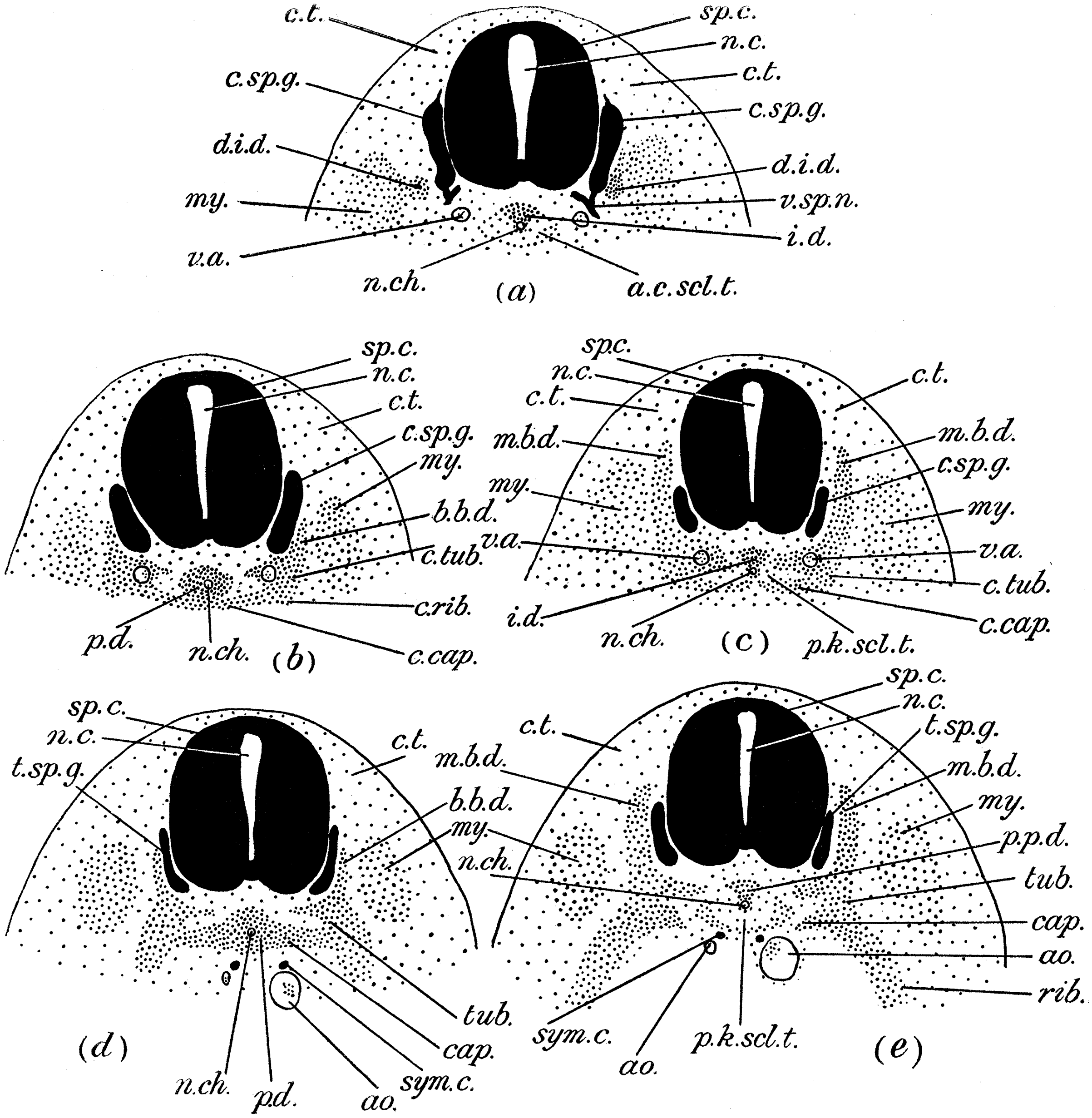
The intersegmental fissure has become almost entirely obliterated, but the boundaries of the primitive segments are still indicated by the intersegmental blood vessels (Plate 12, fig. 8, *i.s.b.v.*). A complete obliteration of the intrasclerotomic fissure has also been effected, but the perichordal disc, which was formed from the ventral portions of both cranial and caudal sclerotomites, still marks the middle of the segment. Frontal sections through the thoracic region indicate clearly, even at this stage, its origin from both sclerotome halves (Plate 12, fig. 8, *p.d.*), so that considering its present intrasclerotomic position and also its ultimate intervertebral position, it must be regarded as a true incipient intercentrum. It is a much larger structure than formerly and its nuclei show a concentric arrangement round the notochord.

In the thoracic region, the lateral part of the ventral division of each caudal sclerotomite, which is even more clearly defined from the dorsal division by its rounded nuclei, is strikingly transformed. It has grown out ventralwards, following the course of the visceral branch of the ventral nerve corresponding to the next posterior segment, to the level of the dorsal boundary of the pericardial cavity and represents the rudiment of the rib. The dorso-lateral part, which appears to be a meristem, lies at the horizontal level of the notochord and abuts on the dorsal division of the caudal sclerotomite. It is the primordium of the tuberculum of the rib (Text-fig. 2 (*e*), *tub.*). The ventro-mesial part forms the rudiment of the capitulum, a few cells from the cranial sclerotomite also contributing to this. The capitula pass into the tissue of the perichordal disc mesially, and the ventral part of the disc is differentiated into a subchordal connection between each pair of such rudiments. Posteriorly, the tissue of the disc merges into the dense tissue of the dorsal part of the caudal sclerotomite (Plate 12, fig. 8, *p.d.*, *k.scl.t.*), which now extends half way up the spinal cord between successive spinal ganglia as the membranous basidorsal (Text-figs. 1 (B), *m.b.d.*, and 2 (*d*), (*e*)). The most posterior part of the caudal sclerotomite, situated behind the diverging bases of the basidorsals, consists perichordally of loose tissue. Similarly, the most anterior part of the cranial sclerotomite, in front of the perichordal disc, consists of loose tissue, but only below the notochord. Above this structure is an apparently independently formed and wedge-shaped cluster of cells, densely crowded together. This cell-condensation, lying behind the intersegmental blood vessels and also between adjacent basidorsals, must represent the fused incipient Interdorsals (Gadow). Above it, the membranous dorsal-interdorsals connect adjacent basidorsals (Text-fig. 1 (B)).

In the cervical region, the perichordal discs have the same composition as in the thoracic, but occupy a larger part of their segments, principally on account of the perichordal crushing of these segments due to the cervical flexure. The relation of the disc to the basidorsal is also the same (Text-fig. 1 (A)). Primordia of interdorsals and dorsal-interdorsals occur in precisely the same position (Text-fig. 2 (*a*)). The boundary between the dorsal and ventral halves of the caudal sclerotomite is marked in this region by the position of the vertebral artery and is quite as well defined as in the thoracic region. The ventral half is differentiated into a ventro-lateral part, which has grown down slightly as the rudiment of a cervical rib, a dorso-lateral part, which forms the lateral portion of the vertebrarterial canal, and a ventro-mesial part, which merges into the tissue of the perichordal disc and which underlies the loose tissue in the most posterior part of the caudal sclerotomite (Text-fig. 2 (*b*), (*c*)). The ventro-lateral part is the homologue of the thoracic rib, the dorso-lateral part of the tuberculum of the thoracic rib and the ventro-lateral part is homologous with the capitulum of the rib. It must be observed, however, that the capitulum of the thoracic rib is formed in part from both sclerotomites, whereas that of the cervical rib is derived solely from the caudal one. The 7th cervical vertebral rudiment does not possess a vertebrarterial



Text-fig. 1.—A stereogram based upon a graphic reconstruction of typical cervical and thoracic segments in a 6·5 mm. embry showing the relative positions and general orientation of the various components of the developing vertebræ. The segme is supposedly cut off at its anterior and posterior limits in each case, and the view is from the side and slightly in front. T basidorsals, dorsal-interdorsals, perichordal discs and the ribs, with their processes, are rendered opaque and are shown one side only. (A), Cervical region; (B), Thoracic region; *a.c.scl.t.*, loose tissue of the most anterior part of the crani sclerotomite; *a.rib*, angle of rib; *b.b.d.*, base of membranous basidorsal; *c.cap.*, cervical capitular process; *c.tub.*, cervic tubercular process; *c.sp.g.*, cervical spinal ganglion; *cap.* capitulum; *d.i.d.*, dorsal-interdorsal; *i.d.*, interdorsal; *m.b.* membranous basidorsal; *n.c.*, neural canal; *n.ch.*, notochord; *p.k.scl.t.*, loose tissue of most posterior part of caud sclerotomite; *p.d.*, perichordal disc; *sh.rib*, shaft of rib; *sp.c.*, spinal cord; *t.sp.g.*, thoracic spinal ganglion; *tub.*, tube culum; *v.can.*, vertebrarterial canal; *v.sp.n.*, ventral spinal nerve-root.



TEXT-FIG. 2.—Transverse sections, from a continuous series, through typical cervical and thoracic segments of a 6·5 mm. embryo, showing the relative positions of the vertebral condensations. (*a*), (*b*) and (*c*) are sections through the anterior, middle and posterior parts respectively of a cervical segment, (*d*) and (*e*) through the middle and posterior parts respectively of a thoracic segment. The nerve cord and spinal ganglia are indicated in silhouette, the condensations by variety in stippling. *a.c.scl.t.*, loose tissue of cranial sclerotomite; *ao.*, aorta; *b.b.d.*, base of membranous basidorsal; *cap.*, capitulum of rib; *c.cap.*, cervical capitular homologue; *c.rib*, cervical rib; *c.sp.g.*, cervical spinal ganglion; *c.t.*, connective tissue; *c.tub.*, cervical tubercular homologue; *d.i.d.*, dorsal-interdorsal; *i.d.*, interdorsal; *m.b.d.*, membranous basidorsal; *my.*, myotome; *n.c.*, neural canal; *n.ch.*, notochord; *p.d.*, perichordal disc; *p.k.scl.t.*, loose tissue of caudal sclerotomite; *p.p.d.*, posterior part of perichordal disc; *rib*, rib; *sp.c.*, spinal cord; *sym.c.*, sympathetic nerve cord; *t.sp.g.*, thoracic spinal ganglion; *tub.*, tuberculum; *v.a.*, vertebral artery; *v.sp.n.*, ventral spinal nerve-root.

canal, but instead has well developed tubercular and capitular processes, although but a rudimentary rib.

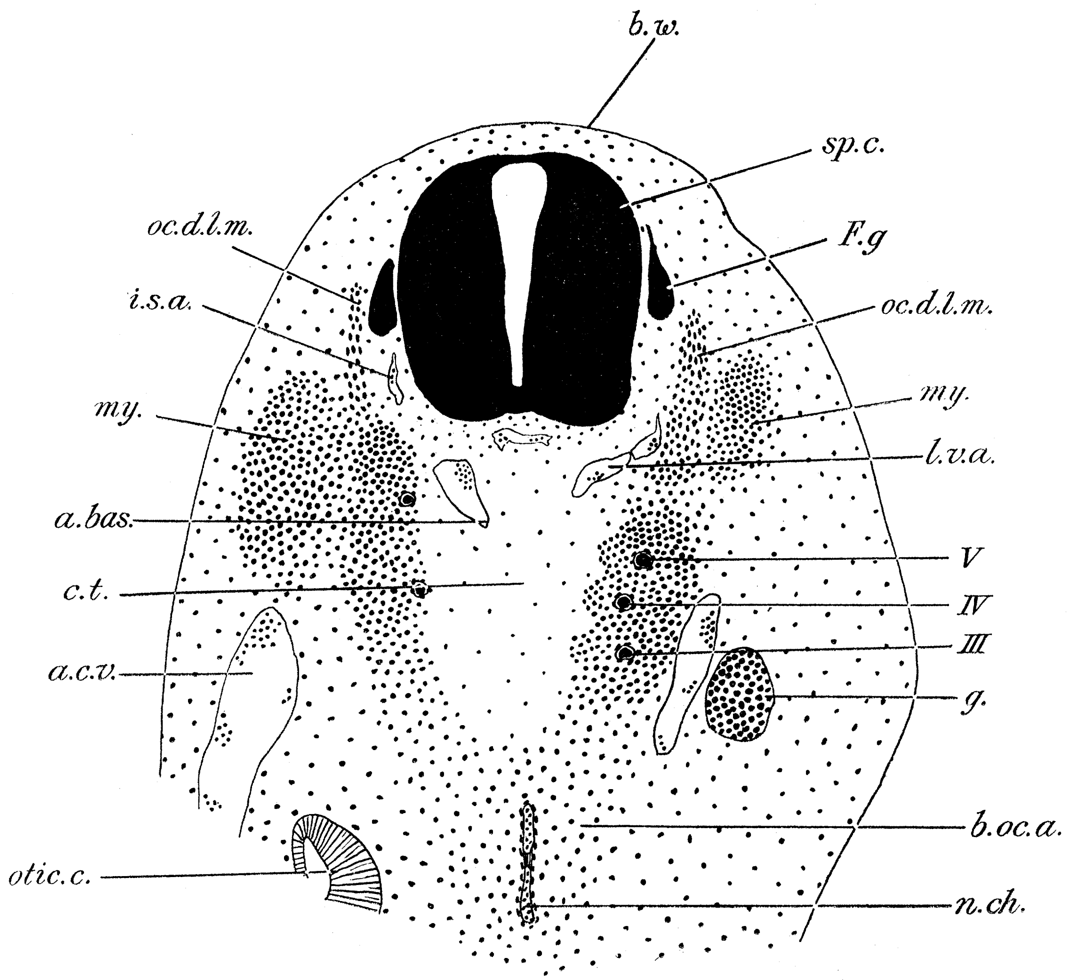
It must be noted also that the membranous basidorsals are not solely derived from the caudal sclerotomite, a few cells from the most posterior part of the cranial sclerotomite also contributing to each. These cells were noted in the previous stage as forming a slight condensation on the anterior border of the intrasclerotomic fissure.

There is no essential difference between the axis segment with its vertebral structures and the more posterior segments in the cervical region, but the atlas presents certain differences. The perichordal disc consists of fewer and less densely aggregated cells, while the subchordal part of the caudal sclerotomite is larger and possesses more cells. Since the differentiation of both atlas and axis segments is best studied in the next stage, a more complete description will be afforded when this stage is dealt with, but it might be mentioned here that there is no incipient dorsal-interdorsal corresponding to the 1st cervical segment.

It will be understood that there is a part of the 1st cervical segment in front of the corresponding perichordal disc, *i.e.*, the major part of the cranial sclerotomite of this segment. This region is distinctly separated from the occipital rudiment by the intersclerotomic fissure, which is persistent here. It is occupied by fairly loose tissue and represents the primordium of the pro-atlas. More will be said about this when the next stage is described.

The cervical flexure is now attaining its maximum development, and the hind-brain is inclined to the spinal cord at an angle of rather less than 90° (Text-fig. 4 (*e*)). The occipital segments thus lie vertically while the cervical ones lie horizontally, and the axis of greatest curvature passes between the two sets of segments. The atlas rudiment lying immediately posterior to this axis of curvature, is greatly modified in consequence, the dorsal part being drawn out so that the basidorsal is directed cranially, while the ventral part is somewhat crushed and displaced caudally. It cannot be too strongly emphasised, however, that the cervical region is distinctly separated from the occipital region.

Four occipital sclerotomes can still be recognised although the sole remaining criterion is the persistence of the 3 occipito-spinal nerves. The intersegmental blood vessels have dwindled to most significant proportions. The most posterior occipital sclerotome is best developed, consisting of a dense lateral part and a looser ventral part which occupies the region below the notochord. The ganglion corresponding to this segment (*i.e.*, FRORIEP's ganglion) persists, but is very small (Text-fig. 3, *F. g*). The 3rd and 4th occipital sclerotomes, which are the ones most crushed in the early stages of development of the cervical flexure, consist of looser tissue both laterally and ventrally and are the most ill-developed of all the occipital sclerotomes. The sclerotomal condensation corresponding to the 1st and 2nd occipital segments consists of dense tissue laterally, but equally loose tissue ventrally. In fact, throughout the whole occipital region, loose tissue occurs perichordally, while laterally it is much denser.

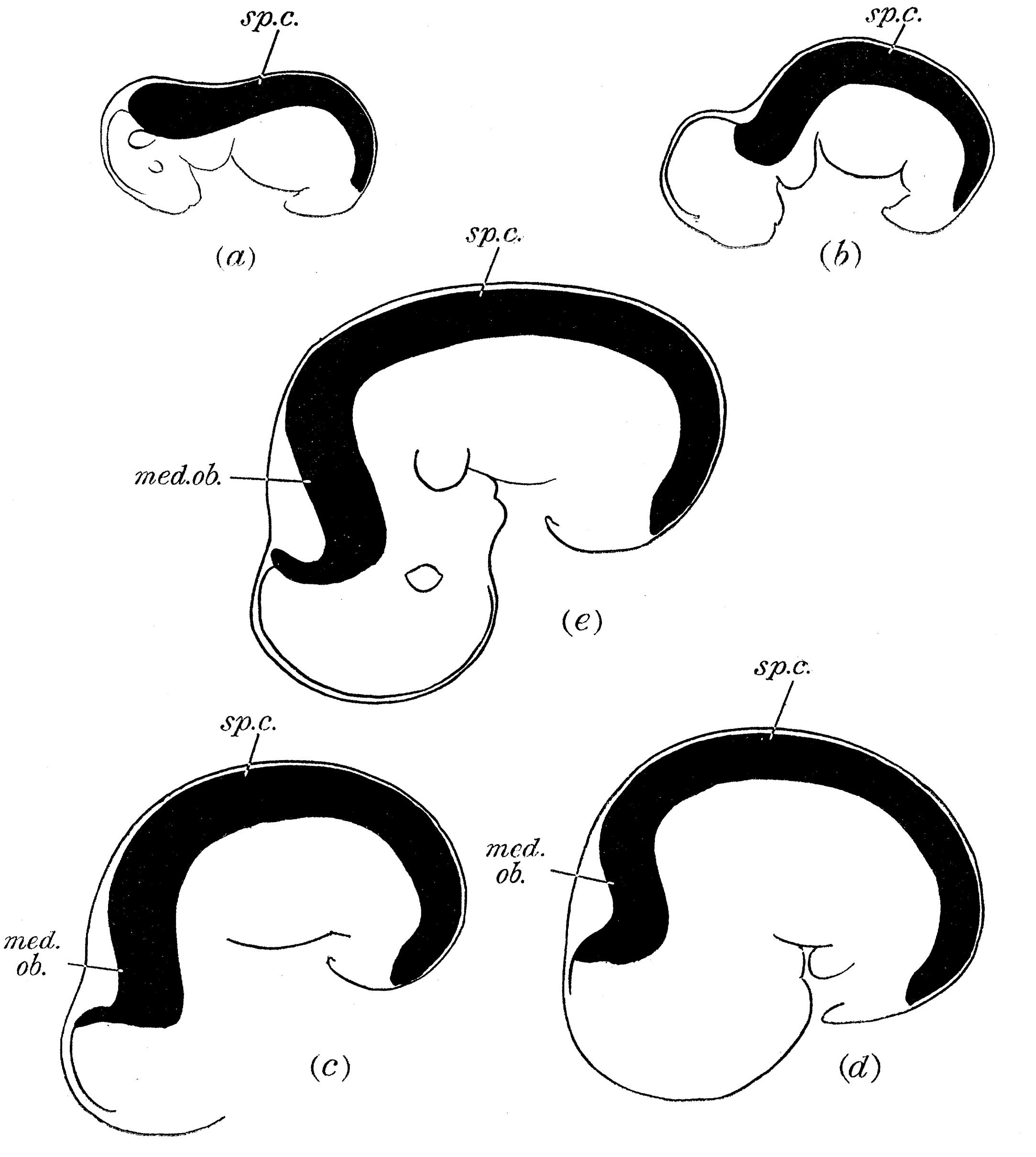


TEXT-FIG. 3.—A section, from a continuous transverse series, through the occipital region of a 6·5 mm. embryo. Because of the cervical flexure, the nerve cord is cut transversely, while the occipital region is cut half frontally. On the right side, the 3 occipito-spinal nerve-roots are seen. *a.bas.*, basilar artery; *a.c.v.*, anterior cardinal vein; *b.oc.a.*, basi-occipital rudiment; *b.w.*, body-wall; *c.t.*, connective tissue; *F.g.*, Froriep's ganglion; *g.*, ganglion; *i.s.a.*, intersegmental artery; *l.v.a.*, loop vertebral artery; *my.*, myotome; *n.ch.*, notochord; *oc.d.l.m.*, dorso-lateral mass of the occipital; *otic.c.*, otic capsule; *sp.c.*, spinal cord; iii, iv and v, occipito-spinal nerves corresponding to the 3rd, 4th and 5th occipital segments.

Embryo of Mus musculus, 7·2 *mm. C.R.Lth.*

The notochord is slightly larger than in the previous stage, its diameter is approximately 0·050 mm., and 10 or 11 nuclei are seen in a transverse section. There has been an enlargement of the vacuoles in the notochordal cells, but these are smaller than the nuclei of the cells in which they appear. Vacuoles are also more numerous at this stage. The notochordal sheath is thin, but conspicuous on account of its intense staining properties.

In the thoracic region, the basidorsal is still membranous, as is the incipient rib with its tubercular and capitular processes (Plate 12, fig. 9, *b.d.*, *rib*, *cap.*, *tub.*). The dorsal-interdorsal and interdorsal rudiments are also membranous, the latter now extending into the posterior part of the next anterior segment. The perichordal discs are slightly larger and more compact. There is no matrix between the densely crowded cells



TEXT-FIG. 4.—Diagrammatic representation of the development of the cervical flexure, based upon camera-lucida drawings of embryos made while these were still in the clearing agent. The general outline is indicated and the nerve-cord is shown in silhouette. The C.R.Lths. of the embryos are as follows :—(*a*) 3·0 mm. ; (*b*) 4·0 mm. ; (*c*) 4·8 mm. ; (*d*) 5·5 mm. ; (*e*) 6·5 mm. ; *med. ob.*, future medulla oblongata ; *sp.c.*, spinal cord. *Note.*—The embryos are not all drawn to the same scale.

(Plate 12, fig. 9, *p.d.*). Posteriorly, each disc is still connected with the diverging bases of the membranous basidorsals of its segments. The tissue of the basidorsal is equally dense throughout, a condition that does not obtain in the cervical region (*vide infra*).

Between the cells of the loose tissue near the anterior and posterior boundaries of the segments, a faint violet-staining matrix has been deposited in the form of a U-shaped mass underlying the notochord. This is the first sign of chondrification and represents the first rudiment of the centrum (Plate 12, fig. 10, *ctr.a.*). It lies between the bases of the membranous basidorsals and is derived from the ventral divisions of both cranial and caudal sclerotomites. It thus corresponds to the most posterior part of the basiventrals and the most anterior part of the interventrals of the next posterior segment.

In the cervical region, the rudiments of the basidorsals are more massive, and in the innermost part of each the nuclei are becoming loosened up and are losing their affinity for hæmatoxylin stains, appearing much less densely stained than the nuclei at the periphery. This loosening up of the central tissue and physiological differentiation of nuclei gives rise to the appearance of a pale patch within the tissue of the basidorsal, which is the first sign of onsetting chondrification in this structure. A faintly stained violet matrix is being deposited between the loosened cells. The basal part of each basidorsal is becoming similarly loosened up, so that the connection with the perichordal disc is being destroyed. The primordia of the cervical centra have the same mode of origin as the thoracic ones, but the crushing action of the cervical flexure has rendered less obvious the origin from portions of adjacent segments, which is readily seen, however, in the posterior part of this region. The primordia of the cervical ribs and their processes are still membranous. The processes form the lateral part of the vertebrarterial canal. A strand of tissue derived from the ventralmost part of the perichordal disc connects each pair of capitula below the most anterior part of the centrum rudiment (Plate 12, fig. 10, *s.s.c.*).

The 1st cervical segment is still clearly separated from the occipital segments by the well marked intersclerotomic fissure (Plate 12, fig. 11, *fis.*). It is also distinguishable from these segments by their advanced developmental condition. Loose tissue is still to be found in the anterior part of the 1st cervical segment below and on each side of the notochord, but above this structure there is a dense cell cluster of independent origin, which is the incipient interdorsal of this segment. This passes into the tissue of the 1st perichordal disc posteriorly. The whole of the tissue lying in front of the disc forms the rudiment of the pro-atlas (Plate 12, fig. 11, *pr.at.*). The tissue of the 1st perichordal disc is looser than that of more posteriorly situated ones and extends throughout the posterior part of its segment, being flanked by the bases of the basidorsals, which are especially massive in their basal parts (Plate 12, fig. 11, *b.b.d.*). The tubercular homologues are slender and pass into the tissue of the basidorsals, while the capitula are greatly enlarged and form the major part of the ventral ring of the atlas. Only a small nodule of tissue, which connects the pair of capitula subchordally, is derived from the perichordal disc of this segment.

The 2nd cervical segment is at this stage an almost typical segment. There is some indication of a bilateral origin of the atlas centrum, which lies partly in the anterior portion of this segment. The 2nd perichordal disc is quite typical, representing the most posterior part of the interventrals fused with the most anterior part of the basiventrals.

The dorso-lateral part of the occipital sclerotomes, which are completely fused, consists of fairly dense tissue with little intercellular matrix. It is generally loosened up and not merely loose in its central part as is the case with the basidorsals of the cervical and thoracic regions. The subchordal and ventro-lateral parts consist of definite pro-cartilaginous tissue. Tiny capsules are being formed throughout. Thus in the occipital region, the notochord lies in a groove above the pro-cartilage which represents the fused occipital centra. It is important to realise that this structure is in an advanced histological condition and clearly distinct from the 1st cervical segment. It is the rudiment of the basi-occipital cartilage (Plate 12, fig. 11, *b.oc.a.*). Mesenchymatous tissue forms a roof over this trough-like rudiment, thus enclosing the notochord. The ventro-lateral portion of the fused occipital sclerotomes consists of fairly loose tissue devoid of intercellular matrix, and it is through this mass that the three main roots of the hypoglossal nerve pass—separately. There is an almost imperceptible gradation into the tissue of the dorso-lateral mass and as yet the condylar foramina are not developed.

Embryo of Mus musculus, 7·8 *mm. C.R.Lth.*

The notochord, although but slightly altered, now affords indications of the onset of profound changes. The number and size of the vacuoles has increased and the nuclei are centrally situated. A complete ring of cytoplasm is closely adherent to the inside of the sheath and is thus separated from the nucleated part of the notochord. The structure has become segmentally differentiated, being constricted intersegmentally and correspondingly dilated mid-segmentally. At present, the respective diameters in these situations are 0·050 mm. and 0·065 mm. The primordia of the centra contain the constrictions. The perichordal tube is conspicuous in the intersegmental position, consisting of a layer of flattened cells 2 or 3 nuclei thick, closely apposed to the notochordal sheath. There is no intercellular matrix between the flattened cells and so the formation contrasts strongly with the developing centrum.

In the thoracic region, the central tissue of the basidorsals has now become loosened up and a matrix is being deposited between the cells, exactly as happened in the cervical region in the previous stage. The tissue of the ribs is becoming looser in the same way, the pale patch extending ventrally as far as the horizontal level of the aorta. The cells which are being crowded to the surface in both basidorsal and rib, form a dense mass, which is the rudiment of the perichondrium. The tissue of the perichordal discs is much more dense than in the previous stage. The anterior part of each of these structures, on to which the capitula abut, is equally distributed ventral and dorsal to the notochord. The ventralmost part is differentiated into a strand of cells with elongated nuclei, which strand subchordally connects a pair of capitula. The centra are more definitely pro-cartilaginous, but the interdorsal remains unchondrified. The centra are encroaching slightly upon the posterior parts of the perichordal discs.

In the cervical region, the process of loosening up of the tissue of the basidorsals and the deposition of matrix between the cells has proceeded much further than in the thoracic region. But the incipient cervical ribs with their processes, which together form the vertebrarterial canal, still consist of dense membranous tissue. The perichordal discs are smaller than the thoracic ones although larger than in the previous stage. The most anterior part of each is confined to the dorsal aspect of the notochord, where it abuts on to the membranous fused interdorsals. The rudimentary capitula of the cervical ribs pass into the disc on the posterior aspect, and not on the lateral one as is the case in the thoracic region. The subchordal strand of tissue connecting the capitula forms a shelf underlying the most anterior part of the centrum rudiment. The dorsal-interdorsals are membranous, like those of the thoracic region.

The 1st cervical perichordal disc occupies the whole of the posterior half of its segment, is still of loose consistency and is still connected with the atlas basidorsals laterally. The capitular homologue is greatly enlarged, while the tubercular one, which forms the lateral part of the vertebrarterial canal, consists of a thin strand of dense tissue.

The basi-occipital rudiment consists of the most developmentally advanced tissue in the whole vertebral column. Cartilage capsules have been developed and the matrix is more copiously deposited and more deeply staining than in any other region. There is a slight deposition of matrix in the loose tissue above the notochord, but this does not persist into the next stage.

Embryo of Mus musculus, 8·0 *mm. C.R.Lth.*

The differentiation of the notochord is slightly more marked than in the previous stage. The diameters of the constricted and dilated segmental regions are approximately 0·040 mm. and 0·060 mm. (Plate 12, fig. 12, *n.ch.*) The sheath is uniformly slightly thicker but the degree of vacuolation is unchanged. In spite of the slight dilatations and constrictions, the segmental waves of the notochord can still be perceived, the ventral troughs being mid-segmental as before. The perichordal tube is still conspicuous and there has been a slight deposition of intercellular matrix between the cells composing it (Plate 12, fig. 12, *p.t.*).

In both cervical and thoracic regions, the pro-cartilaginous basidorsals are well developed, although the proximal and distal portions are still unchondrified. The basal part of each, which is still in connection with the perichordal disc, is becoming loosened up and amalgamated with the antero-dorsal portion of the centrum rudiment, so that the density of nuclei in this part is now greater than was formerly the case (Plate 12, fig. 12, *b.b.d.*). The centra consist of fairly loose tissue however, but with a copious intercellular matrix. The thoracic centra are more extensive antero-posteriorly than the cervical ones, the interbasalia playing a greater part in their formation. The interdorsals and the dorsal-interdorsals remain membranous.

The thoracic ribs extend much further ventrally, the distal portions having grown out along the intersegmental septa, the chondrified part advancing correspondingly. The capitular and tubercular processes are becoming pro-cartilaginous, but retain a strictly membranous connection with the anterior portions of the perichordal discs and basidorsals respectively. The 11th rib has a very rudimentary tuberculum, while the 12th and 13th ribs possess only capitular processes, which abut on the anterior parts of the corresponding perichordal discs. Thoracic and cervical discs alike are entirely membranous. The tissue of the cervical ribs and the vertebrarterial canal remains unchondrified. The 7th ribs retain both capitular and tubercular processes, which are similarly membranous.

The perichordal disc of the 1st cervical segment consists of rather more dense tissue than formerly (Plate 13, fig. 13, *p.d.at.*), and is still in membranous connection with the bases of the basidorsals, which unlike more posterior ones show no signs of loosening up. These are now co-extensive with the now laterally extended membranous fused interdorsals of the 2nd cervical segment, which form the primordium of the articular facet on the axis for the atlas ring and also the atlanto-axial ligament. The pro-atlas, which is still unchondrified, overlies the most posterior part of the occipital rudiment and is even more distinct and separate from it than was formerly the case (Plate 13, fig. 13, *pr.at.*, *b.oc.a.*).

The capitular component of the vertebrarterial canal is extremely well developed. It is in membranous connection with the basidorsal on the mesial aspect of the vertebral artery and shows a similar connection with the perichordal disc. This component forms the larger part of the ventral ring of the atlas, which is evident at this stage, although in but a rudimentary condition. The tubercular component is relatively very slight and forms a slender strand of tissue enclosing the vertebral artery laterally.

The 2nd cervical perichordal disc is similar to more posterior ones, but between its component cells a small quantity of intercellular matrix has been deposited (Plate 13, fig. 13, *p.d.ax.*). The interventral component of the centrum corresponding to this segment, which lies immediately in front of the disc, is much wider than in the typical cervical segment and underlies the equally wide membranous interdorsal mentioned above.

The 1st cervical spinal ganglia are smaller than in previous stages, but the corresponding ventral nerve roots are well developed. The ganglion of Froriep is still to be found lying on the spinal accessory (Cranial xi) nerve, but only in a vestigial condition.

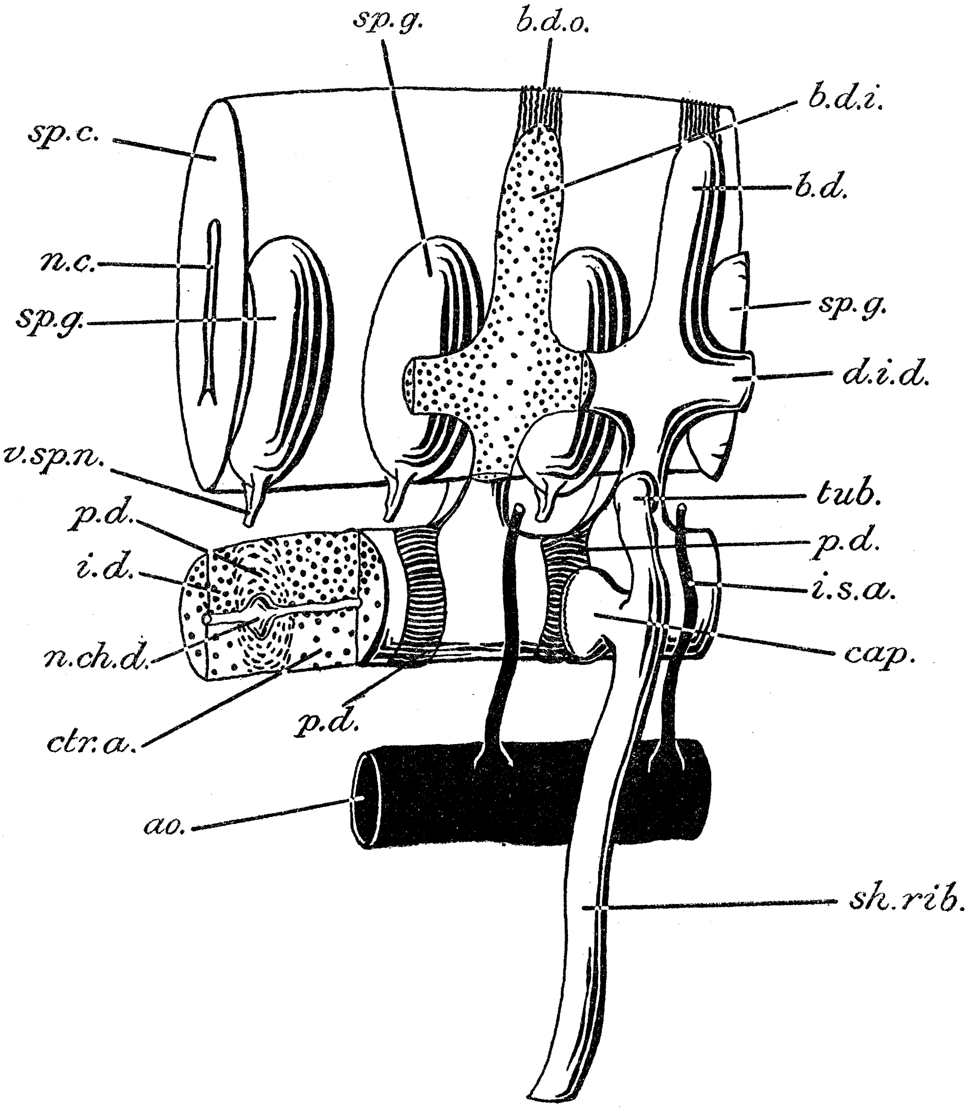
In the occipital region, the cartilaginous basi-occipital rudiment is still separated from the pro-cartilaginous dorso-lateral mass by the dense membranous ventro-lateral mass. It is through this last that the components of the hypoglossal nerve pass. There is no definite foramen in this stage.

Embryo of Mus musculus, *9·0 mm. C.R.Lth.*

The regional differentiation of the notochord has proceeded considerably. In the cervical region, the diameter intersegmentally, and therefore within the centrum, is 0·025–0·030 mm., only 2 or 3 nuclei being present in a single section. The mid-segmental diameter within the intercentrum is 0·095–0·100 mm., about 16 nuclei being shown in section. In the thoracic region, the dilatation of the notochord is greater, the mid-segmental diameter being about 0·15 mm. The notochordal sheath is thick and conspicuous within the centrum rudiment, but thin and attenuated in the region of the perichordal disc. The notochordal cytoplasm has lost its cellular appearance ; it now shows up as a mass of brownish nuclei and of vacuoles, set in a cytoplasmic ground medium in which there are no cell boundaries. This syncytial nature persists and characterises the notochordal mass until development of the vertebral column is almost complete.

In the thoracic region, the basidorsals extend dorsally almost to the upper limit of the nerve cord, each pair being united above this structure by a thin strand of fibrous connective tissue, so that a completed but rudimentary neural arch is formed (Text-fig. 5, *b.d.*). Each basidorsal consists of very advanced pro-cartilage with characteristic angular or irregularly oval nuclei (Plate 13, fig. 14, *b.d.*). The cells earlier displaced towards the periphery have given rise to a definite perichondrial layer which stains an intense blue colour. The cells of the basal portion of each basidorsal, which at an earlier stage became loosened and fused with the anterior part of the centrum rudiment, have now become chondrified, so that the perichondrium of the posterior margin of the basidorsals is continuous with that on the dorsal surface of the rudimentary centrum. The perichondrial layer on the anterior aspect of each basidorsal is still in perfect continuity with the postero-dorsal part of the perichordal disc (Text-fig. 5, *p.d.*). Thus the basidorsal contributes to the centrum anteriorly. The basidorsals have been extended posteriorly as a result of the loosening of their cells consequent upon chondrification, and there is a well defined notch in the posterior margin of each, through which the ventral spinal nerve of the next posterior segment passes. This is the intervertebral notch rudiment.

The interdorsal has become chondrified and completely fused with the interventral component of the developing centrum lying immediately below it. It has also fused with the basidorsals of the next anterior segment, which in turn have fused with the basiventrals of the same segment. Thus the centrum rudiment at this stage is built up of the basiventral and basidorsal components of one segment together with the interventral and interdorsal components of the next posterior segment. The dorsal-interdorsals are still membranous and continuous with the perichondrial layer of the basidorsals. The thoracic ribs have extended below the level of the heart ventrally, and the ten most anterior ones are connected ventro-laterally on each side by a membranous structure which is the bilateral rudiment of the sternum. Chondrification is almost complete in the rib, a definite perichondrium having been formed. The capitular and tubercular processes consist of pro-cartilage less advanced developmentally than the tissue of the rib proper. The capitulum is still in membranous connection with the anterior part of the corresponding perichordal disc, while the tuberculum has a similar connection with the perichondrium of the basidorsal. The capitulum lies in



TEXT-FIG. 5.—A stereogram based upon a graphic reconstruction from transverse serial sections of the vertebral and other structures in the thoracic region of a 9·0 mm. embryo. The structures are shown in surface view, except where supposedly cut to show internal structure. One basidorsal is cut down longitudinally and the axial part of the vertebral column is cut in the sagittal plane, the left side having been removed. The cell-condensations are indicated by variety in stippling. The view is from the left side and slightly in front. *ao.*, aorta; *b.d.*, basidorsal; *b.d.i.*, loose tissue of inner part of basidorsal; *b.d.o.*, dense tissue of outer part of basidorsal; *cap.*, capitulum; *ctr.a.*, centrum rudiment; *d.i.d.*, dorsal-interdorsal; *i.d.*, interdorsal; *i.s.a.*, intersegmental artery; *n.c.*, neural canal; *n.ch.d.*, notochordal dilatation; *p.d.*, perichordal disc; *sh.rib*, shaft of rib; *sp.c.*, spinal cord; *sp.g.*, spinal ganglion; *tub.*, tuberculum; *v.sp.n.*, ventral spinal nerve.

front of the tuberculum, just as the perichordal disc lies in front of the basidorsals (Text-fig. 5).

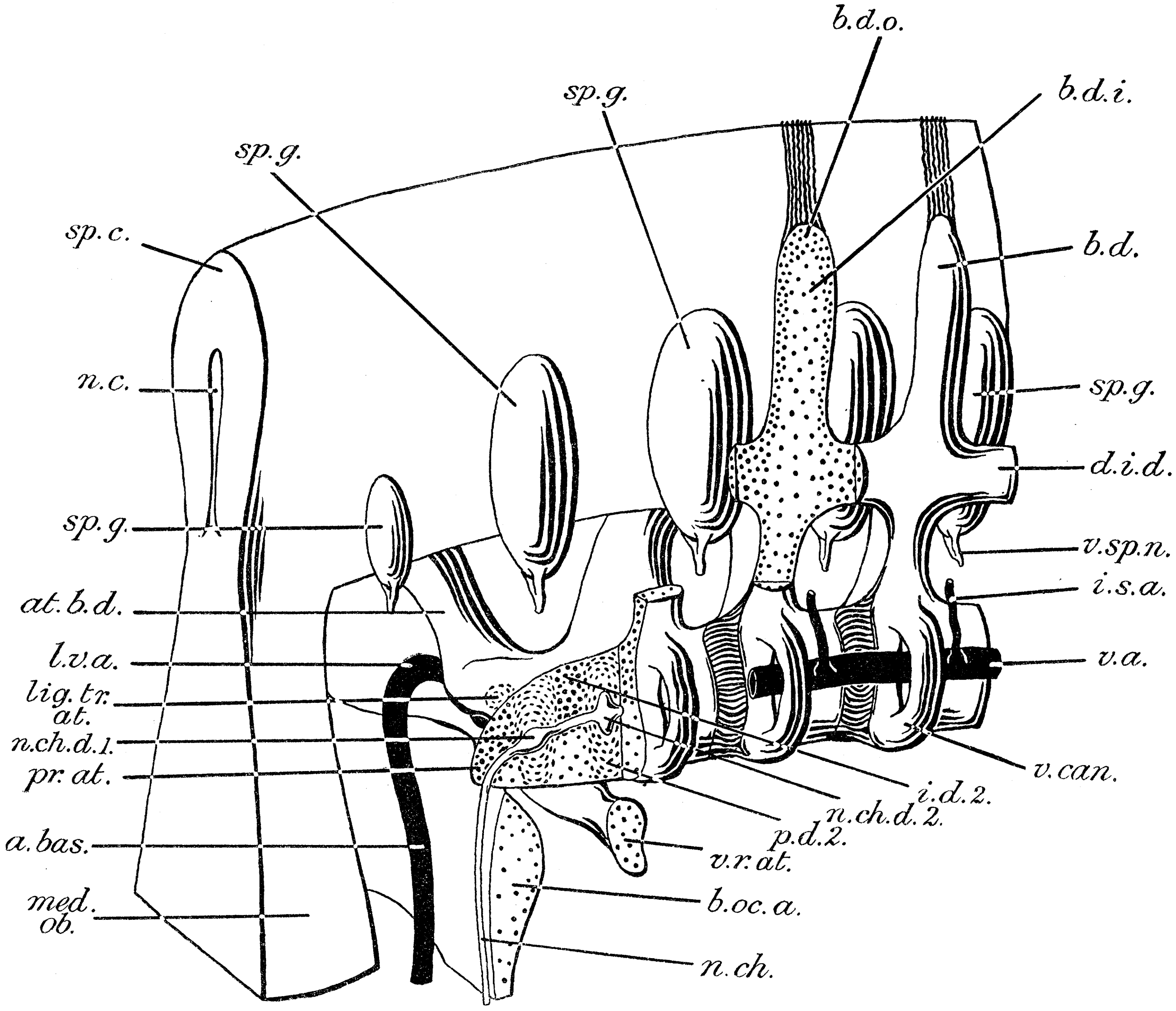
In the cervical region, the basidorsals extend slightly further dorsally than in the thoracic region, and are similarly united with their fellows of the opposite side above the nerve cord. The perichordal discs are largely densely membranous, but in the anterior parts the subchordal tissue is slightly chondrified (Plate 13, fig. 16, *p.d.*). The mid-ventral portion of each forms a strand of dense fibrous connective tissue uniting each pair of capitula. The capitular and tubercular homologues, which form the vertebrarterial canal, consists of dense pro-cartilage. The developing centrum has the same composition in this region as in the thoracic, but the interventral and interdorsal components are not as large.

In both cervical and thoracic regions, the perichordal tube has undergone change. Whereas in the previous stage it formed an investment round the notochord which was conspicuous, in the present stage it is not recognisable as such. The cells of which it was composed have become scattered and merged with the developing centrum, the intercellular matrix of which is now continued up to the notochordal sheath. In this way the developing centrum is cemented to the notochordal sheath. The dorsal-interdorsals of the cervical region, like those of the thoracic region, are unchondrified (Plate 13, fig. 14, *d.i.d.*).

The perichordal disc of the 1st cervical segment still occupies the whole of the posterior half of its segment and it is interesting to find a notochordal dilatation within it (Text-fig. 6, *n.ch.d.*1). The dorsal portion of the disc is slightly chondrified, but the ventral part is membranous. It is still in fibrous-tissue connection with the bases of the atlas basidorsals, which are fused with the greatly enlarged capitular homologues of the corresponding segment. These latter give rise to the ventral ring of the atlas, and it might be mentioned here that this is effected by the maintenance of a fibrous tissue connection between the bases of the basidorsals and the axial portion of the developing vertebral column. The tubercular homologue is still thin and membranous.

The pro-atlas now consists of a U-shaped and chondrified ventral portion, which is the interventral component in front of the 1st perichordal disc, and a dense membranous mass dorsal to the notochord, which represents the fused interdorsal components (Plate 13, fig. 15, *i.v.pr.*, *i.d.pr.*). It lies immediately in front of the 1st perichordal disc and the fibrous tissue which connects this structure with the bases of the atlas basidorsals. It is from this fibrous tissue that the lig. transversum atlantis becomes differentiated. At this stage, it appears as a dense mass attached to the bases of the basidorsals and looping over the posterior part of the pro-atlas (Plate 13, fig. 15, *lig.tr.at.*). The remaining tissue connecting the 1st perichordal disc and the bases of the atlas basidorsals is loosely arranged (Plate 13, fig. 15). It is in this loose tissue that the synovial cavities between the ventral part of the atlas ring and the odontoid process, and this latter process and the lig. transversum atlantis respectively, will be developed subsequently.

In the 2nd cervical segment, the interdorsal, which it will be remembered is very



TEXT-FIG. 6.—A stereogram based upon a graphic reconstruction from serial transverse sections of the vertebral and other structures in the anterior cervical region of a 9·0 mm. embryo. As in Text-fig. 5, the nerve cord, spinal ganglia, and vertebral components are shown in surface view, except where supposedly cut to show internal structure. The left halves of the atlas and occipital rudiments have been removed and the axis basidorsal is cut off short. One basidorsal is cut down longitudinally and the axial part of the vertebral column is cut in the sagittal plane anteriorly, the left side having been removed. The cell-condensations are indicated by variety in stippling. The vertebral artery is cut away on the left side anteriorly, and the view is from this side, and slightly in front. *a.bas.*, basilar artery; *at.b.d.*, atlas basidorsal; *b.d.*, basidorsal; *b.d.i.*, loose inner tissue of basidorsal; *b.d.o.*, dense outer tissue of basidorsal; *b.oc.a.*, basi-occipital rudiment; *d.i.d.*, dorsal-interdorsal; *i.d.*2, interdorsal of second cervical segment; *i.s.a.*, intersegmental artery; *l.v.a.*, loop of vertebral artery; *lig.tr.at.*, lig. transversum atlantis; *med.ob.*, medulla oblongata; *n.c.*, neural canal; *n.ch.*, notochord; *n.ch.d.*1, notochordal dilatation of first cervical segment; *n.ch.*2, notochordal dilatation of second segment; *p.d.*2, perichordal disc of second segment; *pr.at.*, pro-atlas; *sp.c.*, spinal cord; *sp.g.*, spinal ganglion; *v.a.*, vertebral artery; *v.can.*, vertebrarterial canal; *v.r.at.*, ventral ring of atlas; *v.sp.n.*, ventral spinal nerve.

extensive laterally and continuous with the basidorsal posteriorly, is still membranous. The interventral mass immediately below it is completely chondrified and merged with the basal parts of the basidorsals of this segment. It is seen to be in chondrogenic connection with the rudiment of the ventral ring of the atlas also. Later, this connection is lost, but for a brief space of time the typical condition obtains in this modified region of the vertebral column (Plate 13, fig. 16, *cart.*). The perichordal disc of this segment is completely chondrified and continued into the corresponding basidorsal, which is also fused with the true axis centrum. This last lies behind the 2nd cervical disc, of course, being derived partly from the posterior portion of the 2nd segment and partly from the anterior portion of the 3rd. The interdorsal component is only incompletely chondrified.

It must be observed here that loose skeletogenous tissue occurs around the axial part of the developing vertebral column. It is the residuum of the skeletogenous layer and is termed the "membrana reuniens." From this tissue, and in the region below the axial vertebral structures, the primordia of the anterior or ventral common ligament, the anterior atlanto-axial ligament and the anterior atlanto-occipital ligament are formed at this stage. The posterior or dorsal common ligament is also formed in the dorsal portion of this tissue.

The basi-occipital rudiment consists of advanced cartilage, especially in the subchordal part. The three main roots of the hypoglossal nerve still pass separately through the tissue of the ventro-lateral mass, but the tissue between these roots is disappearing, to give rise to some slight indication of the developing condylar foramena (Plate 13, fig. 14, *oc.*, *h.* xii). These lie in the region corresponding to the 3rd and 4th occipital sclerotomes. Between the occipital rudiment and the pro-atlas is a region of loose tissue through which the notochord passes, encased in dense fibrous tissue. This is the rudiment of the lig. apicis dentis, or middle odontoid ligament, to which therefore the notochord contributes. The membrana tectoria is formed in this region also, from the dorsal portion of the "membrana reuniens" (Plate 13, fig. 16, *m.t.*).

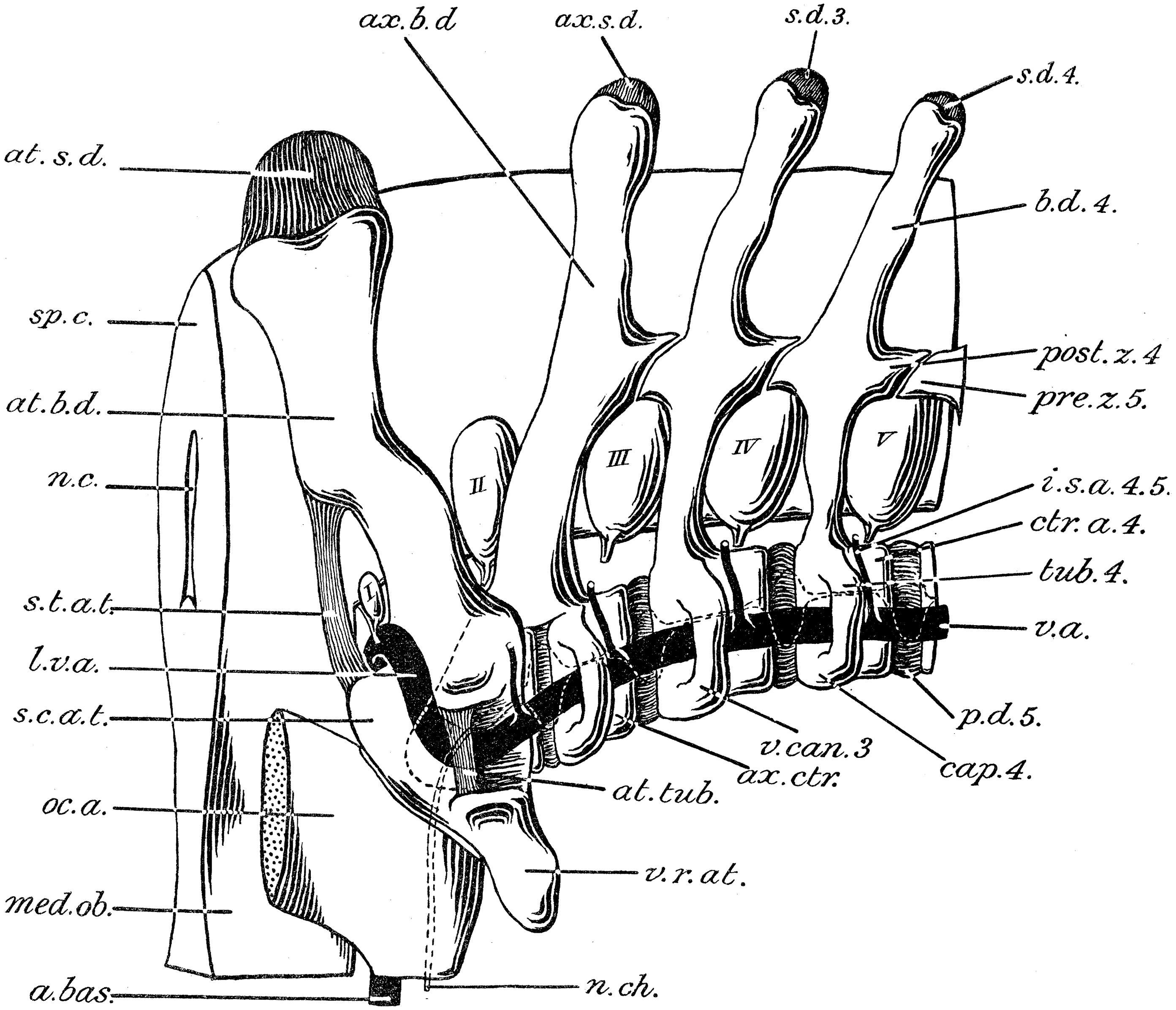
Embryo of Mus musculus, 12·5 *mm. C.R.Lth.*

The notochord is no longer a complete rod of cells. In the intersegmental position and within the developing centrum the sheath alone is seen, the syncytial mass having been completely squeezed out into the region of the perichordal disc (Plate 13, fig. 18, *n.ch.*). The sheath here is laterally compressed, its axes measuring 0·040 mm. and 0·025 mm. respectively. In the middle of the disc, the diameter of the notochordal mass is about 0·320 mm. and about 80 nuclei are seen in section. The dilatation extends for about 0·09 mm. antero-posteriorly, so that the nuclei of the notochordal syncytium must have been actively engaged in dividing since the previous stage. It is obvious that there has been no real degeneration of this tissue, but, on the contrary, the cytomorphosis of these cells has been accompanied by an increased activity of their nuclei. Vacuoles are rather more numerous and the sheath appears extremely thin but quite complete in the dilated part of the notochord.

The basidorsals of both cervical and thoracic regions now extend slightly above the upper limit of the nerve cord (Text-fig. 1, *b.d.* 4) and consist of advanced cartilage with large cells. They are connected by dense fibrous tissue which passes over the nerve cord. A few cells have come to lie on this fibrous tissue, thus giving rise to the primordium of a median dorsal formation, histologically different from the basidorsals and later in time. As it completes the neural arch, this formation must correspond to the Supra-dorsal (GADOW). The dorsal-interdorsals are largely membranous, and an oblique cleft has appeared in each, which is the first indication of the interneural arthrodial diarthrosis. The portions of each interdorsal which are proximal to the posterior aspect of one basidorsal and the anterior aspect of the next posterior one respectively, have become converted into early embryonic cartilage with angular nuclei and very little intercellular matrix (Plate 13, fig. 17, *d.i.d.* 3). These are the rudiments of future pre- and post-zygapophyses respectively (Text-fig. 7, *post.z.* 4, *pre.z.* 5).

The cervical and thoracic perichordal discs have become differentiated each into 3 zones, an anterior, a middle, and a posterior one (Plate 13, fig. 18, *p.d.a.z.*, *p.d.p.z.*, *lig.i.v.*). The anterior zones have become chondrified and have fused with the most posterior part of a developing centrum and the most anterior part of the next posterior one respectively. The two zones are readily distinguishable from the tissue of the earlier centrum rudiment, which consists of much more advanced tissue, especially in the core (Plate 13, fig. 18, *ctr.a.*). The middle zone consists of a narrow ring of fibrous tissue stretched between the rudiments of consecutive definitive centra (each definitive centrum = the posterior zone of the disc, plus the earlier centrum rudiment, plus the anterior zone of the disc next posterior) and extending around the notochordal dilatation. Some slight intercellular matrix has been deposited, but it cannot be regarded as a chondrified structure. It is the rudiment of the fibro-cartilagines intervertebrales, or definitive intervertebral disc (Plate 13, fig. 18, *lig.i.v.*).

The thoracic ribs consist of very advanced cartilage which extends into the capitular and tubercular processes proximally. The most mesial part of each capitulum consists of dense early cartilage, which abuts on to the postero-lateral part of the definitive centrum of the vertebra next anterior and partly on to the definitive intervertebral disc. The costo-central diarthrosis consists of a very dense fibrous tissue (Plate 13, fig. 18). The tip of the tuberculum likewise consists of dense cartilage and the costo-transverse diarthrosis of dense fibrous tissue. The diapophysis is indicated at this stage as a small pro-cartilaginous peg-like process on the basidorsal. The mesial strand of tissue connecting each pair of capitular processes subchordally in the previous stage, is now chondrified and is added partly to the anterior zone of the perichordal disc and partly to the tissue of the intervertebral disc. Ossification centres have appeared in the angles of the ribs. These are best developed at this stage in the 5th pair of ribs, having a cross-sectional area approximately equal to that of the centrum. The



Text-fig. 7.—A stereogram based upon a graphic reconstruction of the vertebral and other structures in the anterior cervical region of a 12·5 mm. embryo. The lateral mass of the occipital is supposedly cut and the distal half removed. The view is from the left side and slightly to the front. The course of the notochord is difficult to show, but an attempt has been made to indicate it by dotted lines. The tip of the odontoid process, which lies within the atlas ring, is similarly shown. *a.bas.*, basilar artery; *at.b.d.*, atlas basidorsal; *at.s.d.*, atlas supra-dorsal; *at.tub.*, tubercular homologue of atlas; *ax.b.d.*, axis basidorsal; *ax.ctr.*, axis centrum; *ax.s.d.*, axis supra-dorsal; *b.d.*4, basidorsal of fourth cervical segment; *cap.*4, capitulum homologue of fourth vertebra; *ctr.a.*, centrum of fourth vertebra; *i.s.a.*,4, 5, intersegmental artery between fourth and fifth segments; *l.v.a.*, loop of vertebral artery; *med. ob.*, medulla oblongata; *n.c.*, neural canal; *n.ch.*, notochord; *oc.a.*, occipital; *p.d.*5, fifth perichordal disc; *post.z.*4, post-zygapophysis of fourth cervical vertebra, *pre.z.*5, prezygapophysis of fifth cervical vertebra; *s.c.a.t.*, sub-condylar portion of atlas; *s.d.*3, third supra-dorsal; *s.d.*4, fourth supra-dorsal; *sp.c.*, spinal cord; *s.t.a.t.*, strand of fibrous tissue enclosing loop of vertebral artery; *tub.*4, tubercular homologue of fourth cervical vertebra; *v.a.*, vertebral artery; *v.can.*3, vertebrarterial canal of third cervical vertebra; *v.r.at.*, ventral ring of atlas (ventralmost part); i, ii, iii, iv and v, first to fifth cervical spinal ganglia.

centres in the 2nd, 3rd, 4th and 6th pairs are smaller, while in the 1st, 7th, 8th and 9th pairs, the perichondrium is breaking down as an indication of onsetting chondrification. In the four most posterior ribs the perichondrium is complete, but blood vessels are arranged around it as if in readiness for the process which will follow its breaking down.

The cervical ribs consist of dense early cartilage, as does the tissue of the vertebrarterial canal (Plate 13, fig. 17). The cores of the cervical centra consist of smaller cells than are seen in the cores of the thoracic centra.

The tuberculum homologue of the atlas is still unchondrified, so that the lateral portion of the vertebrarterial canal consists of fibrous tissue (Text-fig. 7, *at.tub.*). The capitular homologues form the large and well-developed ventral portion of the atlas ring (Text-fig. 7, *v.r.at.*). The tissue on the mesial aspect of the vertebral artery and near its junction with the basilar artery, which is the tissue resulting from a fusion of the basidorsals with the enlarged capitular homologues, is now seen to be growing forwards towards the ventro-lateral portion of the occipital rudiment (Text-fig. 7, *s.c.a.t.*). Here, together with the above portion of the occipital, it forms the rudiment of the double-condyloid joint on each side. And from this tissue, a strand of fibrous connective tissue extends to the antero-lateral part of the atlas basidorsal, passing in front of the ventrally directed loop of the vertebral artery, which is thus enclosed in a rudimentary foramen (Plate 14, fig. 19, and Text-fig. 7, *s.t.a.t.*). The 1st cervical spinal nerve also passes through this foramen, which persists even in the adult animal.

The interdorsal portion of the pro-atlas is now completely chondrified, as is also the 1st perichordal disc, the tissue of which has become merged into that of the pro-atlas anteriorly and that of the atlas centrum rudiment posteriorly. A slight notochordal dilatation is still to be seen within it (Plate 14, fig. 20, *n.ch.d.* 1). The 2nd perichordal disc is also completely chondrified and merged into the general perichordal mass, which is the rudiment of the odontoid process. The 3rd cervical perichordal disc marks the posterior limit of the axis body, its anterior zone being added to the posterior extremity of it (Plate 14, fig. 20, *lig.i.v.* 2, 3). Thus the odontoid process and the body of the axis develop from the axial sclerotomic derivatives of the first two segments plus half of the third.

While the mesial subchordal portion of the occipital rudiment consists of advanced cartilage (Plate 14, fig. 20, *b.oc.a.*), the ventro-lateral and dorso-lateral portions consist of early embryonic cartilage, the latter being rather more advanced than the former. The condylar foramina appear well developed. The rudiments of the condyles have been developed on the posterior aspect of the ventro-lateral portion of the occipital mass. They occupy a primitive ventral position behind and below the floor of the hind-brain.

Embryo of Mus musculus, 14·0 *mm. C.R.Lth.*

The diameter of the notochordal mass in the region of the intervertebral disc has increased to 0·38 mm. (Plate 14, fig. 22, *n.ch.d.* 3) and although the sheath cannot clearly be seen, yet the syncytium is quite distinct from the fibrous tissue of the disc. The sheath can still be seen in the middle of each centrum, through which it pursues an unbroken course.

In the thoracic region, ossification centres are appearing in the basidorsals. These are best marked at this stage in the 9th, 10th and 11th pairs, where they are fairly small however. The 12th and 13th pairs show still smaller centres, while in the 2nd and 8th pairs, the perichondrium is in the act of breaking down. The 1st, 3rd, 4th, 5th, 6th and 7th pairs do not show any sign of onsetting ossification. The supradorsals consist of embryonic cartilage cells loosely arranged on a fibrous tissue basis. All the thoracic ribs are considerably ossified, the centres extending from the bases of the tubercula and capitula through half the length of the rib, the angle of each of which is completely evacuated of cartilage (Plate 14, fig. 21, *b.c.r.*). The tubercula consist of fairly dense cartilage which passes into dense fibrous tissue at the costo-transverse joint, and the capitula have a similar composition (Plate 14, fig. 21, *tub.*, *cap.*).

All the cervical basidorsals except the 7th possess well defined ossification centres just above the level of the vertebrarterial canals and below the zygapophyses. Those in the atlas basidorsal are largest, extending half way to the dorsal boundary of the spinal cord. The pre- and post-zygapophyses consist of dense cartilage and the supradorsals, which are widest in the case of the atlas and axis, are also composed of similar cartilage. The ribs of the 7th cervical vertebra are well developed and possess ossification centres. The vertebrarterial canals of all the cervical vertebræ are densely cartilaginous, with the exception of the lateral part of that of the atlas, which is still fibrous. The tissue forming the anterior margin of the secondarily derived foramen in the atlas arch for the vertebral artery and the 1st cervical spinal nerve is now chondrified. The pro-atlas still consists of dense cartilage.

The cores of cervical and thoracic centra alike are becoming characterised by very large cells, and the anterior and posterior zones of former perichordal discs are densely cartilaginous, so that a striking contrast is afforded between the ends of future vertebræ and the middle parts. There has been a slight deposition of matrix between the cells of the intervertebral discs, but these are certainly not " chondrified."

The mesial, subchordal and dorso-lateral portions of the occipital rudiment consist of very advanced cartilage. A median ossification centre has appeared in the subchordal part of the former, while in the latter a single centre occurs on each side. The ventro-lateral mass still consists of dense cartilage, in which the condylar foramina of each side are situated. The condylar portion of the ventro-lateral mass still occupies its primitive ventral position below the level of the nerve cord and behind the hind-brain.

Embryo of Mus musculus, 23·0 *mm. C.R.Lth.* (*the newly-born animal*).

The notochordal mass within the intervertebral disc has once more become cellular. It is readily distinguished from the tissue of the disc by its feebly stained cytoplasm (Plate 14, fig. 24, *n.pulp.*), and forms the basis of the nucleus pulposus of the adult.

The tissue of the dorsal part of the disc is more dense and possesses a more copious matrix than the dorsal part (Plate 14, fig. 24, *lig.i.v.*).

The portion of the neural arch corresponding to the basidorsals is completely evacuated of cartilage and entirely bony in both cervical and thoracic regions (Plate 14, fig. 23, *b.c.n.a.*). The bone masses extend slightly into the bases of the pre- and post-zygapophyses (Plate 14, fig. 23, *pre.z.*, *post.z.*), which are densely cartilaginous, as are the transverse processes. Ligg. intertransversalia have been formed from the dorsal part of the " membrana reuniens." From the supradorsals, which are densely cartilaginous, the incipient neural spine has grown out as a median process. The thoracic ribs are completely ossified, only the tips of the tubercula and capitula remaining cartilaginous. There is a slight connection between the transverse processes and the tubercula. The vertebrarterial canals are still cartilaginous, except in the case of the atlas, where the lateral portion is still fibrous.

Both cervical and thoracic centra possess well-marked centres of ossification, which occupy the cores, and the perichondrium shows signs of having been broken down dorsally and ventrally (Plate 14, fig. 24, *b.c.ctr.*). The centra have collapsed slightly, as a result of ossification, each now having the form of a catenoid. The notochordal sheath still passes unbrokenly through the centres of ossification (Plate 14, fig. 24, *n.ch.sh.*). The epiphyseal plates, which are derived from the anterior and posterior zones of former perichordal discs, consist of regular rows of cartilage cells (Plate 14, fig. 24, *epip.*). These plates are ill-defined in the region of the atlas and axis, the intervertebral disc having chondrified as well, so that the odontoid process is now completely fused with the body of the axis. There is no ossification centre in the pro-atlas, which forms the tip of the epistropheus.

Bone has completely replaced cartilage in the base of the neural arch of the axis. A separate centre of ossification has appeared in the ventralmost part of the atlas ring, which centre is median and unpaired. The 1st cervical spinal nerve still exists, passing through the secondary foramen in the atlas neural arch along with the vertebral artery. It is the N. sub-occipitalis, which has lost its dorsal ganglion.

The median, subchordal and the lateral portions of the occipital rudiment are completely ossified, but the two portions are still connected by the densely cartilaginous ventro-lateral portion. The condyles are well developed, having grown considerably anteriorly and dorsally, and reach to the dorso-lateral part of the hind-brain. The subcondylar portions of the atlas have closely followed the condyles in their growth dorsally and are now as extensive as these structures. The foramen through which the N. sub-occipitalis and the vertebral artery pass has been shifted to the horizontal level of the dorsal part of the hind-brain by the condylar growth. In the posterior occipital region, the notochord has been preserved. It possesses a perichondrial investment, which stains an intense blue colour, and it lies above the basi-occipital rudiment, to which it is firmly attached. In the region between the occipital and the pro-atlas, the notochord is included in the tissue of the lig. apicis dentis.

Discussion—Notochord.

Carlier (1890) observed that in a sheep embryo $1\frac{1}{4}$ inches long, the notochord is becoming slightly bent into a V-shape in the region of the future intervertebral discs. The bends he attributed to the production in this region of a spindle-shaped body of dense tissue set in a jelly-like matrix. By subsequent growth, the spindle-shaped body displaces the notochord still further. Minot (1907), who does not cite the work of Carlier, examined the notochord in a number of mammals. In the sheep he observed a V-shaped displacement of the notochord in the intervertebral region, but also noticed a displacement in the opposite direction in the regions of the future vertebræ, and it is to his credit to have derived the series of alternate and opposite displacements from an original series of segmental flexures in the notochord. He maintained that this segmental wave is of general occurrence in mammals, showing that it occurs in the pig and the cat, and to a slighter degree in the dog, rabbit, and in man. The crests or dorsal curves of the waves in all these types, excepting the pig, occur in the future intervertebral region ; and in the pig, the position changes until the crest ultimately comes to lie near this region. Carlier did not observe the segmental waves, but maintained that the notochord shows an irregular curve which closely follows the curve of the definitive vertebral column and which, he suggested, plays some significant part in the determination of this curve. In Mus, there is no trace of this irregular curve, which cannot be more than transitory, since the curvature of the whole embryo and consequently of the notochord is greater than that of the adult animal, and is subject to almost constant change during the early stages of development when the vertebral structures are being formed. But the segmental flexures observed by Minot undoubtedly occur, although the ventral troughs occur in the region of the future intervertebral discs and not the dorsal crests as in the types studied by this worker. The series persists until chondrification is well advanced, when it is masked by the dilatations and constrictions.

Carlier also maintained that the spindle-shaped body opens out and gradually becomes indistinguishable from the rest of the tissue of the disc. The external causes to which he attributes the first displacement of the notochord are different from those indicated by the present work. In Mus, the earliest differentiation of the notochord is contemporary with the deposition of an intercellular matrix between the cells of the centrum rudiment. The cause seems without doubt to be the pressure exerted upon the notochord by this matrix as it is formed, the notochord being squeezed out into the future intervertebral disc region (Plate 12, fig. 12). It is thus a direct consequence of the process of chondrification and is not due to a change in the constitution of the perichordal disc, in which there does not occur a spindle-shaped body. There is no trace in Mus of an early series of very slight notochordal dilatations and constrictions, such as Froriep (1886) figured in the case of the ox and Piiper (1928) described in the bird. Such small displacements are difficult if not impossible to observe with certainty, where the notochord is of such small diameter as in the mouse (0·050 mm. at the onset of chondrification). In the ox, according to Froriep, the first dilatation of the notochord in the region of the disc occurs at the onset of ossification (commencement of the *definitiver Zustand*), and this he does not attempt to explain. Merely must I emphasize, in parenthesis, the enormous difference met with in the mouse, where at the onset of ossification the intervertebral diameter of the notochordal mass has increased from 0·05 mm. to at least 0·40 mm., while at the same time the centrum mass has long been devoid of notochordal tissue, excepting the sheath. The persistence of the notochord in the posterior occipital region, where it forms the basis of the lig. apicis dentis, is an important fact which I have not encountered in the literature.

Nucleus Pulposus.—The early work bearing upon the origin of the nucleus pulposus is considerable, but also confusing. The evidence brought to light was most unsatisfactory and it failed to give ground for any settled opinion. It was thought that the notochord played some part in the formation of this structure, although the controversy entered into by LUSCHKA, R. VIRCHOW, KÖLLIKER, ROBIN, DURSY, LOWE and LEBOUCQ was centred on the part played by the intervertebral disc, rather than that played by the notochord. It was generally held that the nucleus pulposus arises by liquefaction of the tissue of the disc and that the notochord may or may not contribute to its formation. GADOW (1895) appreciated the confusion and maintained that either such liquefaction occurs or that the notochordal expansion continues to grow, or that both these facts hold good. WILLIAMS (1908) gave what is probably the most correct account and maintained that the notochord loses its cellular nature and becomes syncytial, vacuoles filled with a mucin-like substance appearing in its cytoplasm. Later, the notochordal tissue comes to resemble cartilage, and persists in the region of the intervertebral disc as the nucleus pulposus. The diverse opinions on the question of the part played by the notochord or the disc, or both these structures, in the formation of the nucleus pulposus suggest that the mode of formation is not the same in all mammals. But it is interesting to find that in Mus, the notochordal tissue is clearly distinct and separate from the tissue of the intervertebral disc during all stages. The development of the notochord closely follows that of the corresponding structure in the pig, as described by WILLIAMS. The cytomorphosis of its cells occurs when chondrification is fairly well advanced (9·0 mm. stage) and it is accompanied by an increased activity of the nuclei within the syncytial mass. That there is no real degeneration of the notochordal tissue is readily seen when it is considered that during a growth of the embryo from 9·0 mm. to 14·0 mm., the bulk of the intervertebral expansion increases at least twenty times, after which it continues to increase, but more slowly (Plate 12, fig. 12, *n.ch.*; Plate 14, fig. 20, *n.ch.d.* 2 and Plate 14, fig. 24, *n.pulp.*).

Discussion—Sclerotomes.

REMAK (1855) was the first to draw attention to the resegmentation of the developing vertebral column (*Neugliederung der Wirbelsäule*) and showed that out of each "Urwirbelkern" or scleromere were developed, (1) the cranial portion of a vertebra together with its arches, (2) the intervertebral ligament, and (3) the caudal end of the next anterior vertebra. EBNER (1888) studied embryos of Tropidonotus and found that the sclerotomes are early divided each by a transverse cleft into anterior and posterior halves. This cleft, or " Intervertebralspalte," appears to be continued into the cavity of the dorsal part of the protovertebra and indicates the vertebral segmentation, the sclerotome halves recombining to form metameres which alternate with the myotomes. SCHULTZE (1896) identified the cleft in Selachians, reptiles and birds, and also in Bos, terming it the " segmentspalte." In reptiles, it is in primary communication with the myocœle, while in birds, a secondary communication with this cavity is established. In the trunk region of Bos, the primordia of the vertebral bodies are derived from the caudal half of one sclerotome and the cranial half of the next posterior sclerotome. WEISS (1901) identified the fissure in the rat, SCHAUINSLAND (1906) and PIIPER (1928) found it in birds, and BARDEEN (1905) observed it in the human embryo, noting that it appeared after the disappearance of the myocœle, at the end of the 3rd week.

In Mus, the intrasclerotomic fissure is first evident in an embryo of 4·0 mm. C.R.Lth. In slightly more advanced embryos it is better developed (Plate 12, fig. 7, *i.scl.f.*), extending from the horizontal level of the spinal ganglia to the level of the notochord, being better developed at the latter level and not at the former, as is the case in Tropidonotus. It is not continuous with the cavity of the myocœle, being formed only after complete differentiation of the myotomes, as PIIPER and BARDEEN found in the cases of the bird and the human embryo respectively. It disappears before the intersclerotomic fissure does, and this suggests that it is more vestigial than in birds, where, according to the most recent work it disappears after the intersclerotomic fissure. In the bird it is more vestigial than in reptiles, so that a series can be arranged from Tropidonotus to Mus in which the fissure is losing its ontogenetic significance. In the occipital region of the latter type it has lost this significance and never appears.

While a number of workers incline to agree with REMAK and v. EBNER, a few others doubt or deny the existence of the intrasclerotomic fissure and dispute the validity of REMAK's theory of " Neugliederung." FRORIEP (1886) failed to identify the fissure in the ox and, briefly discussing the views of REMAK, referred to "the now discarded theory of resegmentation." He believed that the resegmentation is apparent and not real, and attributed it to the oblique orientation of the primitive vertebral arch. The primitive axial skeleton he described, in contrast, as being quite continuous and unsegmented. The failure of this competent and careful observer to identify the intrasclerotomic fissure is almost inexplicable (his smallest embryo was 8·7 mm. and, according to his figure of it, possessed 36 somites). But it is conceivable that his nonchalant treatment of REMAK's theory is attributable to his misfortune in studying only the development of the cervical vertebræ. In Mus, the resegmentation is much more evident in the thoracic region than in the cervical region, a larger part of the loose tissue of the cranial sclerotomite contributing to the formation of the centrum rudiment in the former region than in the latter (*vide* Discussion, Sclerotomal Derivatives).

This is partly due to the relative shortness of the cervical region, especially in mammals where the individual vertebræ are shortened and tend to coalesce into a rigid mass of bone. Although FRORIEP treated the theory of resegmentation so lightly, yet his figures tend to substantiate it (particularly his fig. IV′) and I have no hesitation in predicting further support of the theory by a subsequent researcher studying the development of the thoracic vertebræ in the ox.

GADOW and ABBOTT (1895) lent support to the views of FRORIEP and firmly refused to be in any way tolerant of REMAK's theory. They assert categorically that nobody has ever seen a transverse splitting within a sclerotome, and that nobody has ever observed a healing up or recombination of these halves. They further maintain that the assumption of such a cleft is a theoretical necessity and an attempt to explain the "actually observed " overlap of myomeres and scleromeres. These conclusions arose as a result of an investigation of the development of the vertebral column in *Scyllium catulus*. In this type, the protovertebral plates curve like an S and overlap, the dorsal half being bent towards the median plane and caudalwards, while the ventral half is bent laterally and cranialwards. Each protovertebra forms a myotome and two sclerotomes, one of which is dorsal and the other ventral. A reconstruction of serial sections shows that, " the dorsal half of sclerotome 2 grows downwards, to come to lie behind the ventral half of sclerotome 1 ; the ventral half of sclerotome 2 lies in front of and below the dorsal half of sclerotome 3 ; and the formation of a physiological unit is effected by the recombination of the unequally numbered sclerotome halves, so that the dorsal half lies behind and above the ventral half." They add, " In the trunk region, the conditions are not so clear as in the tail, owing to the advanced mixing up of the dorsal and ventral cell clusters." And, " If A and B mean two successive sclerotomes, a and b their dorsal, and α and β their ventral halves, then the new scleromere is composed of $b + \alpha$ and not $\frac{A + B}{2}$, because $b + \alpha = \frac{\text{B dorsal}}{2} + \frac{\text{A ventral}}{2}$." The ventral sclerotome with its dorsal upgrowth is referred to as the ventral pyramid, the dorsal one with its ventral downgrowth, the dorsal pyramid. The basal portions of dorsal and ventral pyramids give rise to the basidorsals and basiventrals respectively. In forms possessing 4 pairs of arcualia in each segment, the second pair, namely the interdorsals and interventrals respectively, are produced by the apical parts of respective ventral and dorsal pyramids, as cell clusters situated between successive basidorsals and basiventrals. These findings are extended to other groups as " they (interbasalia) are present in Fish and in every other vertebrate, although by no means as independent typical interbasalia, but frequently modified into seemingly quite heterogeneous parts."

The theory of GADOW and ABBOTT cannot be applied to the development of the vertebral column of Mus, because, in the first place, the intrasclerotomic fissure, the existence of which these workers vehemently deny, is indubitably present (Plate 12, fig. 7, *i.scl.f.*). That this fissure is not merely an attempt to explain the growth dorsalwards of the cells of a ventral pyramid and the growth ventralwards of the cells of a dorsal pyramid is shown by the fact that the derivatives of these so-called pyramids do not occur on opposite sides of it. On the contrary, the basidorsal (or the chief component of the neural arch) and the basiventral (or rib-bearing element) occur in each segment on the posterior aspect of the intrasclerotomic fissure, which separates them from the elements giving rise to the interbasalia. If the views of GADOW and ABBOTT were correct, the fissure would separate the basidorsal from the basiventral. It is only by applying their nomenclature to structures according to the position of these when the elements are in the position they finally occupy, without concerning oneself with the assumed existence of "pyramids," that any degree of conformity is obtainable. Nevertheless it must be emphasised that the position of the elements suffers no change such as is maintained, all parts of the caudal sclerotomite consisting *ab initio* of dense tissue clearly distinct from the looser tissue of the cranial sclerotomite. Further, the protovertebræ or primitive somites are not curved into an S-shape in Mus, but on the contrary are arranged almost perpendicularly to the long axis of the body of the embryo, varying slightly according to the particular position. Thus in the anterior cervical region, the dorsal parts are inclined slightly cranially, in the posterior thoracic region slightly caudally, while in the anterior thoracic and posterior cervical regions they are truly perpendicular to the long axis of the body. The variation in orientation is a direct consequence of the curvature of the embryo.

Chondrification of the Sclerotomal Derivatives.—FRORIEP observed that in the ox, hyaline cartilage first appears in the region of the developing body of each vertebra, and later quite separately in the "Wirbelbogen." In the mouse, this is the case in the thoracic region, but in the cervical region, the loosening up of the tissue of the basidorsal (or "wirbelbogen") is contemporary with the deposition of violet-staining matrix between the loosely arranged cells of the centrum rudiment. He adds that, "die Bildung des Wirbelkörpers beginnt in der Cranialen Hälfte des im primitiven Zustand als Körperbezirk unterschiedenen Raumes zwischen den diesen Raum beiderseits umfassenden Interprotovertebralarterien." This accords well with the conditions in Mus, where chondrification first occurs in the loose tissue of the most posterior part of the caudal sclerotomite, *i.e.*, of the basiventralia. He adds, "Da dieser Raum cranial- und theilweise auch ventral- und lateralwärts von Derivaten des primitiven Wirbelbogens umgeben ist, so könnte die Vermuthung nahe liegen, dass die Bildung des Körperknorpels auf einem Wachsthumsprocess des Bogens beruhe." In his opinion two facts show that this is not the case. Firstly, the histological differences between the two rudiments and secondly the fact that the "Körperknorpel" is produced "mit einem Schlage." These differences hold good in the case of the mouse and probably in all other mammals, possibly possessing a phylogenetic significance. But it must not be overlooked that in Mus the basal portions of the basidorsals also contribute to the formation of the definitive centrum, being fused with the early centrum rudiment. Similarly, the interdorsal, which for a time remains membranous, also contributes to the formation of the definitive centrum, as does a portion of the perichordal disc (*vide infra*).

It was maintained by early workers on this problem that the centra arise as bilateral rudiments. Robin (1864) rejected this idea and maintained that the circular appearance of the rudiment of the body of a vertebra is specific for the mammalian embryo. Froriep (1886) and Bardeen (1905) found suggestions of this bilateral origin, the centrum rudiment at first consisting of a pair of lateral masses subchordally united by means of a strand of pro-cartilage, the whole appearing as a U-shaped structure. In Mus this U-shape is characteristic, and in the case of the axis, the earliest rudiment of the centrum shows no subchordal connection and is therefore truly bilateral. Froriep maintains that the U-shaped "Körperknorpel" later becomes closed dorsally, to form a ring; Bardeen that the perichordal septa (= interdorsalia) break down so that a complete ring is formed. In Mus, the fused interdorsals persist as a membranous formation until the centrum rudiment is considerably advanced and, becoming loosened up and chondrified contemporarily with the bases of the basidorsals, contribute to the formation of the postero-dorsal portion of the definitive centrum. Of an interdiscal membrane, such as Bardeen described as arising "where neural and costal processes spring from the primitive disc (= perichordal disc)," there is no trace in the mouse. When the centrum rudiment is formed, however, the nuclei are largely dispersed towards the periphery, to give rise to the perichondrium, which might be regarded as the "interdiscal membrane." That this view is probably correct is shown by a consideration of Bardeen's following statement, that "during the development of the interdiscal membranes, the primitive discs become hollowed out on the posterior surface." In Mus, the centrum rudiment early encroaches upon the tissue of the posterior part of the perichordal disc to a slight extent, so that this stage, when the perichondrium is being formed, is the stage at which the interdiscal membrane is being formed in the human embryo.

As regards the formation of zygapophyses, the present author cannot agree with Froriep, who maintained that the dorsal arch-piece grows backwards to form the posterior zygapophysis, towards which the arch piece of the next posterior vertebra grows out as the pre-zygapophysis, and that later dense connective tissue aggregates around these processes to form a "syndesmose" out of which the zygapophyseal joint arises. In Mus, the tissue which is destined to form the zygapophyses is laid down as dense membranous tissue in the dorsal part of the cranial sclerotomite before the obliteration of the intrasclerotomic fissure (Plate 11, fig. 4, on right of *v.sp.n.*). When the fissure disappears, it remains to connect adjacent basidorsals between the horizontal levels of the dorsal and ventral nerve roots. It remains in this position until the basidorsals come to consist of advanced cartilage, by which time it has come to occupy a supra-ganglionic position (Plate 13, fig. 17, *d.i.d.* 3). When centres of ossification appear in the basidorsals, the incipient dorsal-interdorsal becomes pro-cartilaginous, at the same time showing an oblique central split bordered by dense fibrous tissue. This is the primordium of the zygapophyseal or interneural diarthrosis. The rudimentary pre- and post-zygapophyses thus formed are not completely chondrified until the basidorsals are entirely evacuated of cartilage (Plate 14, fig. 23, *pre.z.*, *post.z.*). The closure of the neural arch in Mus is also dissimilar from that described for the ox by FRORIEP. In this latter type the cartilaginous arch pieces, growing dorsally on each side, pass over the nerve cord to fuse in the median dorsal line. In Mus, the closure is first effected by the development of a strand of fibrous connective tissue, uniting the distal extremities of each pair of basidorsals above the nerve cord. This median strand remains fibrous until the cartilage of the basidorsals is of the advanced hyaline type, when pro-cartilage cells come to lie on it, to form the primordium of a formation representing the supra-dorsal of GADOW. When the basidorsals are completely bony, the supra-dorsal still consists of dense early cartilage.

In the human embryo, according to BARDEEN, the laminar processes of the neural arches completely encircle the nerve cord, and from the region of fusion in the median line a pair of spinous processes grow out. The case in Mus is very different, for in it the spinous processes are developed as median dorsal outgrowths of the dense cartilage of the supra-dorsal. This occurs about the time when the basidorsals are almost completely evacuated of cartilage. In Sphenodon, according to SCHAUINSLAND (1906), the spinous processes are formed as median unpaired structures and are derived from a median cartilage which completes the neural arch. Thus the development of these processes in Mus conforms closely to the primitive mode, as displayed by Sphenodon, and differs considerably from the mode in Man.

Briefly recapitulating, the contributions made to the formation of the definitive centrum in Mus are as follows :—The early centrum rudiment is formed as a U-shaped structure below the notochord in the region of the intersegmental blood vessels. It is derived partly from the tissue of the most posterior portion of the cranial sclerotomite and partly from the tissue of the most anterior portion of the caudal sclerotomite, and thus is derived partly from the basalia and partly from the interbasalia. The basiventral component of this rudiment is more considerable in the cervical region than in the thoracic one. Later the interdorsal, which lies above the notochord in the anterior part of each segment, becomes chondrified and added to the centrum rudiment, and at the same time the bases of the basidorsals become loosened up and chondrified also to fuse with the centrum rudiment. Still later the perichordal disc becomes differentiated into three zones, and also partly contributes to the formation of the definitive centrum (*vide* Discussion, Perichordal Disc).

There remains to be considered the structure referred to as the perichordal tube. This structure arises as a membranous investment, 2 or 3 cells thick, around the notochordal sheath (Plate 12, fig. 8, *p.t.*). It remains membranous until the bases of the basidorsals and the interdorsals become chondrified, when the cells of which it is composed also become scattered and a matrix deposited between them. By this means, the notochordal sheath is no longer separated from the vertebral elements by membranous tissue but on the contrary is in cartilaginous connection with them, *i.e.*, the bases of the so-called arcualia have become effectively welded to the notochordal sheath. The perichordal tube is always most insignificant, but this is to be expected in a type where the notochord itself assumes microscopic proportions. It is certain that the notochord is the nucleus of the vertebral column but never in itself a supporting structure. The sheath, probably impregnated with chondrogenous substances, persists within the centrum as a continuous tube even when this structure is almost completely bony (Plate 14, fig. 24, *p.ch.sh.*). The process of ossification commences with a breaking-down of both dorsal and ventral perichondria, although ROBIN and BARDEEN maintain that the dorsal perichondrium alone is ruptured.

In conclusion to this part of the discussion, it might be pointed out that the bulk of the centrum is not derived from the interventralia, which contribute only to the postero-ventral portion of each vertebra. Hence, if the criterion which determines a Gastrocentral vertebra is the fact that interventralia form the major portion of the centrum, then the mammalian vertebra as illustrated by that of Mus is not of this type, as GADOW maintained.

Discussion—Perichordal Disc.

FRORIEP found that in the early stages of development of the vertebral column, the "primitiver Wirbelbogen" halves (=basidorsals) are fused with each other in the median line, both above and below the notochord. He says that, "der perichordale Theil des primitiven Wirbelbogen ist an der ventralen Seite der Chorda mächtiger und dichter gefügt als an der dorsalen. Ich habe das dadurch hergestellte festere Verbindstück der beiden Bogenhälften die hypochordale Spange bezeichnet." Later, the "primitiver Wirbelbogen" halves become disconnected from the perichordal part dorsally and in the "Uebergangsperiode," "der primitive Wirbelbogen ist kein einheitliches Gebilde mehr. Der perichordale Theil desselben hat sich durch longitudinalfaserige Auflockerung von den übrigen Bestandtheilen differenzirt, als Anlage des Lig. intervertebrale." The "hypochordale Spange" still connects the membranous arch-halves subchordally however. In the most anterior cervical segments this structure becomes chondrified, while in the most posterior segments of this region it diminishes in size ultimately to disappear. It is clear from the above that what FRORIEP refers to as the "hypochordal spange" is merely the ventralmost portion of the perichordal disc, which, becoming differentiated off from the rest of this structure, forms a strand of tissue subchordally connecting the rudiments of thoracic and cervical capitular processes. Undue stress appears to have been laid upon this insignificant "structure" in the literature on this subject. In Mus, it does not dwindle, eventually to disappear, but at one time underlies the anterior part of the centrum rudiment as membranous tissue, later to become absorbed into the mass of the centrum. In the thoracic region it partly contributes to the intervertebral ligament and partly to the posterior aspect of each vertebra along with the remaining portion of the anterior zone of the perichordal disc (*vide infra*). The fate of the remaining portion of the perichordal disc is not as simple in Mus as it is in the ox according to FRORIEP. Instead of becoming the rudiment of the lig. intervertebrale, it becomes differentiated into 3 zones, anterior, middle, and posterior in position. The anterior and posterior zones become chondrified and added to the posterior and anterior aspects respectively of adjacent vertebræ. The middle zone alone remains fibrous as the rudiment of the intervertebral ligament (Plate 13, fig. 18, *lig.i.v.*).

GADOW accepted FRORIEP'S observations and conclusions, but in addition maintained that the " hypochordale Spange " represents the ventralmost part of the basiventrals, the remaining portion of the disc representing the axial portion of these elements. In Mus, this is not the case, since the perichordal disc is derived from both cranial and caudal sclerotomites and thus cannot represent pure basiventralia, but instead represents fused inter- and basiventralia. The discs do not absorb the whole of these elements however, but strictly only the most posterior portion of the former and most anterior portion of the latter.

WEISSE (1901) applied the term " Horizontalplatte " to the perichordal disc, and he maintained that "aus der Horizontalplatte geht nur vorn die Anlage des Annulus fibrosus hervor; der übrige Theil der Bandscheibe ist eine sekundäre Bildung." He could not find a trace of the " hypochordale Spange," except in the case of the atlas, in the rat. Thus, the mode of formation of the intervertebral ligament in the rat is strikingly different from that observed in the case of the mouse.

BARDEEN has shown that " the cartilage of the vertebral bodies grows at the expense of the perichordal disc," which obtains in Mus as well as in Man. In dealing with the origin of this structure, he suggested that in all probability the most anterior portion is derived from the tissue of the anterior sclerotome half, since each disc is formed about the rudiment of the fissure of v. EBNER. It is gratifying to find that what he suggests as a probability in Man is actually the case in the mouse, where cells from both cranial and caudal sclerotomites are observed to enter into the composition of the disc (Plate 12, fig. 8, *p.d.*).

SCHULTZE (1896) found that the tissue of the perichordal discs becomes entirely chondrogenous, so that for a time the centra of the vertebræ are fused into a continuous cartilaginous axial column, and BARDEEN observes that this does not appear to be the case in Man, where, in all stages examined, some membranous tissue separates adjacent vertebral bodies. According to MÜLLER (CH.) (1906) capsules are developed in the chondrified disc tissue immediately around the notochord. In Mus as in Man, there is never a continuous axial column of cartilage. The middle zones of the discs remain fibrous, and although some slight matrix is deposited between the cells, this zone is never converted into tissue resembling cartilage.

Discussion—Atlas and Axis.

FRORIEP states that " die hypochordale Spange der ersten Halswirbelanlage sich in hyalinen Knorpel umwandelt und mit den knorpeligen Bogenhälften zu einem einheitlichen hypochordal geschlossenen Bogenknorpel zusammenfliesst." The "Bogenhälften" do not fuse with the centrum and so the atlas ring is obtained. In Mus, the origin of this ring cannot be explained in such simple terms. The bases of the basidorsals become chondrified but retain a fibrous tissue connection with the 1st perichordal disc, so that instead of contributing to the formation of the definitive centrum, they remain in isolation. The capitular homologues of the segment are greatly enlarged and, passing below the vertebral artery, meet the bases of the basidorsals. Chondrifying contemporarily with these structures, they contribute to an extensive and massive formation passing below the notochord, being here connected with each other by means of the now chondrified subchordal strand to form the ventral ring of the atlas. The strand of tissue connecting the capitular homologues, which is the "hypochordale Spange" of Froriep, forms only a small nodule of chondrified tissue immediately below the notochord and not the whole of the ventral ring of the atlas. The tubercular homologues are small and remain fibrous lateral to the vertebral artery, so that the vertebrarterial canal is never completed in cartilage and bone. The foramen in the atlas ring for the loop of the vertebral artery is not the vertebrarterial canal but is a secondarily developed foramen, resulting from a chondrification of a strand of tissue which is developed in front of this loop, extending from the ventral part of the ring to the dorsal part. The position of this strand of tissue is shown in Text-fig. 7, *s.t.a.t.* By subsequent growth it contributes to the formation of the atlas ring. The atlas ring subsequently ossifies by three centres, one for each lateral portion and a median centre for the subchordal nodule.

Weisse states that the "Horizontalplatte" is only slightly developed in the rat, and that the "Vertikalplatte," which connects this with the membranous "Bogenhälften," is undeveloped in the 1st cervical segment. We have seen that, in Mus, the perichordal disc (= Horizontalplatte) is well developed and extends throughout almost the whole of the posterior half of this segment. The ventralmost portion of this structure gives rise to the mid-ventral nodule of the atlas ring. The bases of the basidorsals (= Vertikalplatten) are also well developed and make a considerable contribution to the formation of the atlas ring. Thus, the development of the atlas is very different in the rat, according to the findings of Weisse, from that in the mouse.

The literature on this subject does not give any precise indication of the segmental position of the posterior limit in the axis. It is to be observed that the centrum of this vertebra is formed in a typical manner, so that the anterior zone of the 3rd perichordal disc becomes fused with the axis centrum rudiment to form the posterior part of this vertebra. The odontoid process is formed from the tissue of the first two perichordal discs together with the centrum rudiments of the atlas and the pro-atlas. The articular facet on the axis for the atlas ring is derived from the considerably widened interbasalia of the 2nd cervical segment.

Discussion—Pro-atlas.

ALBRECHT (1880) pointed out that " der n Spinalnerv eines amnioten Wirbelthiere durch n-1 Wirbel desselben austritt." He reasoned that if we make $n = 1$, then the 1st cervical spinal nerve of the amniote passes through the 0th vertebra. This 0th vertebra is situated between the occipital region and the atlas and is called the " pro-atlas." He wrongly believed that the amphibian 1st spinal or nerve sub-occipitalis is non-existent, and accordingly stated, " mithin ist der sogenannte Atlas der Amphibien dem Atlas der Amnioten nicht homolog. Und mithin ist überhaupt der sogenannte Atlas der Amphibien in Wirklichkeit der Proatlas derselben." It was pointed out that while the N. sub-occipitalis is not present in the Anura, yet it occurs in the Urodela ; and GADOW, speaking of the odontoid-like process in the latter group, said that it belongs to a scleromere the rest of which, namely, the dorsal arcualia, has been added to the occipital part of the cranium. The N. sub-occipitalis is the nerve of this scleromere. PETER (1895) showed that this nerve occurs in Bufo and in the Gymnophiona, but is lost in Rana* amongst anuran types. ALBRECHT thought he had found remnants of the pro-atlas in the form of a free dorsal piece on the top of the atlas of crocodiles, of Hatteria, and even of Erinaceus, and called this piece the " eparcuale pro-atlantis." He was supported by HOFFMANN, WIEDERSHEIM, DOLLO, BAUR and HOWES, but failed to receive support from GADOW and CORNET. GADOW believed this piece to be the neural spine of the atlas. Making use of the work of FRORIEP, he criticised ALBRECHT'S attempt to homologise the amphibian pro-atlas, stating that " in the Amniota at least two neuromeres and scleromeres have been added to the cranium, the first two post-cranial nerves of the Amphibia being converted into the hypoglossus of the Amniota. Consequently neither the ' atlas ' nor the second vertebra of Amphibia is strictly homologous with the atlas of the Amniota." He referred to these vertebræ as " pro-atlantic vertebræ." In Mus the evidence points to the fact that at least 5 segments contribute to the formation of the occipital region of the cranium, so that, accepting GADOW'S nomenclature, at least five of the most anterior vertebræ of Amphibia are "pro-atlantic vertebræ." The formation of the hypoglossal nerve is intimately connected with the development of the cervical flexure, and although little that is not extremely speculative can be advanced regarding the inclusion of the pro-atlantic vertebræ into the occipital region of the cranium, yet it seems that in mammals generally, an explanation of the phylogenetical significance of this flexure would provide the key to an elucidation of the problem.

But the question of the so-called pro-atlas is quite distinct from that of the pro-atlantic vertebræ. The predominant view is expressed by HAYEK (1923) when he says that in many mammalian types, " bildet sich aus dem Körper des Proatlas die Spitze des Dens Epistrophei." He states that, " die Ausdrücke Proatlas und Occipitalwirbel werden heute vielfach als Synonima verwendet," but finds that the last

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* In Rana it can be discovered in the tadpole stage, according to unpublished work of Dr. H. K. MOOKERJI.

occipital vertebra has a well-developed "hypochordale Spange" and contributes to the formation of the point of the odontoid process. Thus he justifies himself in stating, "danach ist also der Proatlas der letzte (kaudalste) Occipitalwirbel und darf nicht als Occipitalwirbel schlechtweg bezeichnet werden." But BARGE (1915) and (1918) gets nearer to the heart of the problem. Accepting the theory of "neugliederung" advanced by REMAK, he questions the position of the cranio-vertebral interval. Considering the work of FRORIEP, WEISSE, GAUPP and BARDEEN, he finds that it is intersclerotomic, *i.e.*, occurs at the anterior limit of a segment. Then he shows that if the resegmentation theory holds good, a half segment remains left over between the 1st cervical vertebra and the occipital region, for, calling the cranial half of a segment *a* and the caudal half *b*, in general the composition of the *n*th vertebra is expressed by the metamerical formula $S.n.b. + S\,(n+1).a.$ In the case of the 1st vertebra, the portion $S1.a.$ remains separated from the caudal half of the vertebra lying cranially to it by the cranio-vertebral interval. He questions the potency of this segment half, but does not give a categorical answer. Instead he gives an opinion that it is connected with variations of the atlas in the region of the sulcus arteriæ vertebralis, with the pro-atlas hypothesis of ALBRECHT, with the concrescentia atlanto-occipitalis and the manifestation of occipital vertebræ. Later (1917), he added to the proofs he obtained from the literature, by an investigation carried out on the sheep, and showed that the anterior sclerotome half of the 1st cervical segment fuses with the neural processes and "hypochordale Spange" of the atlas to form the "massæ laterales." There is no vertebral body corresponding to this half segment as in the case of more posterior segments.

The work of BARGE can be applied in part to the case of Mus, where the cranio-vertebral interval is undoubtedly intersegmental in position and where evidence is afforded in support of the resegmentation theory. But in the cranial sclerotomite the elements characteristic of more posterior centra are laid down, *i.e.*, the interdorsalia and interventralia. Thus, although BARGE was unable to prove that the pro-atlas is thus derived yet the proof is afforded by a consideration of the development in Mus. There is no dorsal-interdorsal or zygapophysis rudiment in this hemi-segment, but as we have seen there is a strand of fibrous tissue stretched in front of the loop of the vertebral artery, which ultimately gives rise to the portion of the atlas ring anterior to the foramen for this artery. This strand is not developed *ab initio,* but is a secondary formation. Nevertheless out of consideration of its position alone one can infer that it is derived from the tissue of the cranial sclerotomite and is thus a derivative of the hemi-segment immediately behind the cranio-vertebral interval. It thus corresponds to the tissue which contributes to the formation of the "massæ laterales" in the sheep.

The pro-atlas chondrifies and ossifies relatively later than the cervical centra, which in turn become transformed later than the fused occipital centra, so that histologically as well as genetically it is distinct from these regions. In the nomenclature of GADOW it may be defined as "the axial portion of the interbasalia of the first cervical segment which remains at the anterior end of the vertebral column after the resegmentation of the cervical and thoracic vertebræ to form the point of the odontoid process, the cranio-vertebral interval being intersegmental in position."

Discussion—Occipital Region.

It has long been known that the occipital region of the cranium is derived from an uncertain number of segments. **Froriep** gives careful consideration to the development of this region in the ox. He found that it arises out of the fusion of 4 vertebræ-equivalents, of which only the most posterior one develops as an independent vertebra prior to fusion. This he terms the " occipital vertebra." Cranialwards of this vertebra he found three arch rudiments, three myotomes and three ventral nerve roots. The sclerotomic derivatives of these metameres give rise to what he calls the " scheinbar ungegliederte Abschnitt " of the occipital rudiment. **Jager** (1924) found that in the occipital region of a chick embryo, 3 to 4 days old, two cartilaginous vertebral bodies are seen to be fused with the more anteriorly situated and unsegmented parachordal rod. These are fused with the atlas, but towards the end of the 4th day become separated from it. One or two arch pieces corresponding to these bodies are formed also. Behind the vagus nerve 4 myotomes are developed. In some cases a 5th is developed beneath the vagus, and rarely a 6th occurs in front of this. Only the two most caudally disposed segments thus indicated possess ventral nerve roots which are retained. There are neither dorsal roots nor ganglia.

In Mus the early stages of development of the occipital region are characterised by the presence in front of the 1st cervical segment of 5 rudimentary somites. Only the most posterior one possesses dorsal ganglia, which are smaller than those of more posterior segments. The rudimentary somites are differentiated into equally rudimentary dermato-myotomes and sclerotomes, excepting the most posterior one, which is almost as well developed as the 1st cervical segment. Intersegmental arteries and veins are developed between the somites and in front of the most anterior one. The most cranially disposed segment does not develop ventral nerve roots and fairly early loses its dermato-myotome (4·0 mm. stage). The second segment develops ventral nerves, but these disappear before the hypoglossal nerve exists as such, the corresponding dermato-myotome persisting however. The remaining three pairs of ventral nerves mark the limits between the four pairs of persistent sclerotomes (Text-fig. 3, iii, iv and v). The first pair of the latter is really compounded of the sclerotomal tissue of the first two of the five segments observed. When all the segments are evident, there is in addition a region anterior to them and posterior to the developing otic capsule which is devoid of segmentation. This suggests that the limiting number of segments involved in the production of the occipital region of mammals is not necessarily fixed at five. The fusion of the perichordal part of the whole occipital region takes place contemporaneously with the development of the cervical flexure, and when this attains its maximum the fusion is complete. This takes place while the vertebral components are still membranous, so that the fusion is relatively earlier in Mus than in the ox, where the "occipital vertebra" is separate and distinct from the "scheinbar ungegliederte Abschnitt" until chondrification takes place.

Throughout the whole occipital region, the perichordal tissue is loose. This obtains in all probability because of the fact that the notochord is widely removed from the hind-brain, especially in the posterior part of the region, as a result of the action of the cervical flexure. When chondrification ensues, the whole of axial tissue becomes converted into a rod of cartilage. The notochord lies in a longitudinal groove on the dorsal aspect of the rod. The dorsal tissue of the fused segments chondrifies slightly later to form what has been referred to as the dorso-lateral mass. At this stage, the ventral nerves of the three most posterior segments still pass separately through the membranous tissue connecting the dorso-lateral mass with the centrum portion of the occipital rudiment, but later a foramen is developed. These nerves form the rudiment of the hypoglossal nerve.

Discussion—Ribs.

FRORIEP found that in the early stages of development, the "primitive Wirbelbogen" grow out until they come in contact with the muscle-plate of the segment to which they belong. The bulge of the muscle-plate causes each of them to expand dorso-ventrally, thus effecting a division into neural and costal processes. The costal processes grow out, each past the edge of its muscle-plate as the rudiment of the rib. In Mus the sclerotome is early divisible into dorsal and ventral, as well as anterior and posterior halves. The lateral portion of the ventral division of the caudal sclerotomite represents the primordium of the rib and its processes. This portion later becomes differentiated into a dorso-lateral part, which is the incipient tuberculum, a ventro-lateral part which is the rib rudiment, and a ventro-mesial part, which is the rudiment of the capitulum (Plate 12, fig. 9, *tub.*, *rib*, *cap.*). In the cervical region, the capitulum consists of tissue derived entirely from the caudal sclerotomite and passes into the posterior part of the perichordal disc (Text-fig. 1 (A), *c.cap.*). In the thoracic region, it consists partly of tissue similarly derived, but partly of cells from the most posterior part of the cranial sclerotomite (Text-fig. 1 (B), *cap.*), and it abuts on the lateral part of the disc. The rib proper grows ventralwards in the most posterior part of the segment to which it corresponds, and as far as can be ascertained does not arise by differentiation of mesenchyme "in loco" as is suggested in some text-books. The incipient tuberculum is from the outset in membranous connection with the primordium of the basidorsal, which connection is retained long after these structures have become cartilaginous (Plate 4, fig. 21, *tub.*). The costo-transverse diarthrosis first appears as a split in the dense fibrous tissue between the cartilaginous tuberculum and neural arch. At this stage there is no definite diapophysis, which develops later as a small peg-like outgrowth of the neural arch. This is not the case in the human embryo, where, according to BARDEEN, the transverse processes are at first short processes, lying at some distance from the corresponding ribs, towards which they grow outwards and forwards. The present research has lent support to the opinion of GADOW, that the cervical and thoracic ribs are homologous structures ; but more than this, it has indicated that in the cervical region, the tubercular and capitular homologues give rise to the vertebrarterial canal. In the atlas vertebra a further modification is seen, the capitular process being greatly enlarged and contributing to the formation of the ventral part of the ring.

Discussion—Ontogeny and Phylogeny.

FRORIEP recognised three stages in the development of the vertebral column of the ox, which he called, "primitiver Zustand," "Uebergangsperiode" and "definitiver Zustand" respectively. During the first stage the vertebral rudiments are being laid down, during the second the centra are becoming formed in cartilage, and, in the last, the arch pieces are chondrifying and are fusing with the centrum to form a cartilaginous vertebra. Tissue substitution takes place during the last stage and the vertebra becomes bony. BARDEEN referred to three stages similarly in the case of the human, as Blastemal, Chondrogenous and Osteogenous respectively. The blastemal period terminates immediately before the first chondrogenic centre is developed; during the chondrogenous period the cartilaginous vertebra is completely formed, to become bony during the third period. Thus, these two authorities set the same final limit to the first period in each case. The "Uebergangsperiode" of FRORIEP, however, corresponds to a part only of the Chondrogenous period of BARDEEN, which includes the major part of the "definitiver Zustand" of the former. In Mus the development of the vertebral column falls naturally into 4 stages, which may be referred to as Mesenchymatous, Sclerotogenous, Chondrogenous and Osteogenous respectively. During the early part of the Mesenchymatous stage, the notochord is surrounded by loose mesenchymatous tissue. It is the nucleus about which the vertebral column will be subsequently formed but is not an axial supporting organ. The somites are becoming differentiated into dermato-myotomes and sclerotomes. About the middle of the period the sclerotomes reach ventrally on each side almost to the level of the notochord, and during the latter part of the period a subchordal union is effected. The intrasclerotomic fissure is developed and reaches its maximum development at the end of the period, which is indicated in an embryo of 5·5 mm. C.R.Lth. The Sclerotogenous stage commences with an obliteration of the intrasclerotomic fissure and the development of the perichordal disc from the tissue of cranial and caudal sclerotomites. Membranous basidorsals, interdorsals and dorsal-interdorsals are formed, and primordia of the ribs are laid down. The cervical flexure reaches its full development towards the close of the period, which occurs between the lengths of 6·5 mm. and 7·2 mm. The Chondrogenous period opens with the formation of the centrum rudiment and contemporary chondrification of the basidorsals and ribs.

About the middle of the period, the pro-cartilaginous basidorsal fuses with the centrum rudiment and the perichordal disc is separated from the tissue of the former. The interdorsals and the rib-processes become chondrified and the supra-dorsal is developed as a fibrous tissue strand completing the neural arch. The zygapophyses are formed towards the end of the period, when the perichordal disc becomes differentiated into 3 zones, the anterior and posterior of which become chondrified and added to the posterior and anterior aspect of adjacent vertebræ respectively. The posterior limit of this period is marked in an embryo of 14·0 mm. C.R.Lth. During the Osteogenous period, bone centres are laid down in the angles of the ribs, the neural arches and the centra in turn, the last being formed just prior to birth.

The ontogenetic Membranous stage is too indeterminate to bear comparison with any phylogenetic stage in the ancestry of the Mammalia, but the division of the sclerotomes by the development of the intrasclerotomic fissure towards the end of the period recalls the conditions characterising the "Ganoid" fishes and certain primitive Stegocephalia. The obliteration of this fissure during the Sclerotogenous period may be taken as indicating the first definite advance from the primitive Ganoid condition. The perichordal disc, which is formed about the vestige of the intrasclerotomic fissure and which lies immediately below and in front of the basidorsal, must be regarded as an intercentrum. There is little resemblance between the disc in Mus and the intercentrum of a Stegocephalian vertebra, but it must not be overlooked that the notochord has dwindled to most insignificant proportions in the former type. The interdorsal is to be regarded as fused pleurocentra. At the end of the second period there are thus three dense condensations of cells which represent the intercentrum, pleurocentra and basidorsals respectively. The condensations are continued one into another, giving rise to an embolomerous arrangement, which, if in an osseous condition would parallel the arrangement found in certain Stegocephalia, *e.g.*, in the caudal vertebræ of Cricotus. The rhachitomous condition is not indicated at all. The basidorsals, interdorsals and the ribs ossify as they stand, but the intercentrum undergoes change. It divides into zones, which contribute to adjacent vertebræ and at the same time form an intervertebral pad of fibrous tissue. This indicates the probable line of advance from the embolomerous arrangement indicated in the vertebral column of the Stegocephalian-like ancestor of the Mammalia. It indicates the development of greater rigidity in the individual vertebræ, and a contemporary increasing of the flexibility of the column as a whole, by the sacrifice of the intercentrum as a bony element. The ontogenetic development of the cervical flexure is completed at the close of the Sclerotogenous period, when the embolomerous arrangement of elements obtains, so that the changes in habits which affected the changes in the structure of the vertebral column mentioned above, are possibly connected with the phylogenetic significance of this flexure. The connection between the development of the flexure and the formation of the highly modified occipital region is a close one ontogenetically. That it is possibly also a close one phylogenetically is the inference one draws from this fact.

Summary.

(1) Sclerotomic cells are liberated from the dorso-mesial portions of the primitive somites. The notochord is very slender and is surrounded by loosely arranged mesenchymatous cells.

(2) Five rudimentary segments are seen in the anterior region of the embryo. Only the most posterior one possesses dorsal ganglion cell-clusters, which are smaller than typical ones. Intersegmental arteries pass between the segments and in front of the most anterior one. Still later, intersegmental veins are formed in front of and slightly lateral to the arteries.

(3) The whole mesial walls of the primitive somites are breaking down to form the sclerotomes, which appear as wedges of tissue filling up the space between the dermato-myotome and the nerve cord. This tissue does not extend as far as the notochord ventrally.

(4) The notochordal sheath is formed and a series of strictly segmental waves appears in the notochord. The dorsal crests lie within a segment, while the ventral troughs occur in the region of the intersegmental blood vessels. The waves do not occur in the region of the anterior rudimentary segments.

(5) Ventral spinal nerve roots appear in the anterior part of each segment, excepting the most anterior of the five rudimentary ones, which has lost its dermato-myotome.

(6) A narrow transverse cleft, the intrasclerotomic fissure, is formed within each sclerotome, dividing this into cranial and caudal halves or sclerotomites. The cranial sclerotomite consists of loose tissue, while the caudal one is formed of densely crowded cells.

(7) The cervical flexure appears in the anterior region of the embryo and effects a slight crushing of the rudimentary segments, which are the occipital segments. The nerves of these segments are closely approximated as a consequence.

(8) The caudal sclerotomite is now histologically divisible into dorsal and ventral halves. The dorsal division, in which the nuclei are elongated and arranged radially with respect to the notochord, is the incipient Basidorsal, while the ventral division, in which the nuclei are round and do not display any definite orientation, is the forerunner of the Basiventral.

(9) Condensed tissue appears in the dorsal part of the cranial sclerotomite between the dorsal ganglia and the ventral nerve roots, and connects adjacent incipient basidorsals. It forms the membranous forerunner of the Dorsal-interdorsal.

(10) A few densely aggregated cells extend around the notochord. These are derived chiefly from the ventral division of the caudal sclerotomite, but partly from the corresponding division of the cranial one. The membranous vertebral elements are thus in connection with the notochord.

(11) The 1st cervical segment is clearly defined by its more massive and extensive ventral division of the caudal sclerotomite. In front of it, three pairs of ventral spinal nerves are still to be seen, the most anterior pair having disappeared. These occipito-spinal nerves are the forerunner of the hypoglossal nerve. They mark the boundaries

of the corresponding sclerotomes, the tissue in front of the most anterior one having been derived from the sclerotomes of the first two rudimentary occipital segments.

(12) The intrasclerotomic fissure has become obliterated, but it is still clearly indicated that cells from both sclerotomites envelope the notochord, to form the rudiment of the perichordal disc.

(13) Occipital, cervical, and thoracic regions are well defined although still entirely membranous. In the cervical region the vertebral arteries are complete. The neck-flexure is well developed, the hind-brain being inclined to the spinal cord at an angle of 90°. The perichordal parts of the segments near the cranio-vertebral interval are more completely crushed, but the interval is clearly marked by the subchordally persistent intersclerotomic fissure. It is found to lie at the anterior extremity of the 1st cervical segment.

(14) The thoracic and cervical ribs with their capitular and tubercular processes become differentiated from the tissue of the ventral division of the caudal sclerotomite, *i.e.*, from the basiventral. In the cervical region, these processes form the lateral portion of the vertebrarterial canal. In the thoracic region, a slight contribution is made to the formation of the membranous capitulum by cells derived from the cranial sclerotomite.

(15) In both cervical and thoracic regions, a dense condensation of cells appears immediately above the notochord in the cranial part of the segments. It is median and passes into the perichordal disc posteriorly. It represents fused Interdorsalia. The tissue below and lateral to the notochord in this portion of each segment, and also in most posterior portion of each segment, is loosely arranged skeletogenous tissue.

(16) In the loose tissue mentioned above, a violet-staining intercellular matrix is deposited, which marks the onset of chondrification. Thus the rudiment of the centrum is laid down and is a derivative of both basalia and interbasalia, arising from the tissue of both sclerotomites, *i.e.*, basiventrals and interventrals.

(17) The inner tissue of the prechondral basidorsals is loosened up and an intercellular matrix is deposited between the loosened cells. This occurs first in the cervical region and slightly later in the thoracic region.

(18) Chondrification is well advanced in the occipital region. The cells of the loose perichordal tissue in this region now lie in tiny capsules. The lateral part of the occipital rudiment is still membranous however.

(19) A layer of cells two or three nuclei thick lies closely apposed to the notochordal sheath, which is thus separated from the early rudiments of the centra. There is no matrix between the cells, so that the formation, or perichordal tube is strikingly contrasted with the pro-cartilage of the centra.

(20) The inner tissue of the thoracic ribs is becoming loosened up as did that of the basidorsals at an earlier stage and a matrix is similarly deposited. Chondrification is extending slightly into the processes of the thoracic ribs, although in the cervical region the vertebrarterial canals are still membranous.

(21) In the 1st cervical segment, the perichordal disc consists of looser tissue than is found in a typical disc, and it extends throughout almost the whole of the posterior half of its segment. The capitular homologues are immense and, together with a strand of tissue which connects the pair subchordally, form the primordium of the ventral ring of the atlas.

(22) The basidorsals are pro-cartilaginous, but are still in membranous connection with the perichordal disc anteriorly. The basal portions are becoming loosened up, chondrified and amalgamated with the antero-dorsal portion of the centrum rudiment.

(23) The membranous interdorsal of the 2nd cervical segment, becoming laterally extended and co-extensive with the basidorsals of the 1st and 2nd cervical segments, forms, together with the equally widened interventrals, the articular facets on the axis for the atlas ring.

(24) In the occipital region, the dorso-lateral mass is pro-cartilaginous. The basioccipital rudiment consists of definite cartilage, but between this and the dorso-lateral mass is a ventro-lateral mass of membranous tissue. Through this tissue the occipito-spinal nerves, or factors of the hypoglossal, pass separately.

(25) The notochord is dilated in the region of the perichordal disc in each segment, so that the notochordal waves are no longer evident. The mid-segmental diameter of the notochord is 3 or 4 times the diameter in the intersegmental region. The notochordal tissue, now syncytial, has been squeezed out of the intersegmental regions into the regions of the perichordal discs by the developing cartilage of the centrum rudiments.

(26) The interdorsal condensations are now chondrified, except in the case of the 1st and 2nd cervical segments, and contribute to the formation of the centrum rudiments in the postero-dorsal parts.

(27) Each pair of basidorsals is connected above the nerve cord by a strand of connective tissue. Pro-cartilage cells come to lie on the strand and a slow process of chondrification ensues, to give rise to a median formation, which is the Supra-dorsal.

(28) The cells of the perichordal tube become dispersed and the cartilaginous matrix of the centrum rudiment becomes continued up to the notochordal sheath. In this manner the centrum is welded to the notochordal sheath.

(29) The pro-atlas consists of a chondrogenous ventral portion derived from the interventrals of the 1st cervical segment and a still membranous interdorsal. The dorsalmost part of this latter, together with tissue derived from the dorsalmost part of the perichordal disc, forms the lig. transversum atlantis.

(30) The diameter of the notochordal mass in the region of the perichordal disc has increased until it is now about 8 times that in the region of the centrum rudiment. The sheath is still unbroken.

(31) Basidorsals and ribs consist of very advanced cartilage, while the supra-dorsals and the rib processes, together with the homologous portions of the vertebrarterial canals, consist of early embryonic cartilage. The dorsal-interdorsals are still membranous,

later they become chondrified, and a cleft arises in each, to form the earliest indication of the interneural diarthrosis between adjacent pre- and post-zygapophyses.

(32) Thoracic and cervical perichordal discs alike have become differentiated into 3 zones, which are anterior, middle, and posterior in position. The anterior and posterior zones chondrify and fuse with the posterior and anterior faces of adjacent centrum rudiments. The middle zone remains fibrous as the rudiment of the intervertebral ligament.

(33) Condyle rudiments are formed from the ventro-lateral part of the occipital rudiment, which now consists of dense cartilage. Formed below the level of the nerve cord and behind the hind-brain, they grow antero-dorsally until they come to lie on each side of the dorso-lateral part of the hind-brain. The parts of the atlas ring adjacent to the condyles grows so as to follow these structures.

(34) A strand of fibrous tissue is developed in front of the loop of the vertebral artery, stretching from the condylar portion of the atlas ring to the dorsal part, thus enclosing the vertebral artery in a secondarily derived foramen. The lateral part of the vertebrarterial canal of the atlas ring remains fibrous.

(35) The 1st and 2nd perichordal discs are completely chondrified so that the rudiment of the odontoid process is formed. The 3rd perichordal disc marks the posterior limit of the axis centrum.

(36) Ossification centres first appear in the angles of the thoracic ribs. Later similar centres appear in the lateral parts of the thoracic basidorsals and still later in the corresponding parts of the cervical basidorsals. All these centres are single ones.

(37) The basi-occipital ossifies by a median subchordal centre, and a single centre appears in each dorso-lateral mass. The ventro-lateral mass consists of dense cartilage and through it passes the hypoglossal nerve.

(38) The notochordal mass becomes cellular again. It is quite distinct from the tissue of the intervertebral ligament.

(39) When the basidorsals and the ribs are completely evacuated of cartilage, a median ossification centre appears in each centrum. There is no centre in the pro-atlas even when the other vertebra show well-developed centres.

(40) The rudiments of the spinous processes grow out from the dense cartilage of the supra-dorsals as unpaired formations. When this occurs, the basidorsals are entirely bony.

(41) A separate median centre of ossification appears in the ventralmost part of the atlas ring.

(42) The tips of pre- and post-zygapophyses and the tissue of the capitular and tubercular processes long remain densely cartilaginous. The vertebrarterial canals are also cartilaginous.

(43) The notochordal sheath persists when ossification is well advanced and passes unbrokenly through the bony core of each centrum.

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DESCRIPTION OF PLATES.

Abbreviations.

a.bas., basilar artery.
a.c.v., anterior cardinal vein.
a.c.v.d., anterior cardinal vein near ductus Cuvieri.
ao., aorta.
ao.l., last aortic arch.
a.r.d.my., most anterior rudimentary dermato-myotome.
a.r.scl., most anterior rudimentary sclerotome.
arth.d., arthrodial diarthrosis.
at., atlas.
at.b.d., atlas basidorsal.
at.cap., capitular process of atlas.
at.ctr.a., atlas centrum rudiment.
at.lat.m., lateral mass of atlas.
at.tub., tubercular process of atlas.
ax.art., articular surface on axis for atlas.
ax.cap., capitular process of axis.
ax.ctr.a., axis centrum rudiment.
ax.tub., tubercular process of axis.
b.b.d., base of basidorsal.
b.c.ctr., bone centre in centrum.

b.c.n.a., bone centre in neural arch.
b.c.r., bone centre in rib.
b.d., basidorsal.
*b.d.*5, basidorsal of 5th cervical vertebra.
b.oc.a., basi-occipital rudiment.
*b.v.*4, basiventral component of 4th cervical centrum.
b.w., body-wall.
cap., capitulum rudiment.
*cap.*3, capitulum rudiment of 3rd cervical vertebra.
cart., cartilaginous connection between atlas centrum and capitulum of axis.
c.s.t., tissue overlying cervical sinus.
c.scl.t., cranial sclerotomite.
*c.scl.t.*1, cranial sclerotomite of 1st cervical segment.
*c.sp.g.*1–*c.sp.g.*5, 1st to 5th spinal ganglia.
c.t., connective tissue.
ctr.a., centrum rudiment.
d., dermatome.
d.C., ductus Cuvieri.
d.h., developing heart.
d.i.d., dorsal-interdorsal.
*d.i.d.*3, dorsal-interdorsal of 3rd cervical segment.
d.m.r., dorsal membrana-reuniens.
d.my., dermato-myotome.
d.r.sp.n., dorsal ramus of ventral spinal nerve.
epip., epiphysis.
epis., epistropheus or axis.
fis., intersclerotomic fissure marking cranio-vertebral interval.
h.xii., hypoglossal nerve (= Cranial xii).
i.d., interdorsal.
i.d.pr., interdorsal component of pro-atlas.
i.s.a., intersegmental artery.
i.s.a.c.o., intersegmental artery between atlas and occipital.
i.s.b.v., intersegmental blood vessel.
i.s.v., intersegmental vein.
i.scl.f., intrasclerotomic fissure.
*i.v.*4, interventral component of 4th cervical centrum.
i.v.pr., interventral component of pro-atlas.
k.scl.t., caudal sclerotomite.
l., lung.
lig.a.d., lig. apicis dentis.
lig.i.v., intervertebral ligament.
*lig.i.v.*2,3, intervertebral ligament between axis and 3rd vertebra.
lig.l.ant., lig. longitudinale anterius.
lig.l.post., lig. longitudinale posterius.
lig.tr.at., lig. transversum atlantis.
m.t., membrana tectoria.
med.ob., medulla oblongata.

my., myotome.
n.c., neural canal.
n.ch., notochord.
n.ch.sh., notochordal sheath.
*n.ch.d.*1, notochordal dilatation of 1st cervical segment.
*n.ch.d.*2, and *n.ch.d.*3, 2nd and 3rd notochordal dilatations.
n.k.scl.t., dorsal division of caudal sclerotomite.
n.pulp., nucleus pulposus.
oc., occipital rudiment.
oc.scl., last occipital sclerotome.
oc.c.d., condylar portion of occipital.
oc.l.m., lateral mass of occipital.
oes., oesophagus.
otic c., otic capsule.
p.d., perichordal disc.
p.d.at., perichordal disc of 1st cervical segment.
p.d.ax., perichordal disc of 2nd cervical segment.
p.d.a.z., anterior zone of perichordal disc.
p.d.p.z., posterior zone of perichordal disc.
p.o.a.lig., posterior occipito-atlantal ligament.
p.o.i.a., point of origin of intersegmental artery.
p.p.ctr., posterior part of centrum.
p.mch., perichordal mesenchyme.
p.t., perichordal tube.
pet.g., petrosal ganglion.
ph., pharynx.
post.z., post-zygapophysis.
pr.at., pro-atlas.
pre.z., pre-zygapophysis.
r.com., ramus communicans.
rib, rib rudiment.
s.ac.n., spinal accessory nerve (= Cranial xi).
s.d., supra-dorsal.
s.g.c.c., spinal ganglion cell-cluster.
s.r.sp.n., sensory root of spinal nerve.
s.s.c., subchordal strand connecting a pair of capitula.
s.t.at., tissue enclosing loop of vertebral artery.
scl., sclerotome.
sp.c., spinal cord.
sp.g., spinal ganglion.
*sp.n.r.*1, 1st cervical spinal nerve-root.
sym.g., sympathetic ganglion.
syn., synovium.
syn.td., synovium of 1st trochoid diarthrosis.
tub., tuberculum rudiment.
u.om.b.v., blood vessel formed by union of umbilical and omphalomesenteric veins.
v.a., vertebral artery.
v.b.sp.n., ventral branch of ventral nerve-root.

v.k.scl.t., ventral division of caudal sclerotomite.
v.lat.m., ventro-lateral mass of occipital.
v.m.r., ventral part of "membrana reuniens."
v.r.at., ventral ring of atlas.
v.sp.n., ventral spinal nerve-root.
ii, iii, iv and v, occipito-spinal nerves corresponding to the 2nd, 3rd, 4th and 5th occipital segments.

PLATE 11.

FIG. 1.—Transverse section through an anterior segment of a 2·0 mm. embryo. × 100.

FIG. 2.—Transverse section through an anterior segment of a 3·0 mm. embryo. × 100.

FIG. 3.—Transverse section through the most anterior segment of a 3·0 mm. embryo. × 95.

FIG. 4.—Parasagittal section through an anterior segment of a 4·0 mm. embryo, showing the division of the sclerotomes. × 100.

FIG. 5.—Parasagittal section through the occipital region (future) of a 4·0 mm. embryo, showing the approximation of the nerves. × 80.

FIG. 6.—Transverse section through the 1st cervical segment of a 4·8 mm. embryo. × 65.

PLATE 12.

FIG. 7.—Frontal section through the future thoracic region of a 5·5 mm. embryo, showing the intra-sclerotomic fissure. × 110.

FIG. 8.—Frontal section through the thoracic region of a 6·5 mm. embryo, showing the development of the perichordal disc. × 100.

FIG. 9.—Transverse section through the middle of a thoracic segment in a 7·2 mm. embryo. × 65.

FIG. 10.—Transverse section through the anterior part of a cervical segment in a 7·2 mm. embryo. × 65.

FIG. 11.—Transverse section through the pro-atlas of a 7·2 mm. embryo ; note the cranio-vertebral interval. × 65.

FIG. 12.—Frontal section through the thoracic region of a 8·0 mm. embryo. × 70.

PLATE 13.

FIG. 13.—Sagittal section through the anterior cervical region of an 8·0 mm. embryo. × 100.

FIG. 14.—Parasagittal section through the anterior cervical region of a 9·0 mm. embryo. × 50.

FIG. 15.—Transverse section through the pro-atlas of a 9·0 mm. embryo. × 60.

FIG. 16.—Parasagittal section through the anterior cervical region of a 9·0 mm. embryo. × 75.

FIG. 17.—Parasagittal section through the anterior cervical region of a 12·5 mm. embryo. × 58.

FIG. 18.—Frontal section through the thoracic region of a 12·5 mm. embryo. Note the enormous dilatation of the notochord. × 60.

PLATE 14.

FIG. 19.—Section, from a transverse series, passing through the anterior portion of the atlas basidorsal of a 12·5 mm. embryo. On the right is seen the tissue enclosing the loop of the vertebral artery and also the condylar part of the occipital rudiment. × 45.

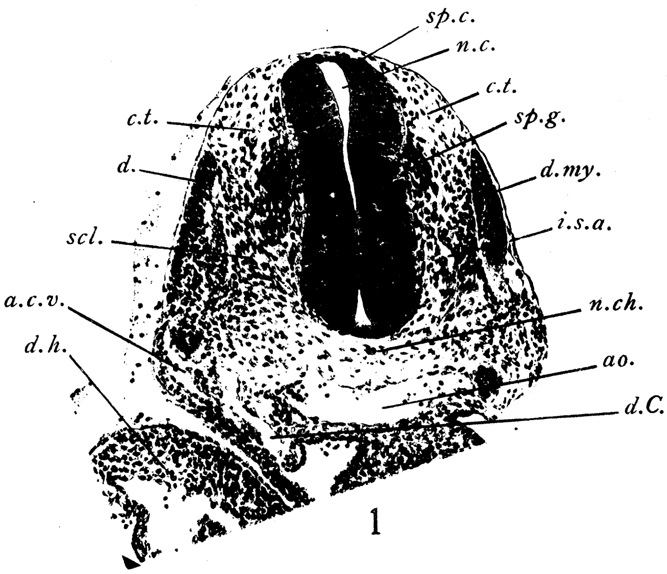
FIG. 20.—Sagittal section through the anterior cervical region of a 12·5 mm. embryo. Again, note the notochordal dilatation. × 65.

FIG. 21.—Transverse section through the thoracic region of a 14·0 mm. embryo. Showing the bone centre in the angle of a rib. × 45.

FIG. 22.—Frontal section through the cervical region of a 14·0 mm. embryo. × 34.

FIG. 23.—Parasagittal section through the base of a neural arch in the newly born animal (23·0 mm.). × 44.

FIG. 24.—Sagittal section through the thoracic centra of the newly born animal, showing the nucleus pulposus and bone centres. Note the unbroken notochordal sheath remnant. × 58.



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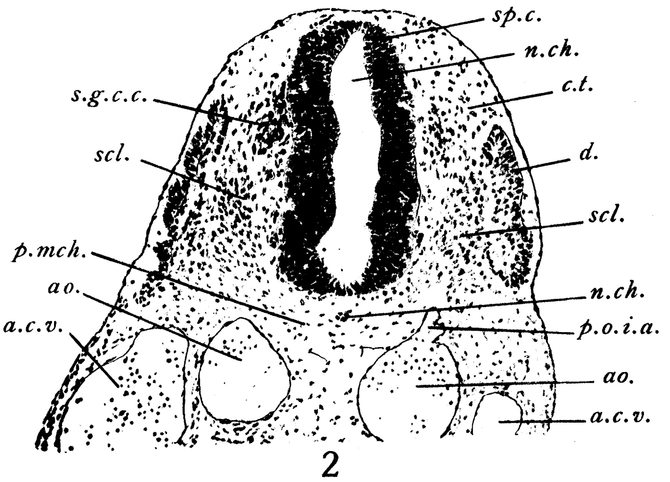

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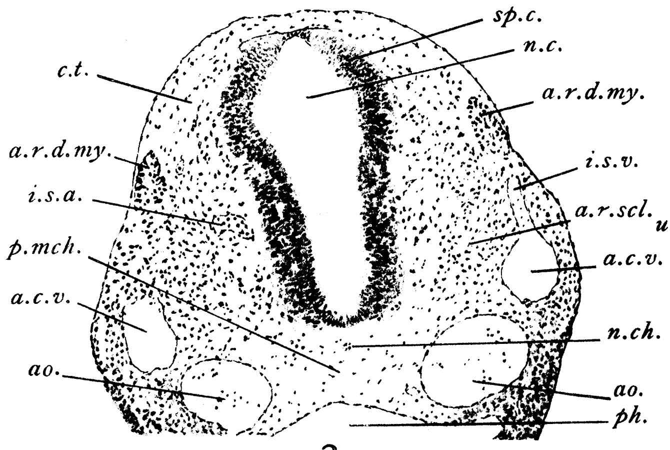

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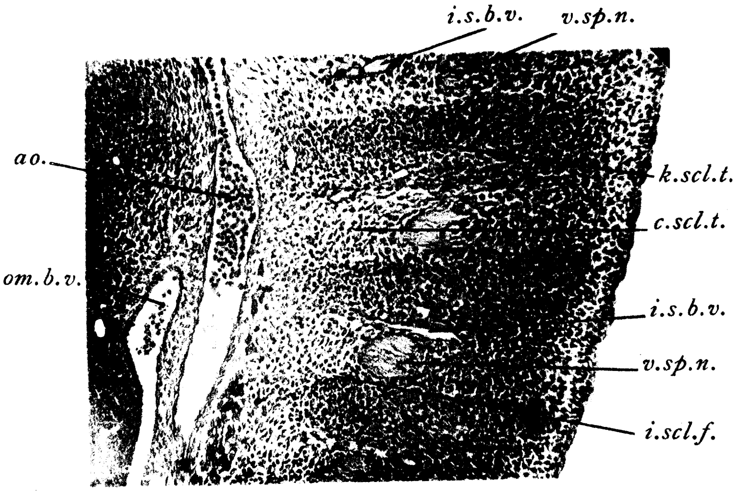

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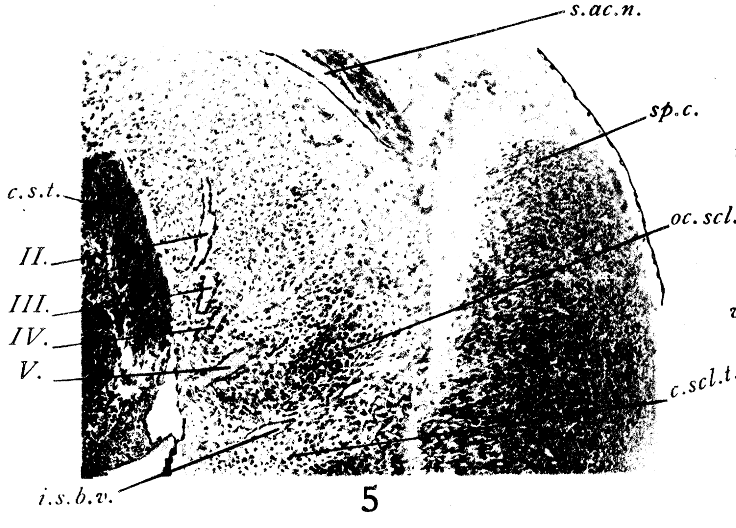

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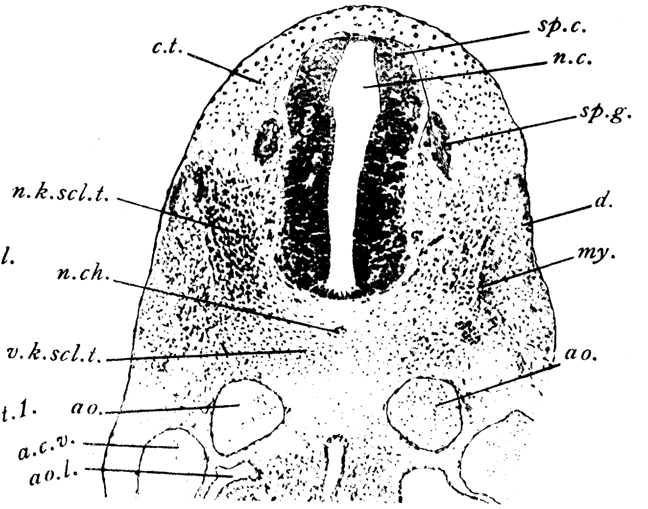

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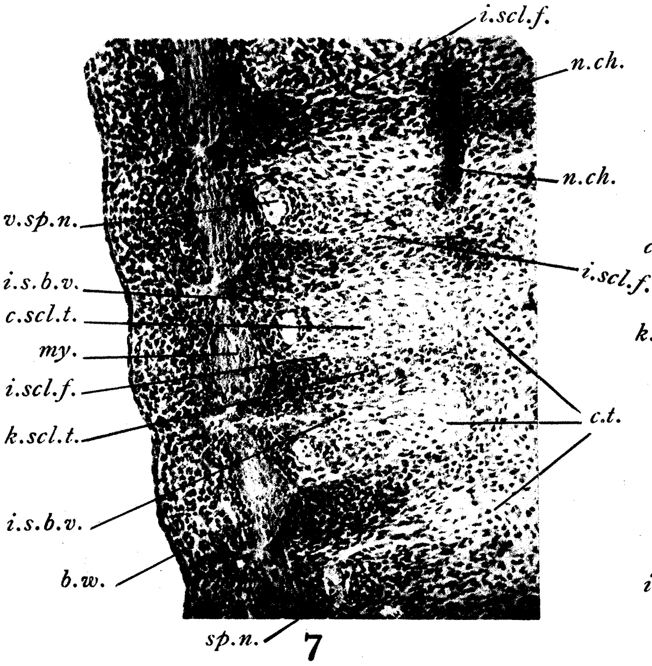

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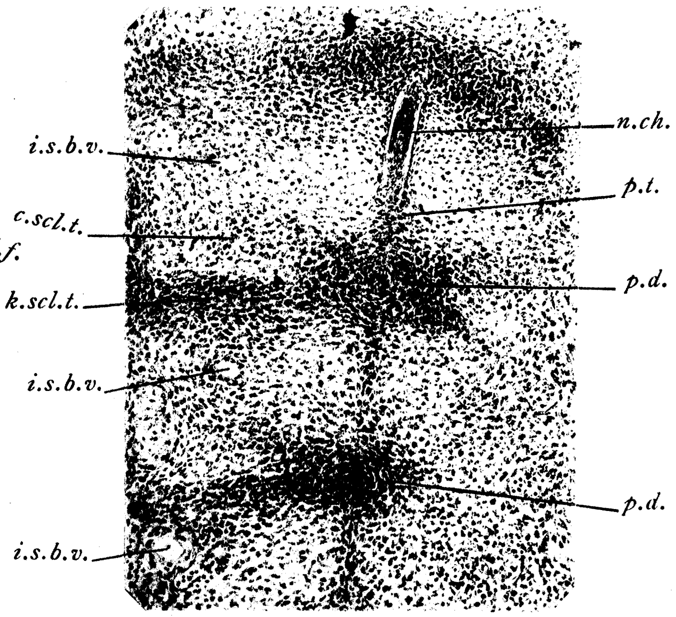

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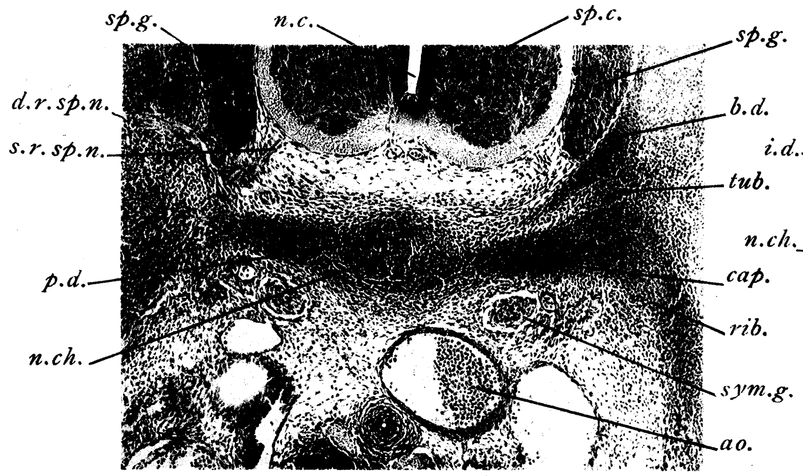

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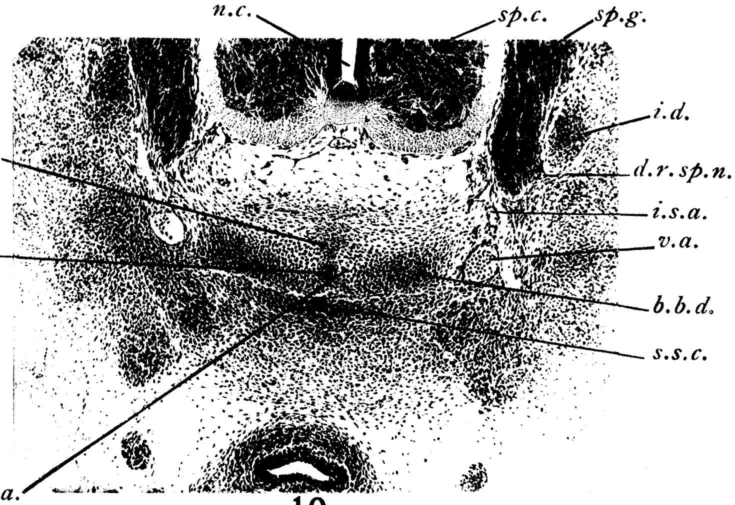

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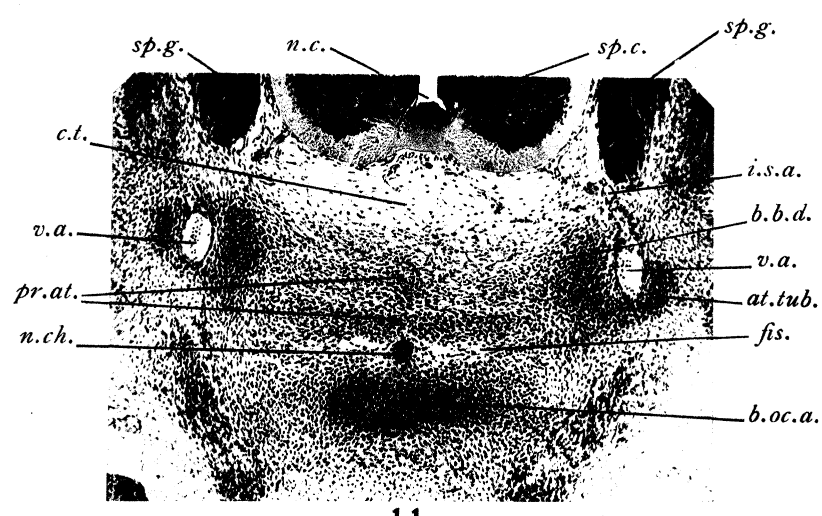

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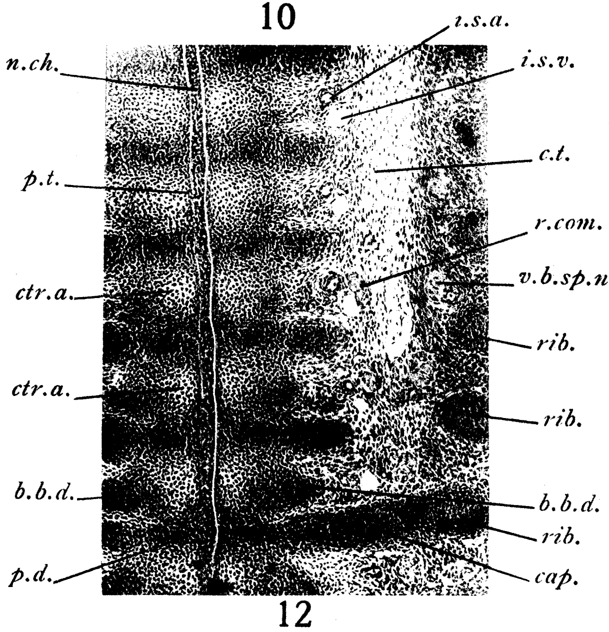

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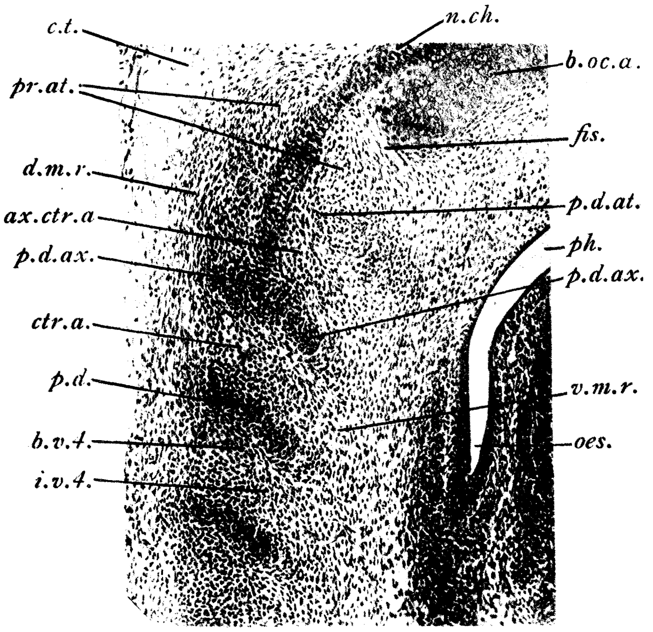

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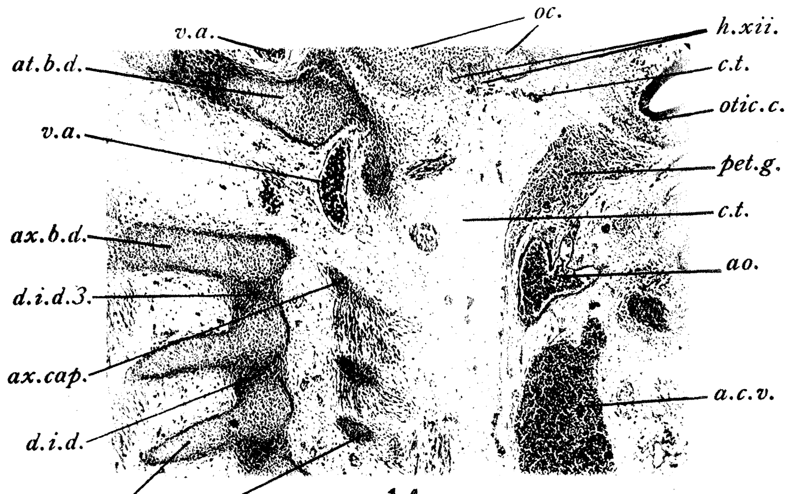

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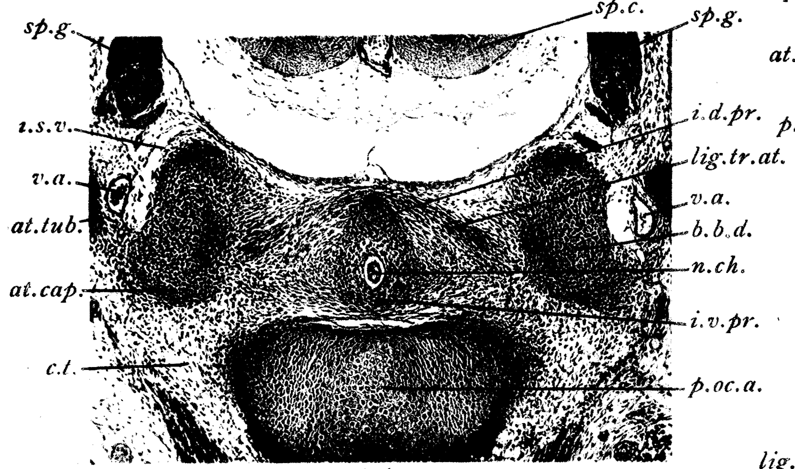

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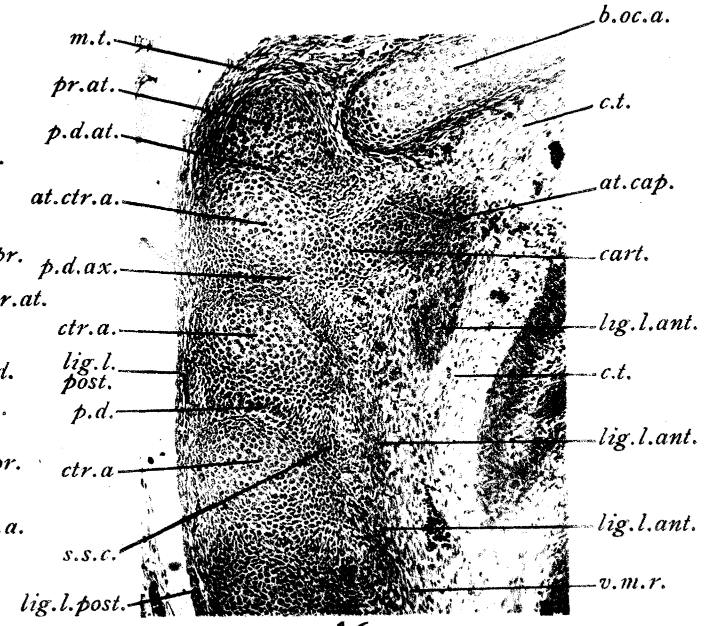

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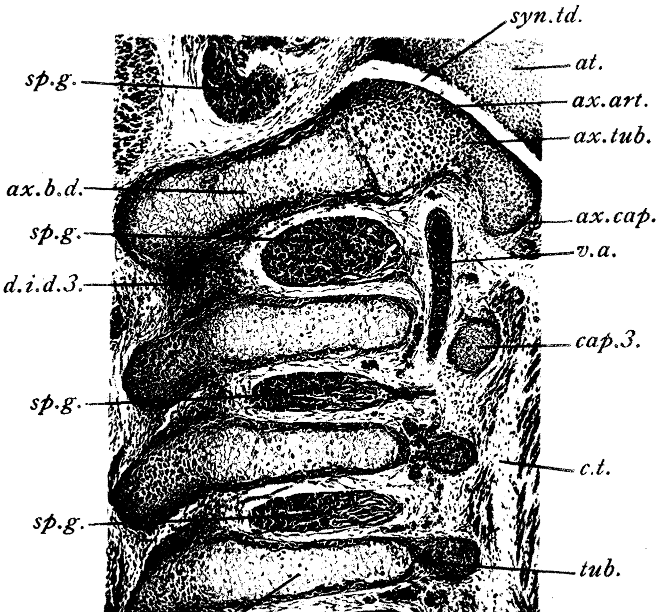

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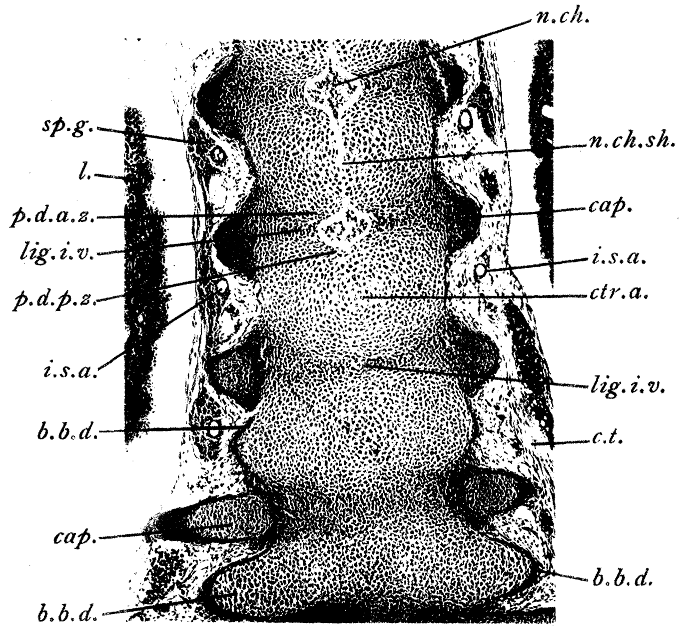

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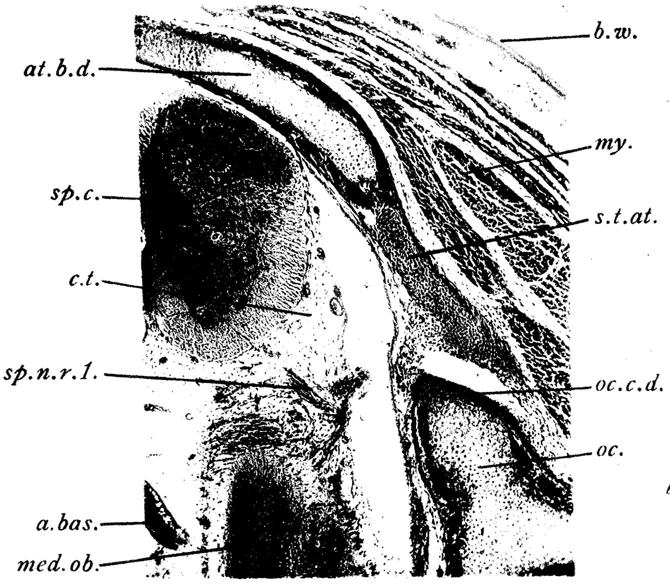

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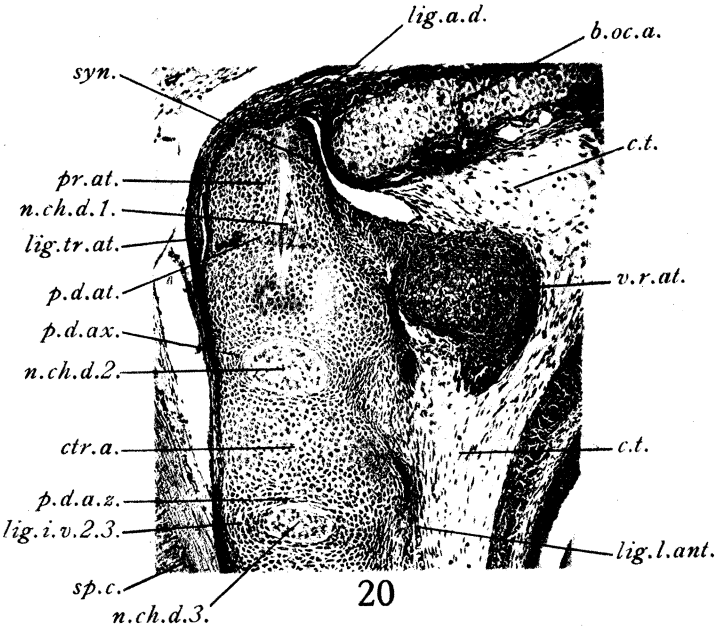

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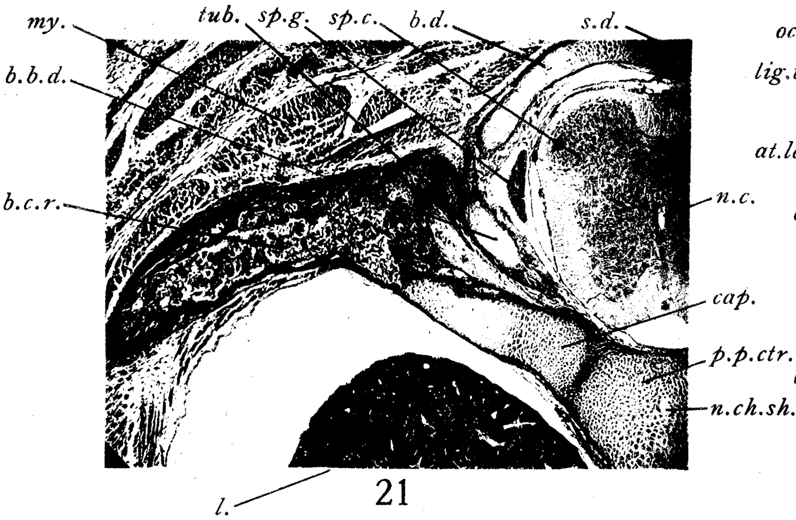

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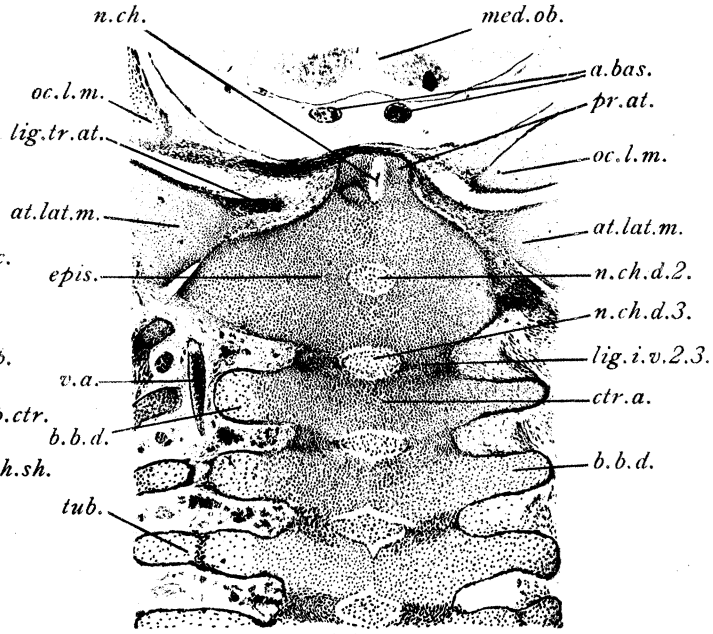

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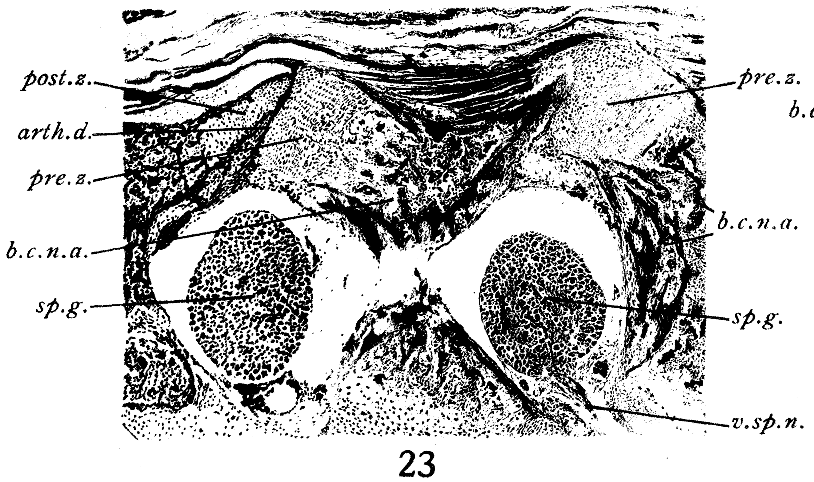

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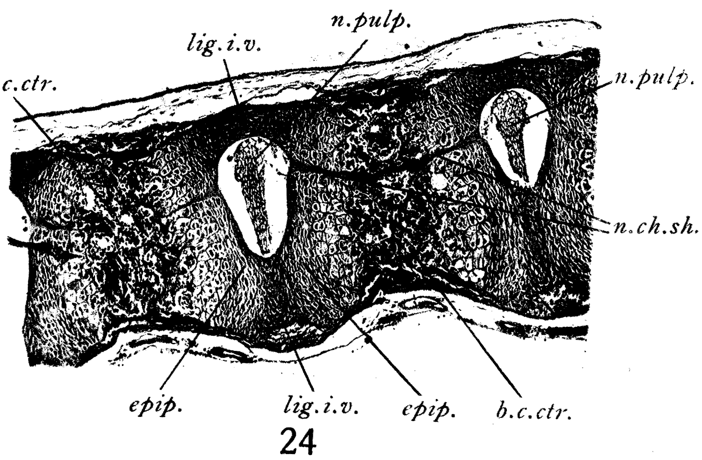

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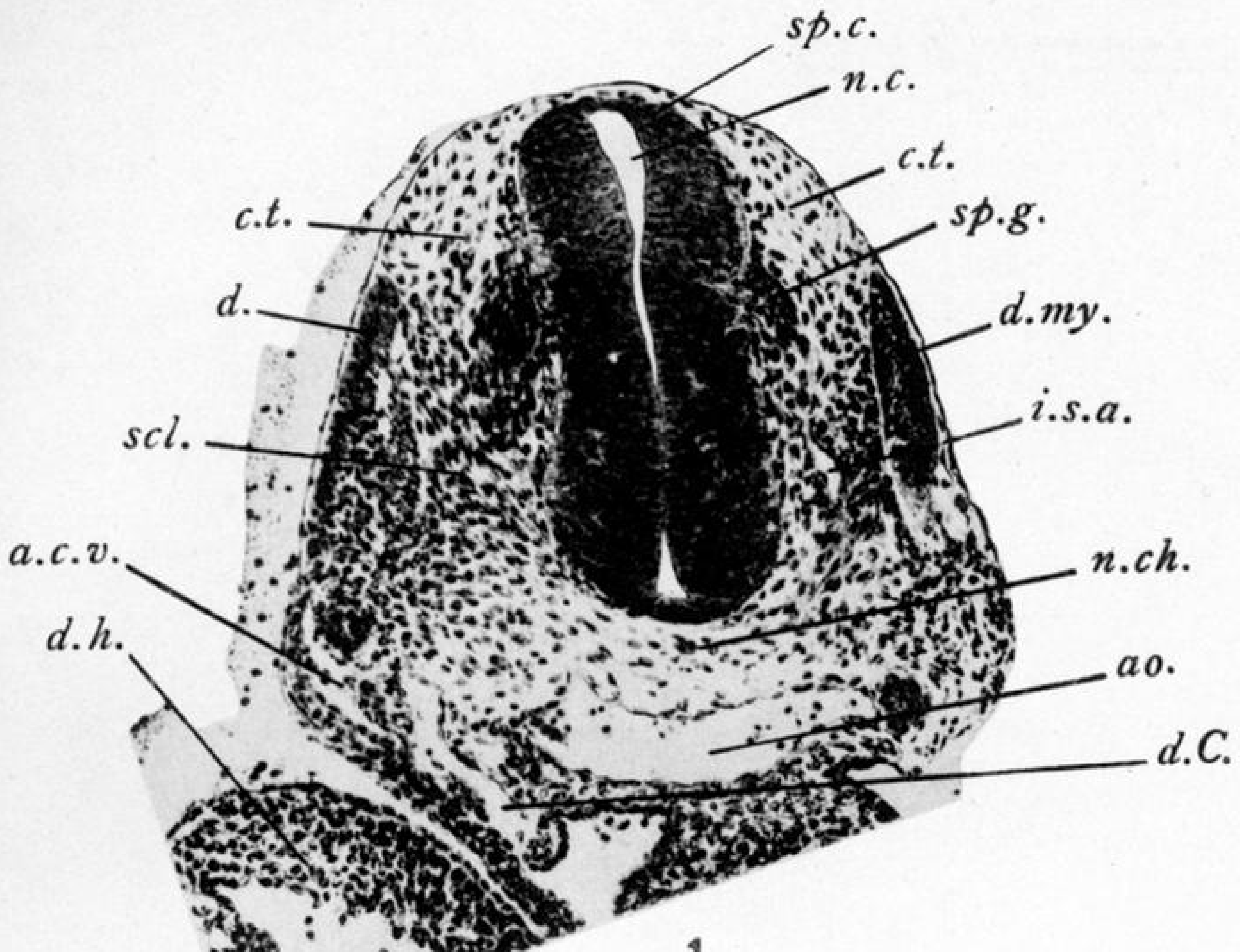

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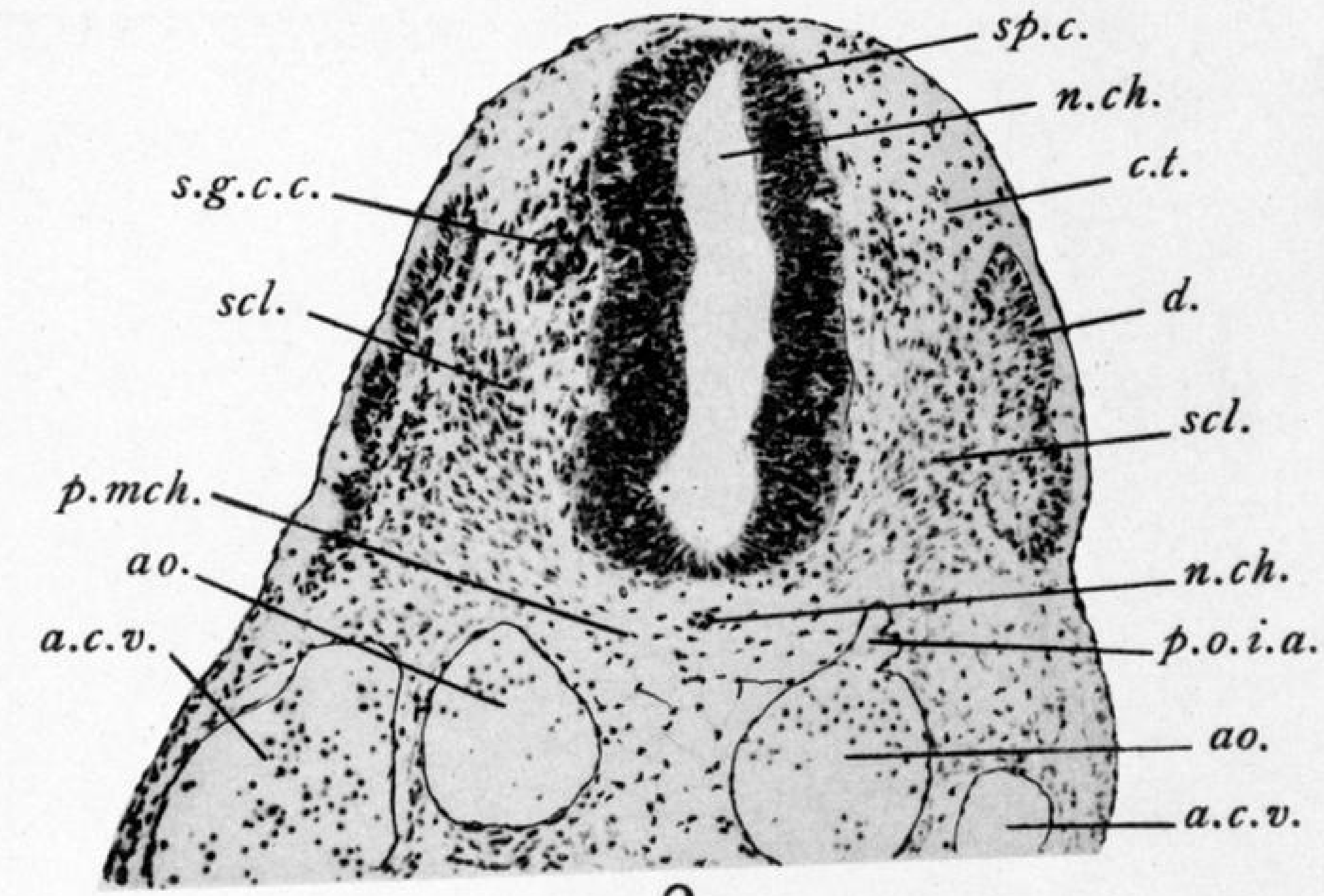

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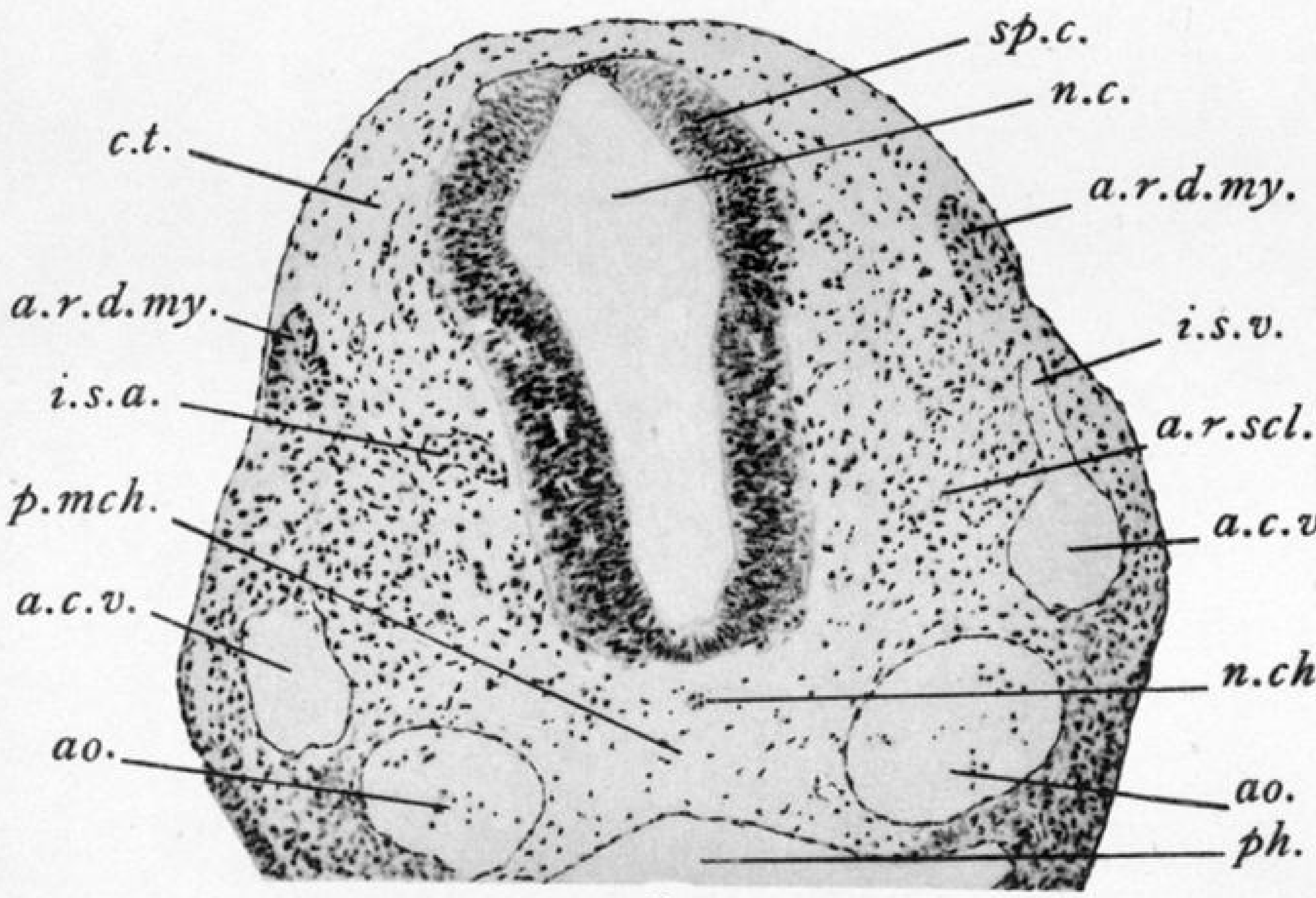

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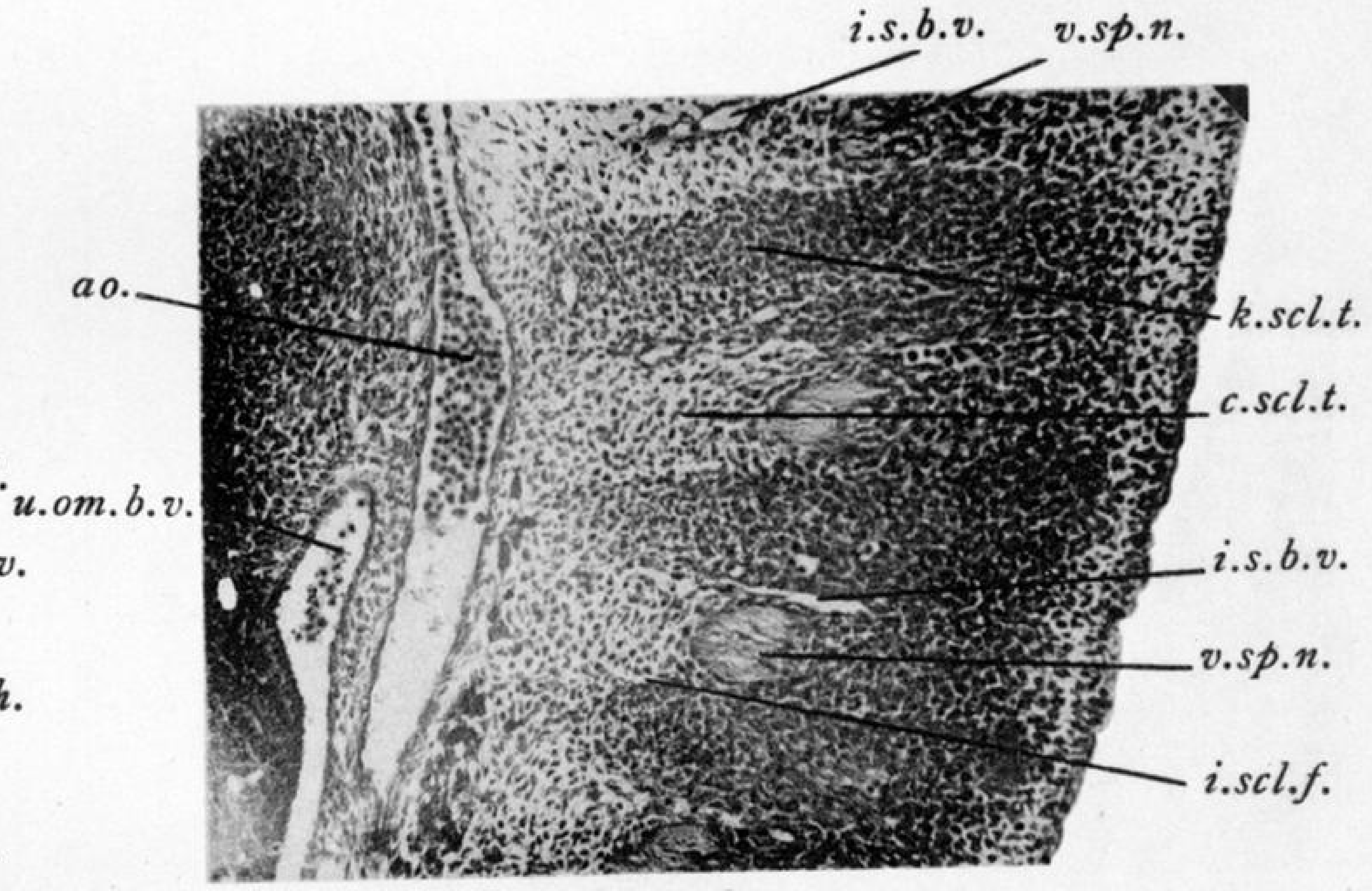

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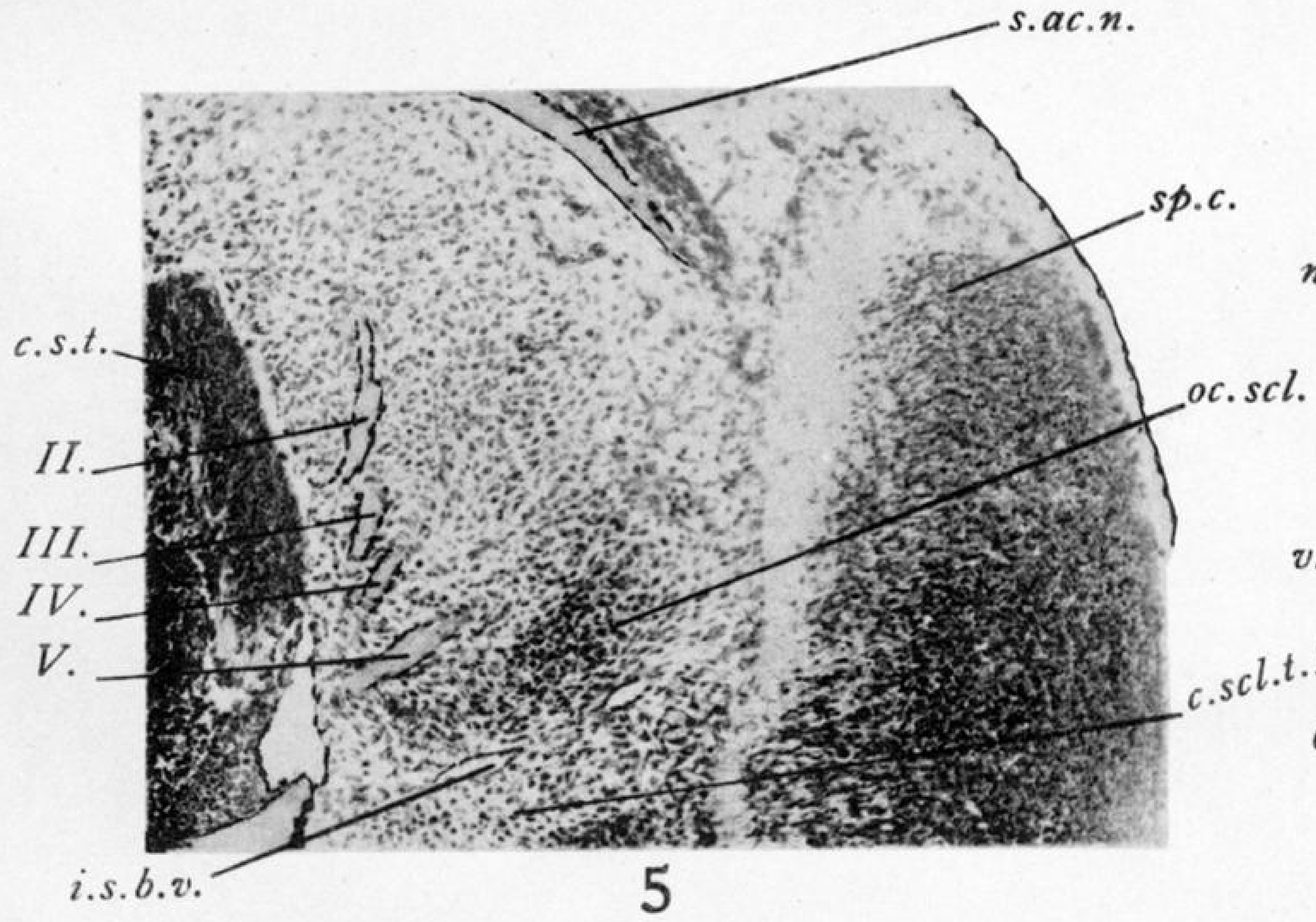

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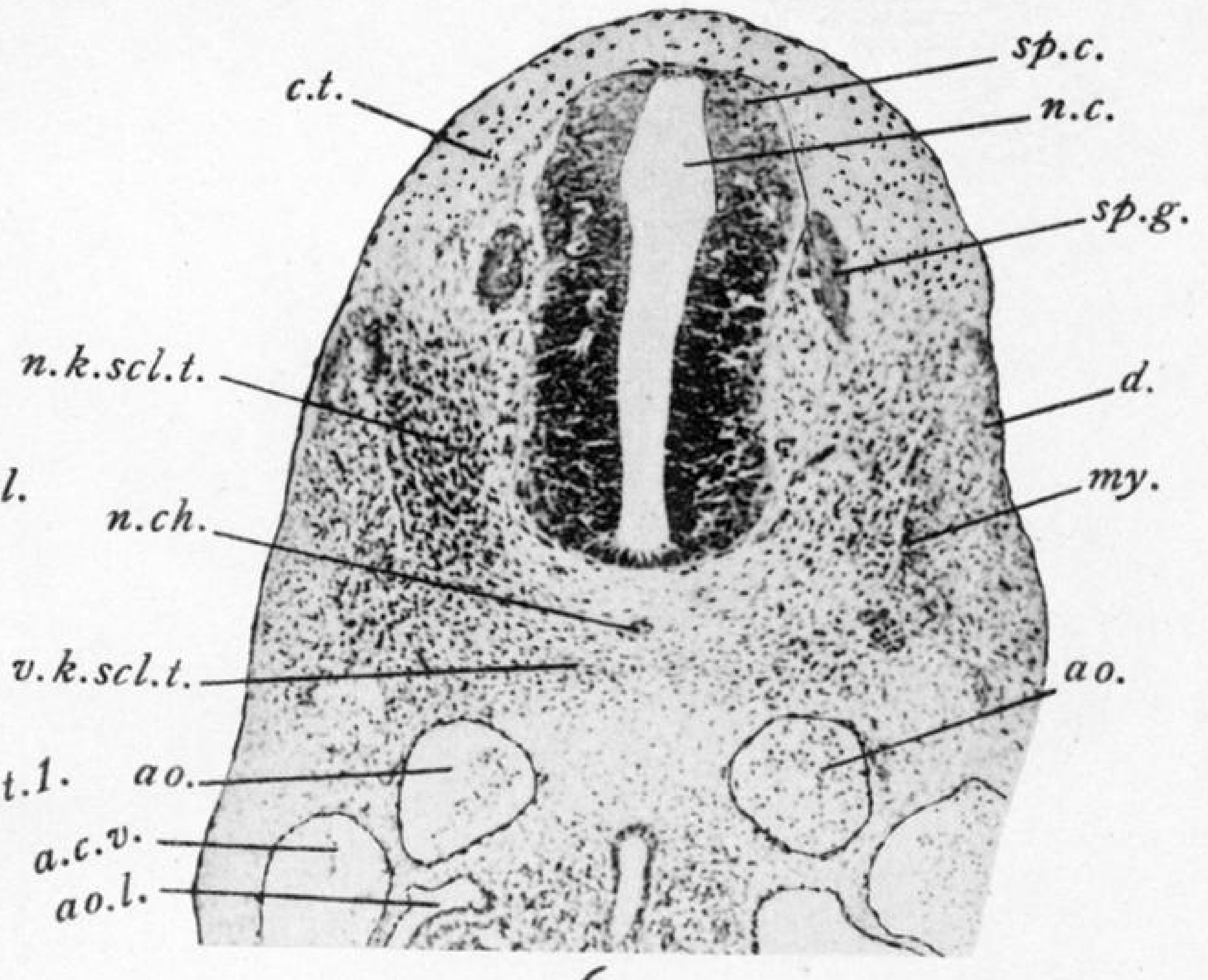

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PLATE 11.

FIG. 1.—Transverse section through an anterior segment of a 2·0 mm. embryo. × 100.

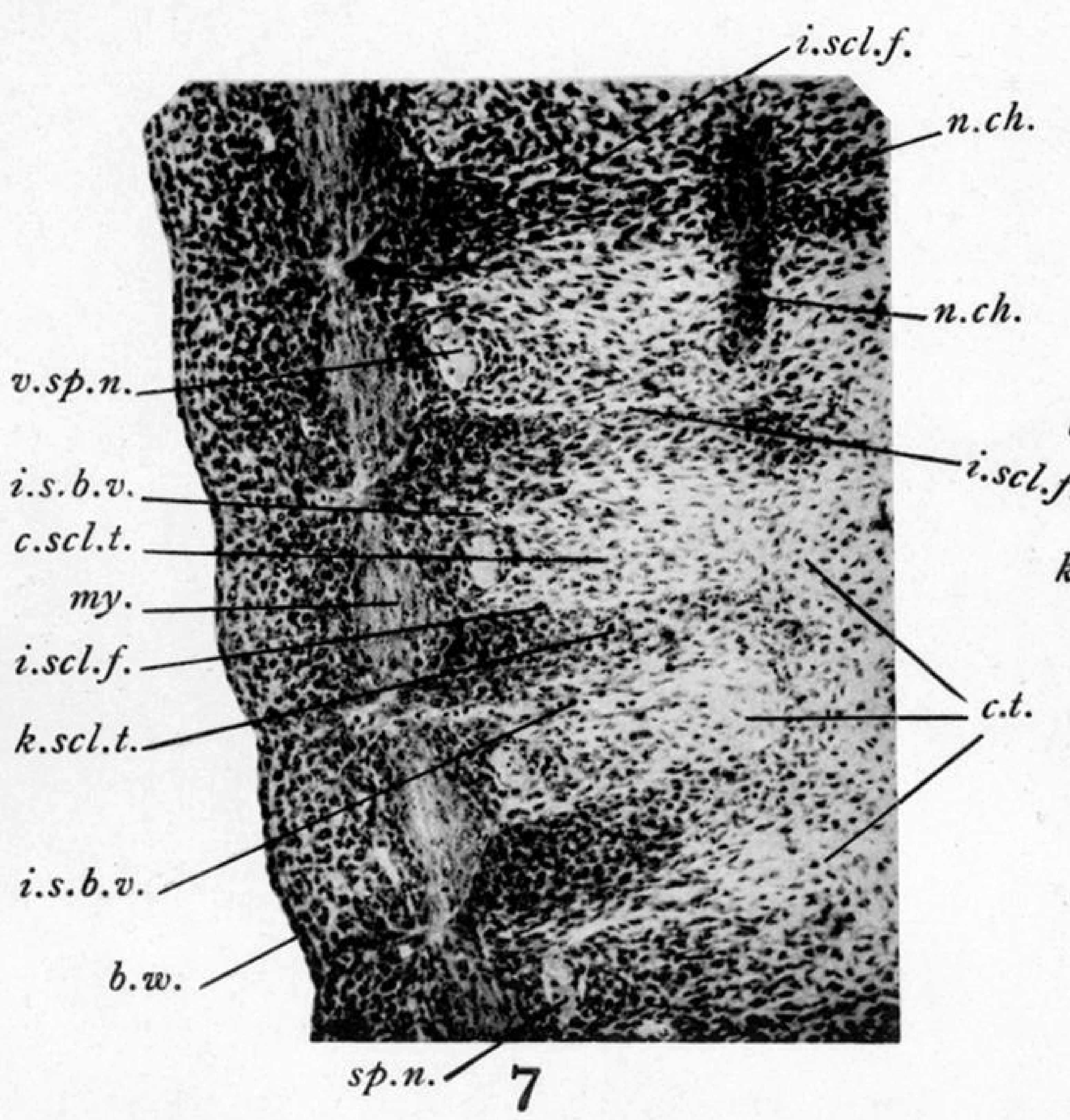
FIG. 2.—Transverse section through an anterior segment of a 3·0 mm. embryo. × 100.

FIG. 3.—Transverse section through the most anterior segment of a 3·0 mm. embryo. × 95.

FIG. 4.—Parasagittal section through an anterior segment of a 4·0 mm. embryo, showing the division of the sclerotomes. × 100.

FIG. 5.—Parasagittal section through the occipital region (future) of a 4·0 mm. embryo, showing the approximation of the nerves. × 80.

FIG. 6.—Transverse section through the 1st cervical segment of a 4·8 mm. embryo. × 65.



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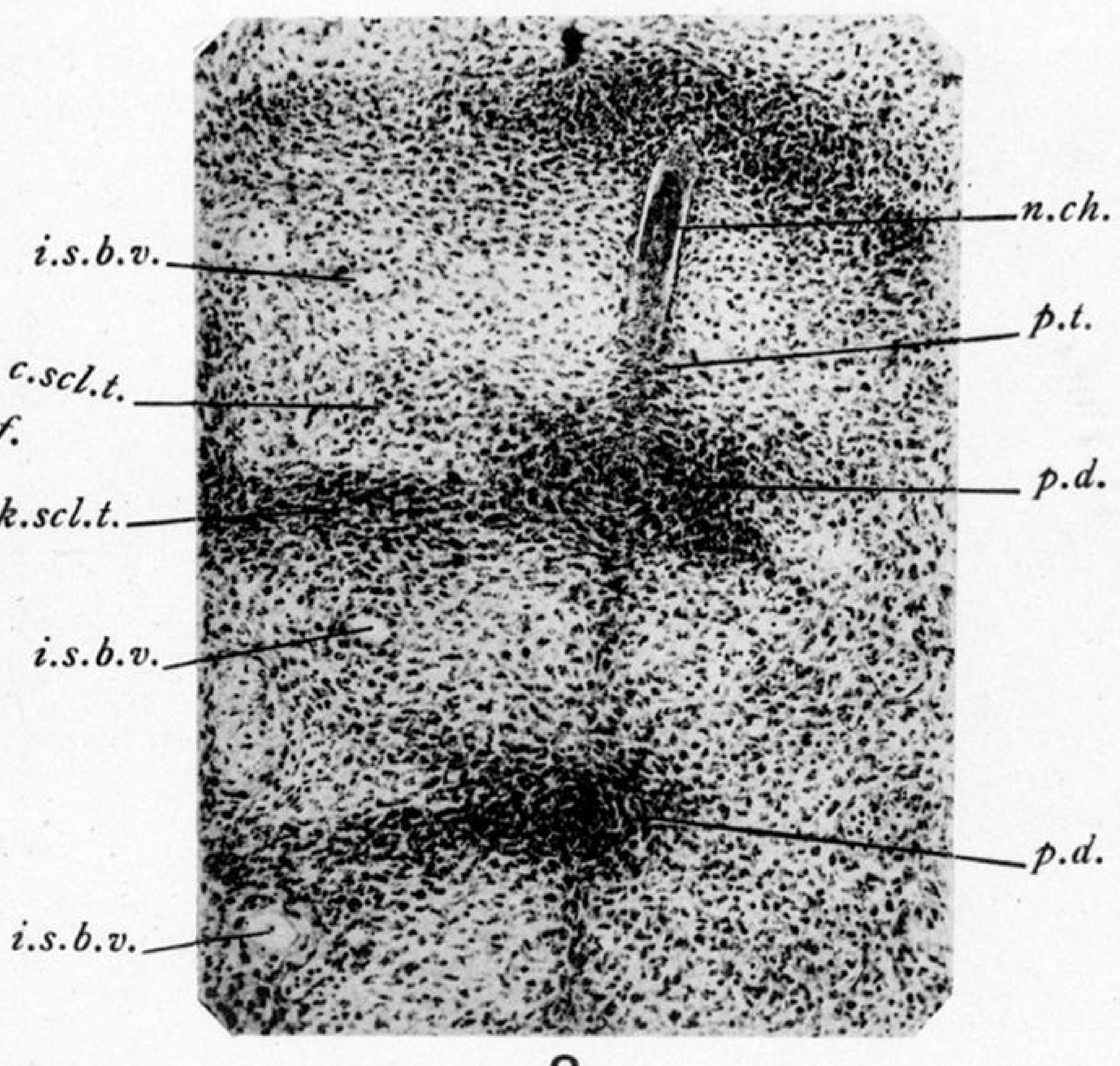

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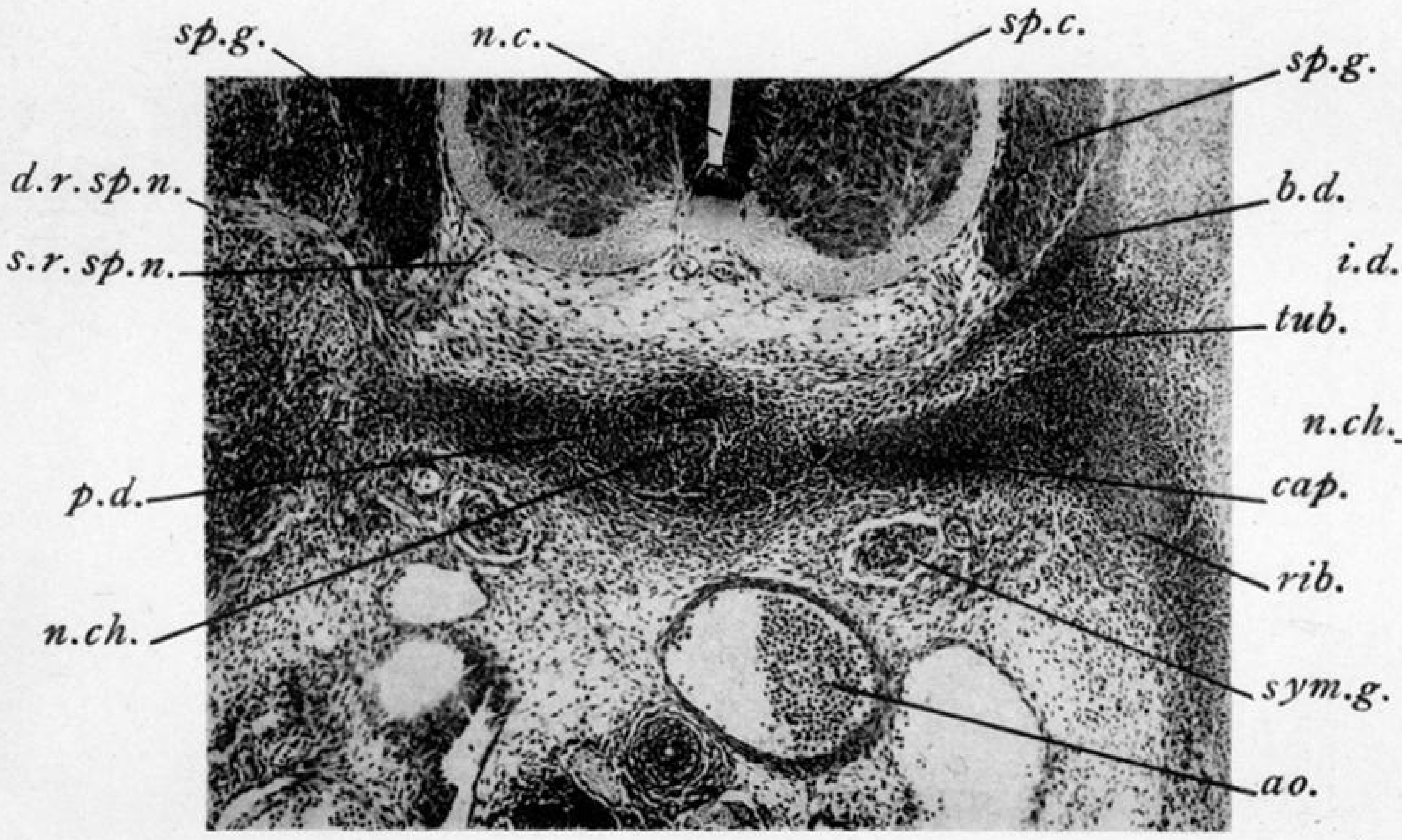

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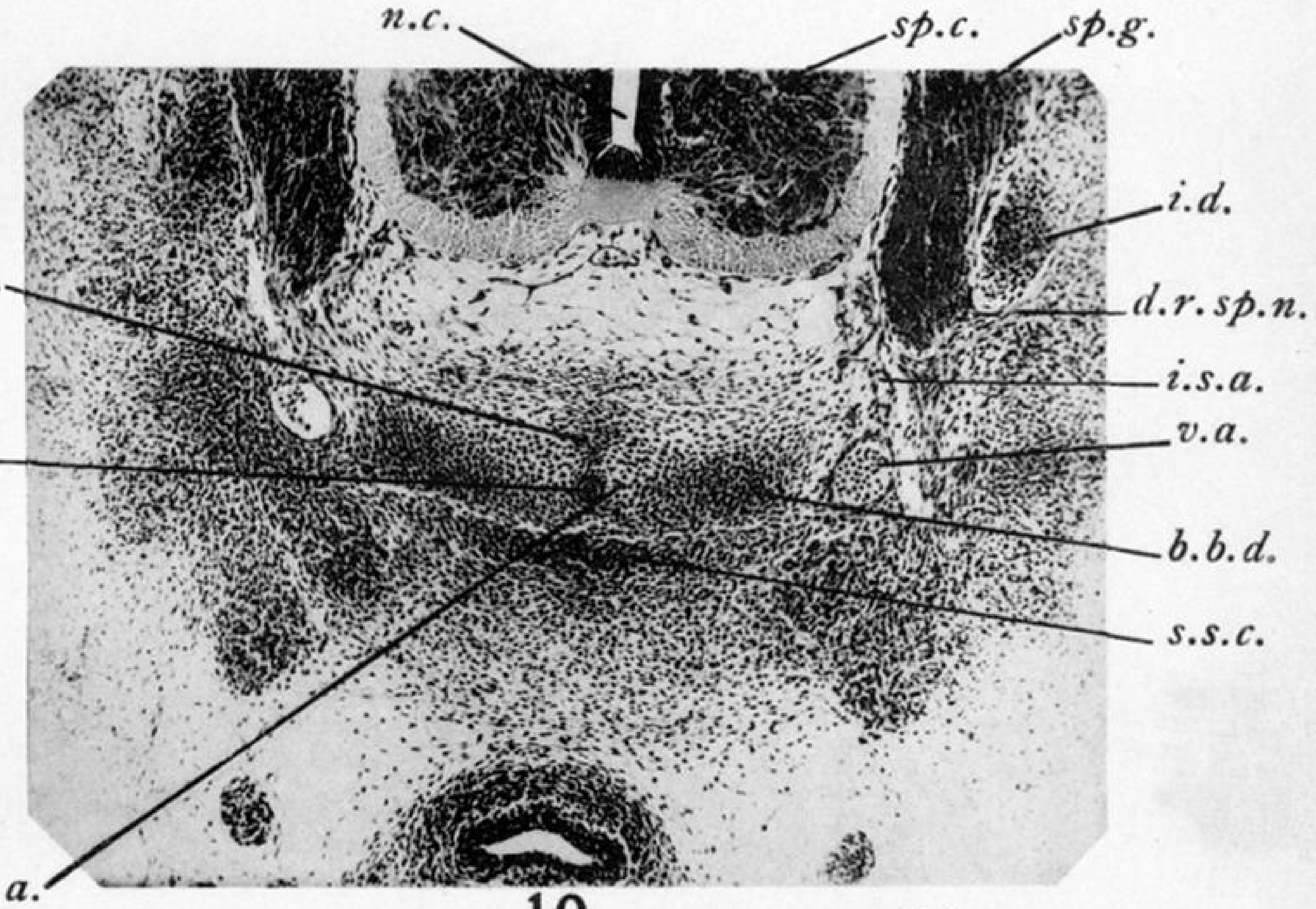

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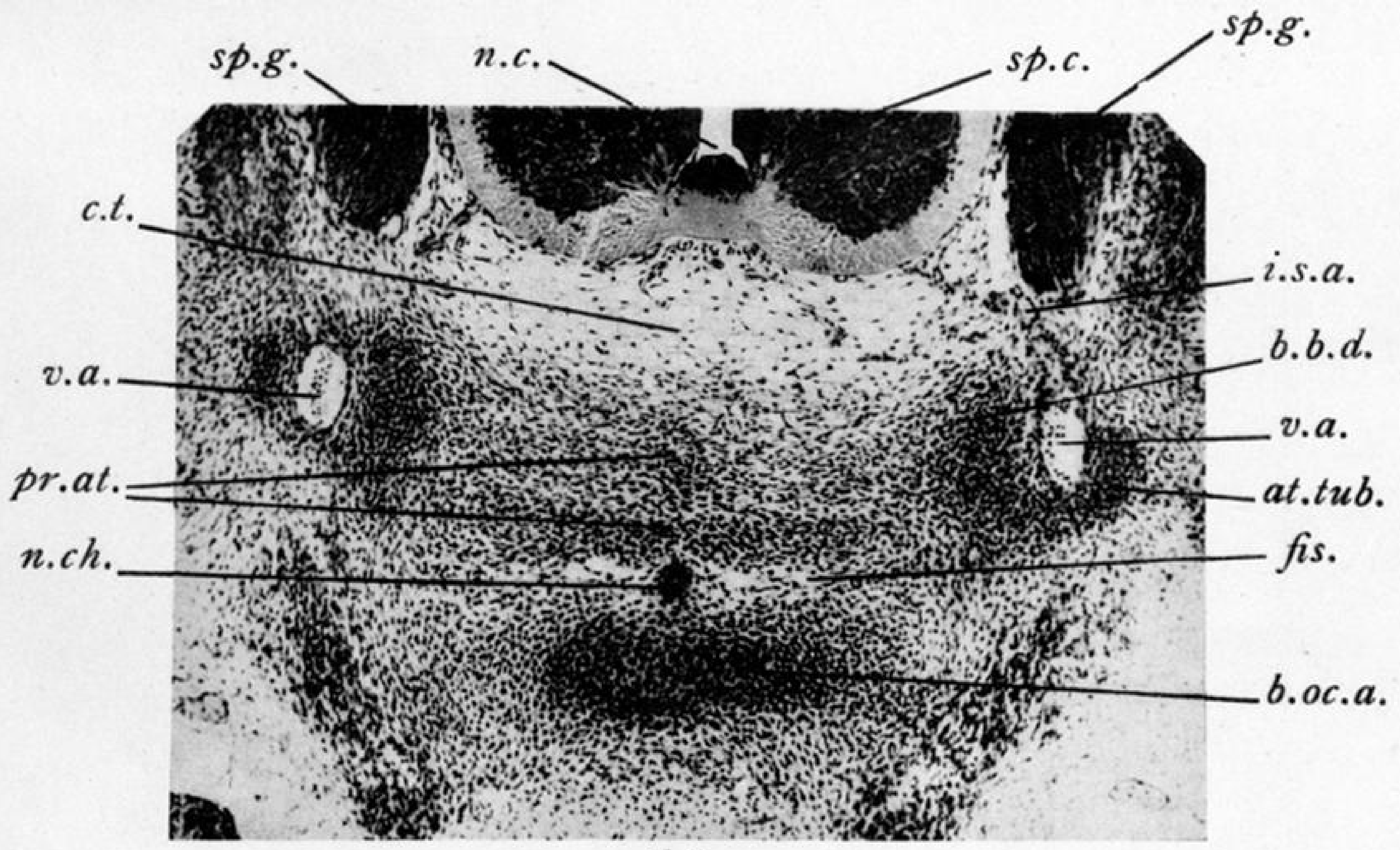

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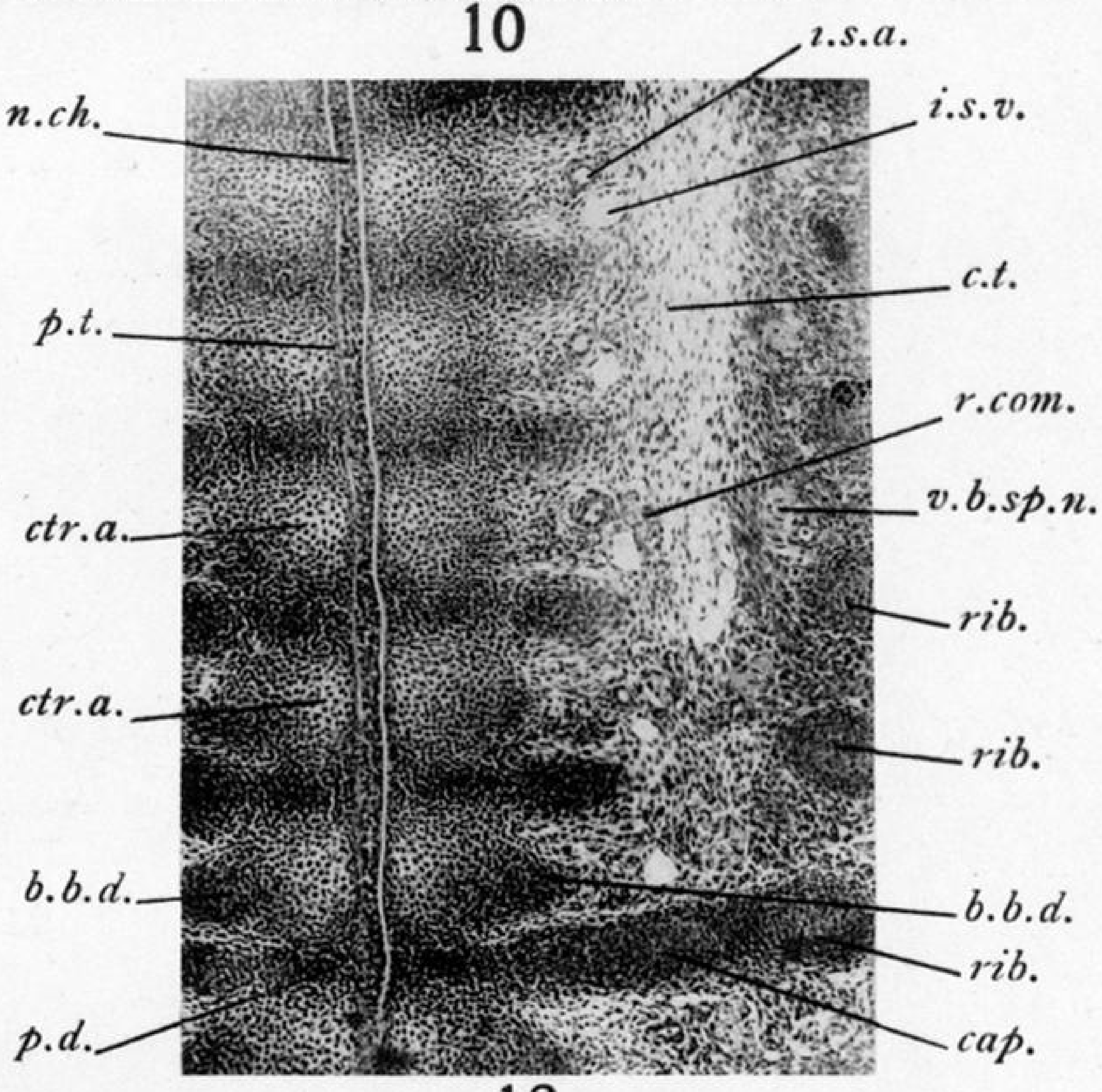

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PLATE 12.

FIG. 7.—Frontal section through the future thoracic region of a 5·5 mm. embryo, showing the intra-sclerotomic fissure. × 110.

FIG. 8.—Frontal section through the thoracic region of a 6·5 mm. embryo, showing the development of the perichordal disc. × 100.

FIG. 9.—Transverse section through the middle of a thoracic segment in a 7·2 mm. embryo. × 65.

FIG. 10.—Transverse section through the anterior part of a cervical segment in a 7·2 mm. embryo. × 65.

FIG. 11.—Transverse section through the pro-atlas of a 7·2 mm. embryo; note the cranio-vertebral interval. × 65.

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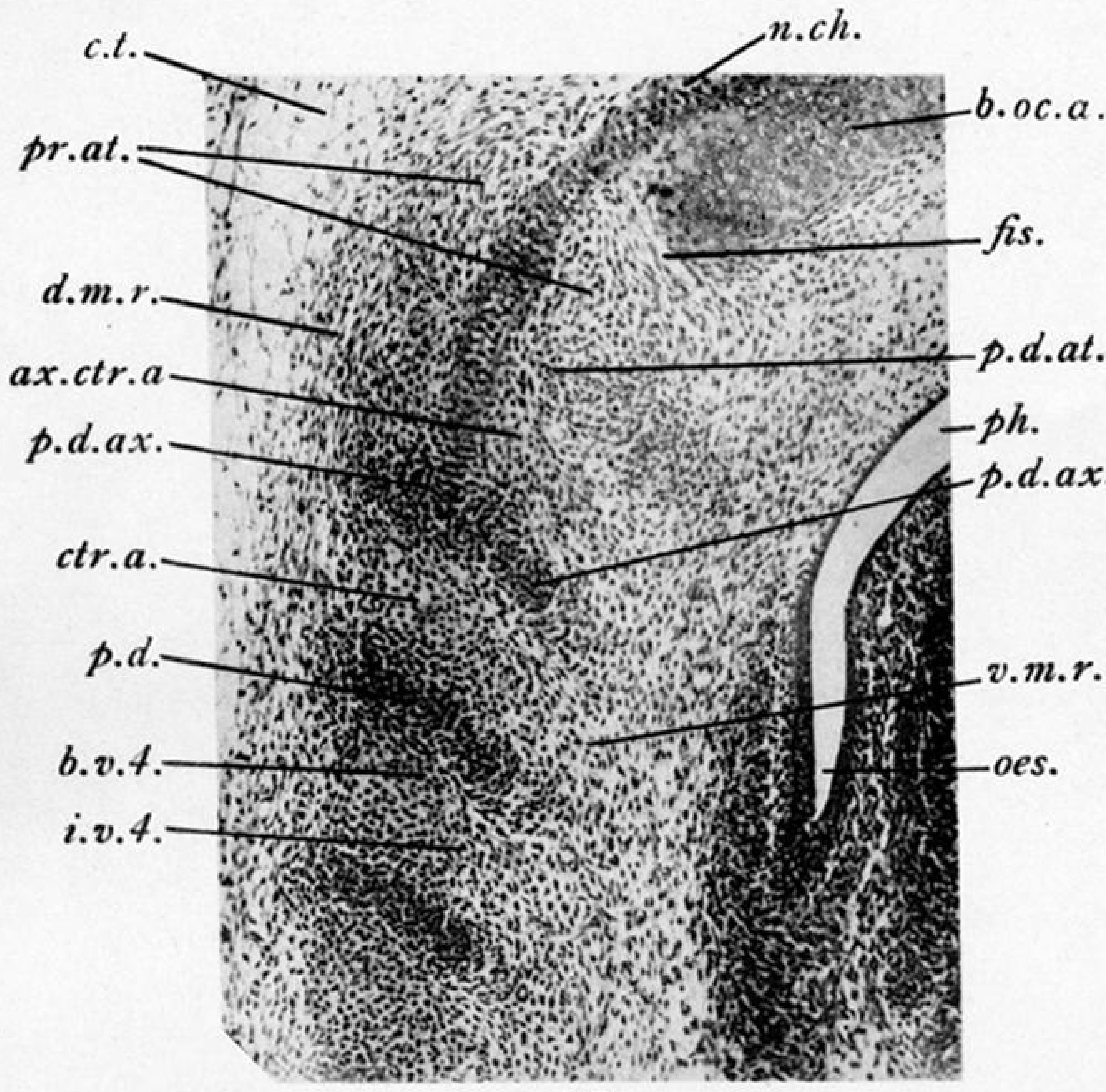

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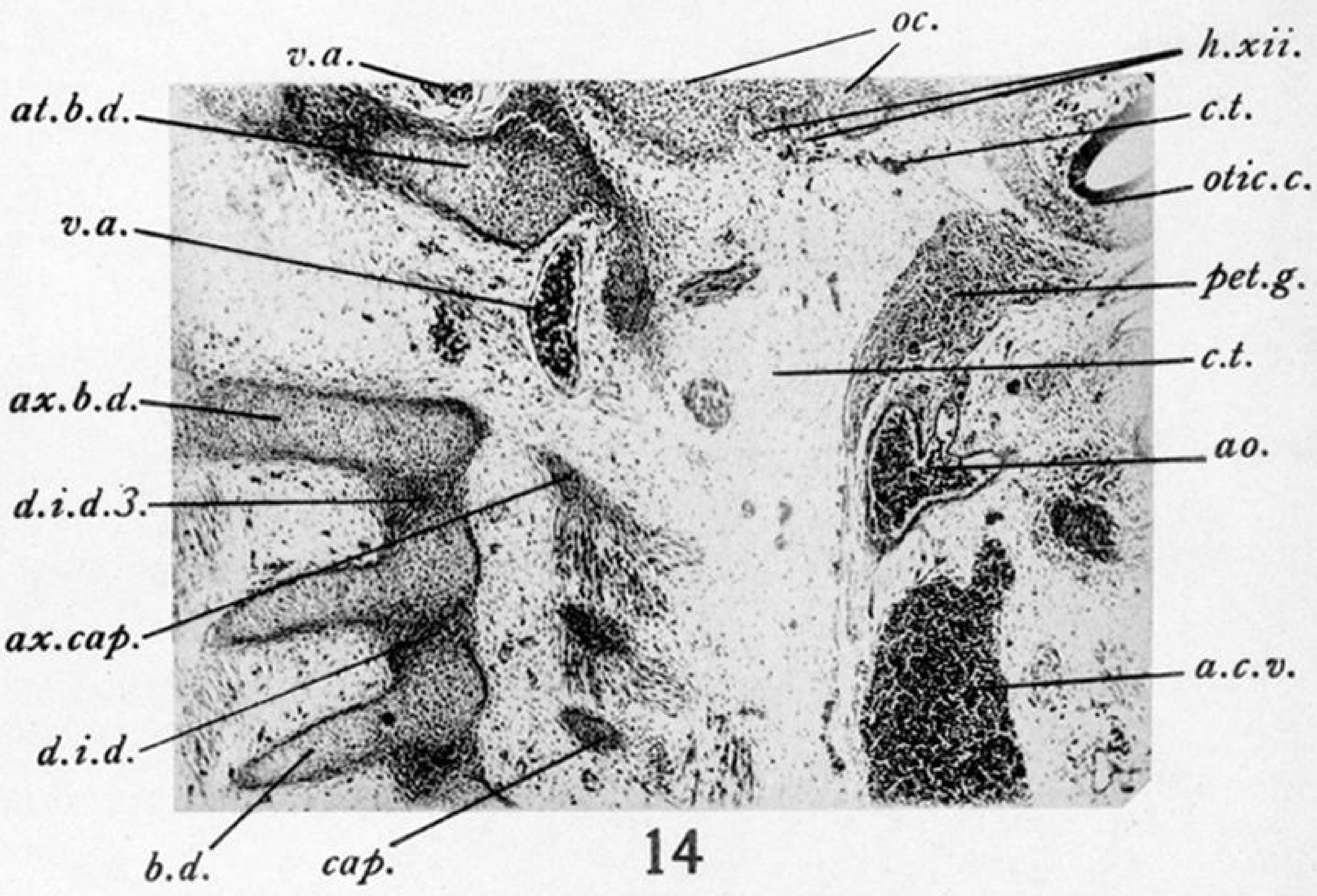

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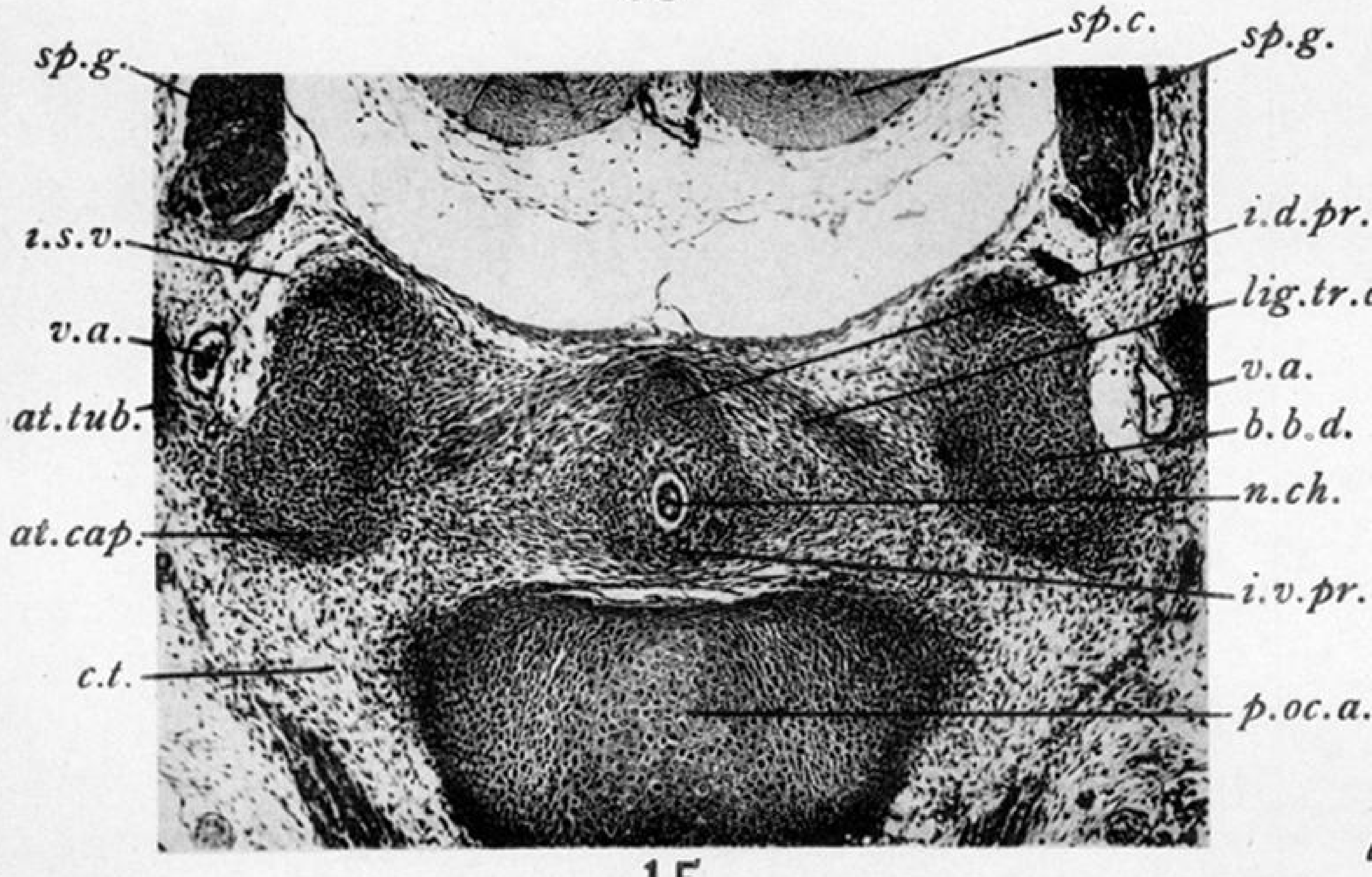

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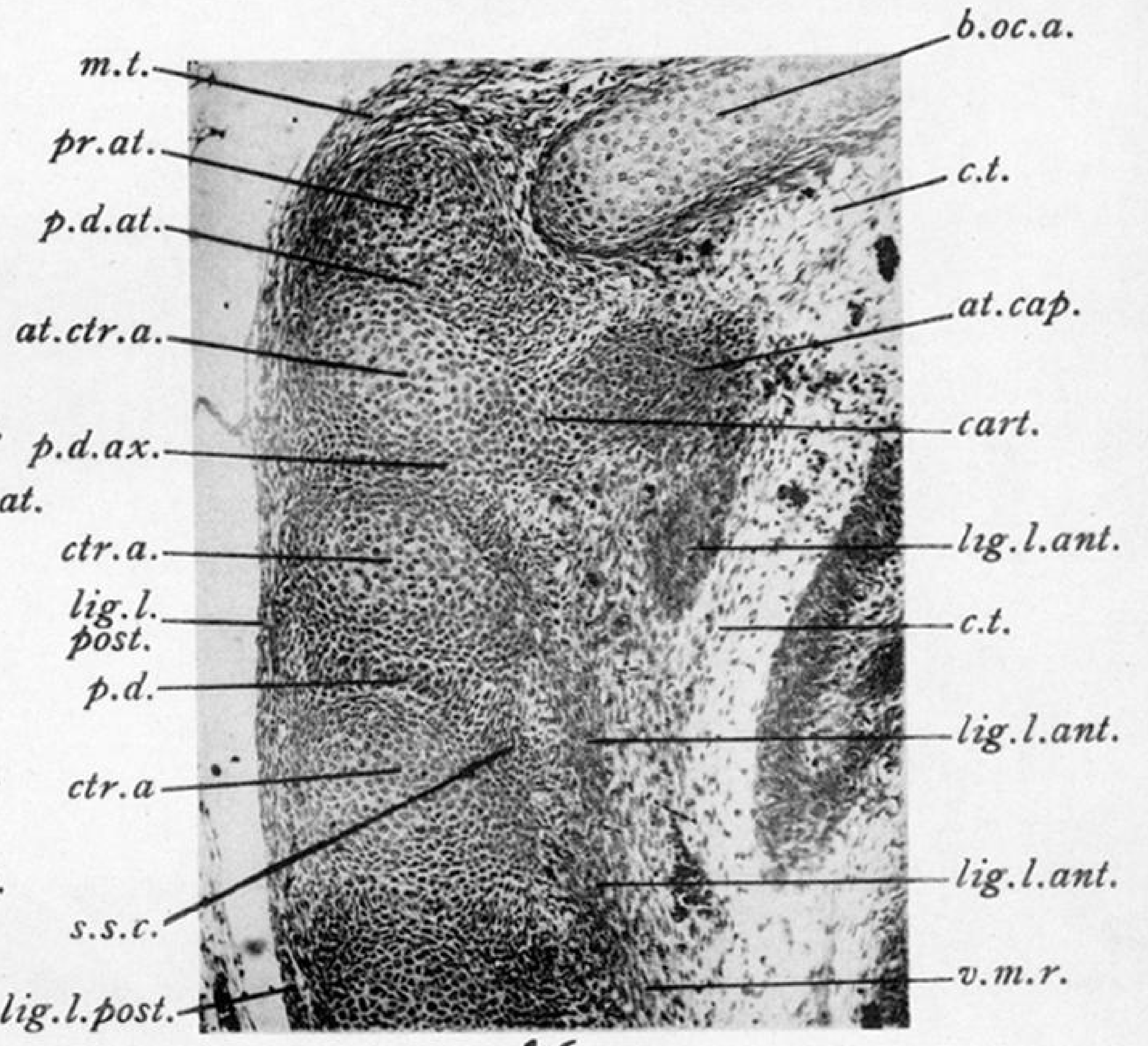

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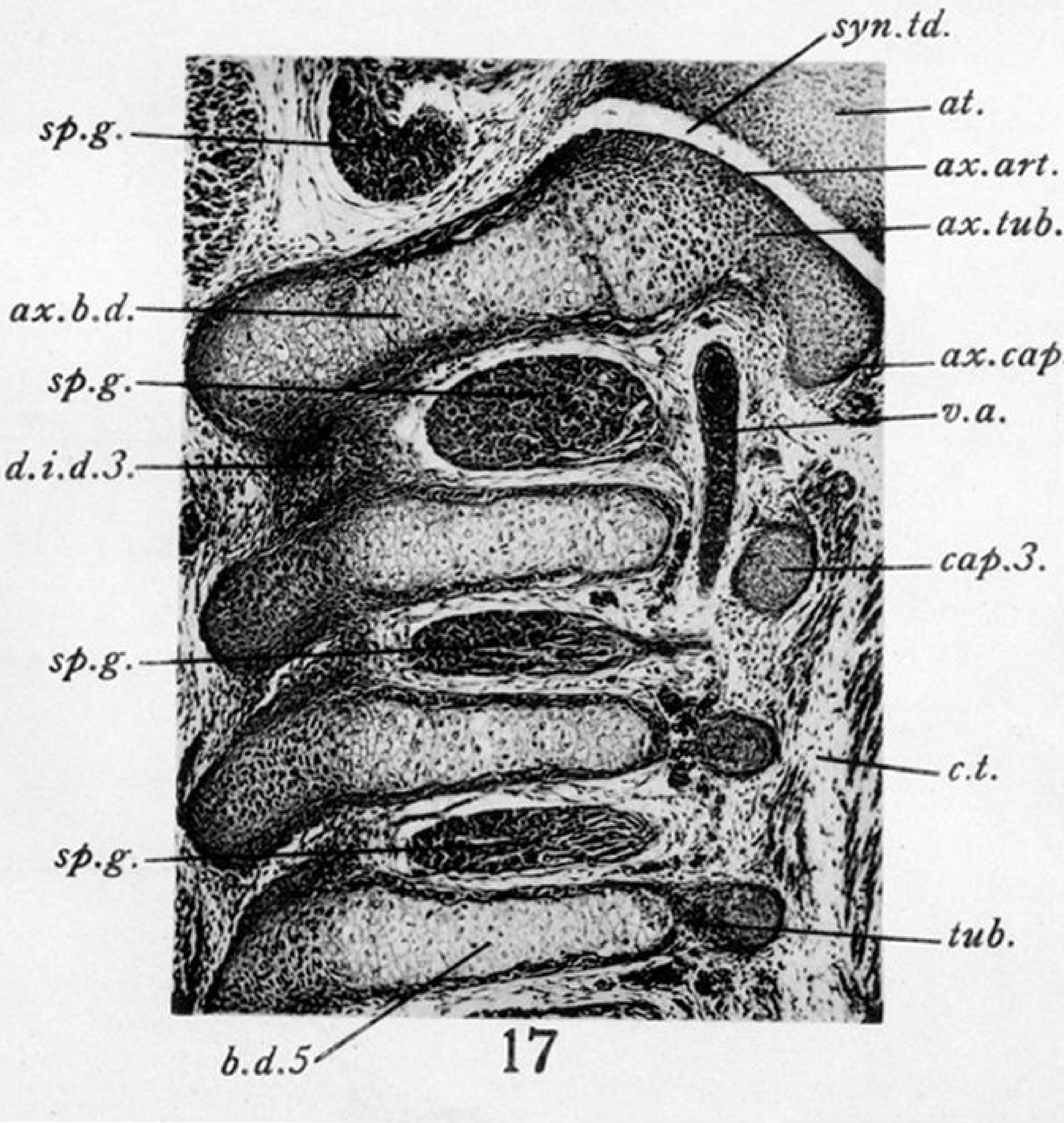

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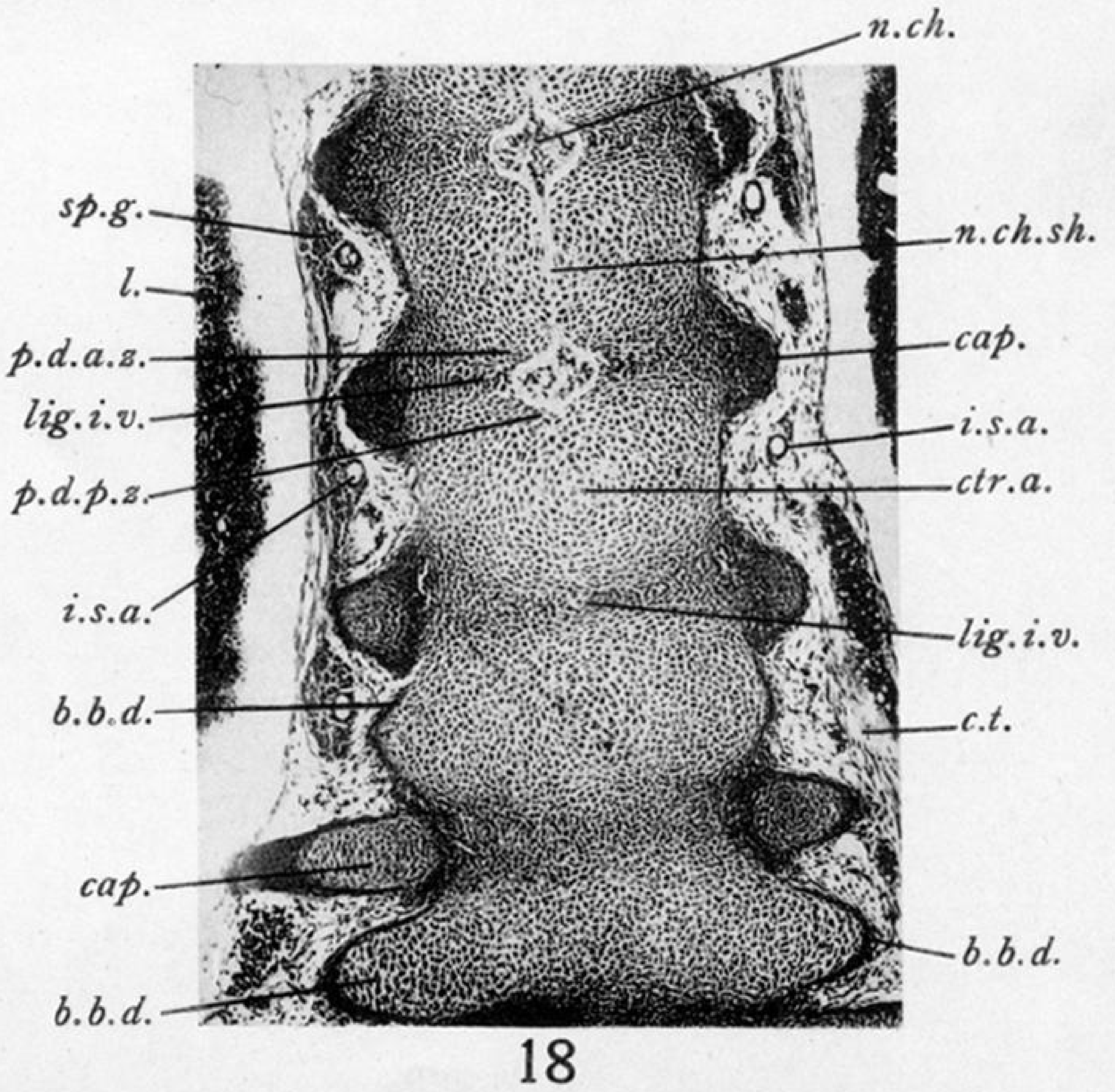

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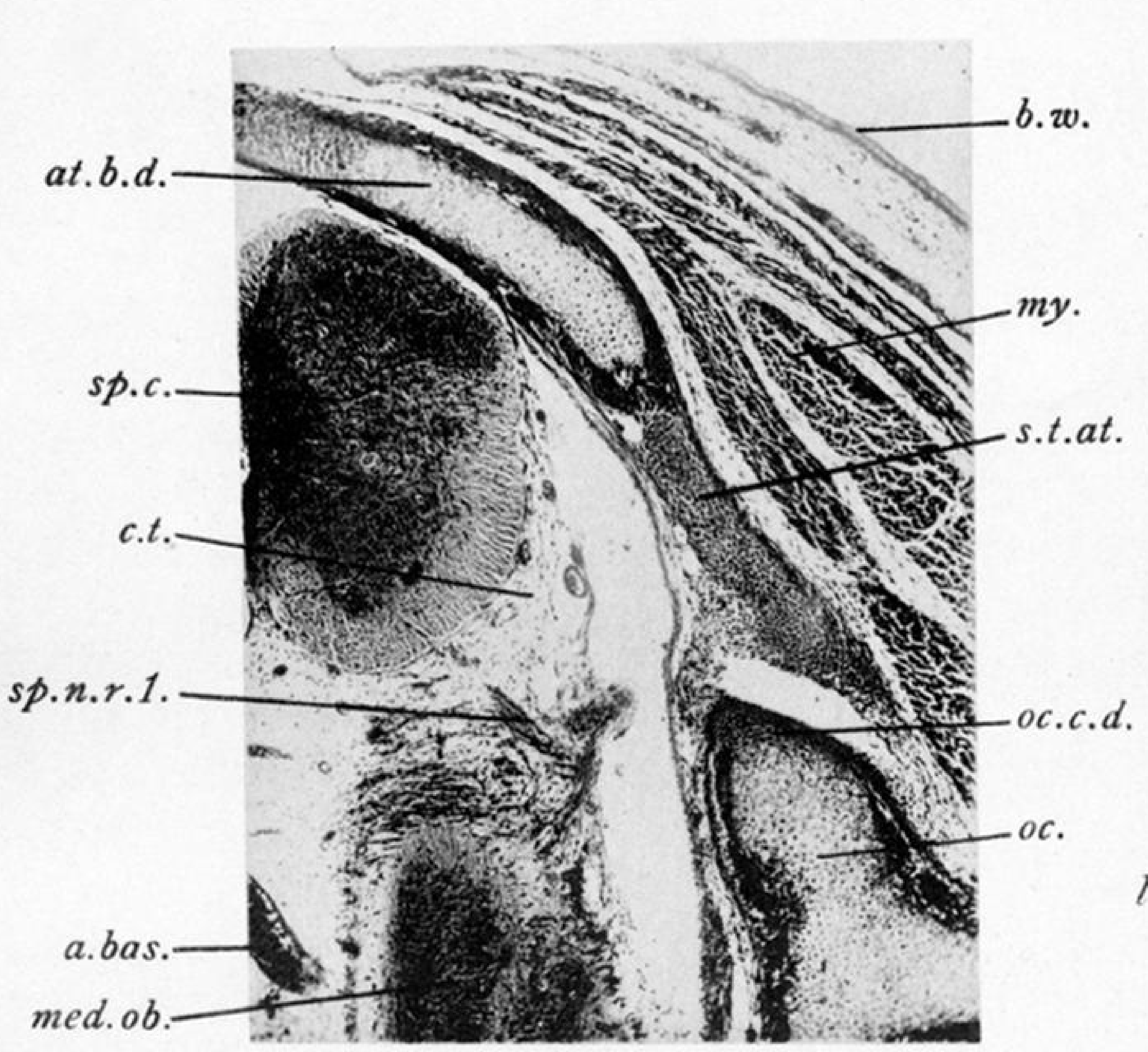

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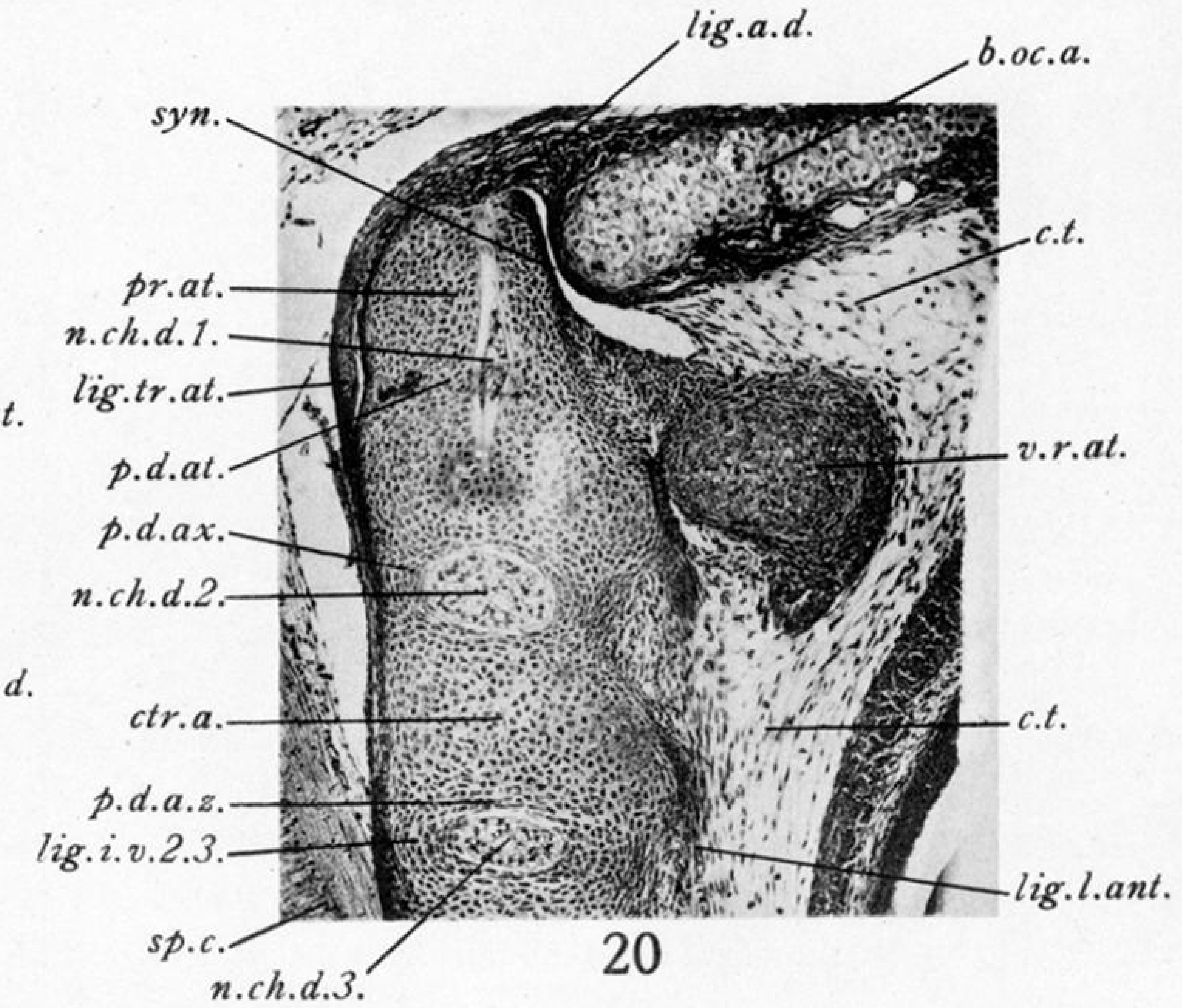

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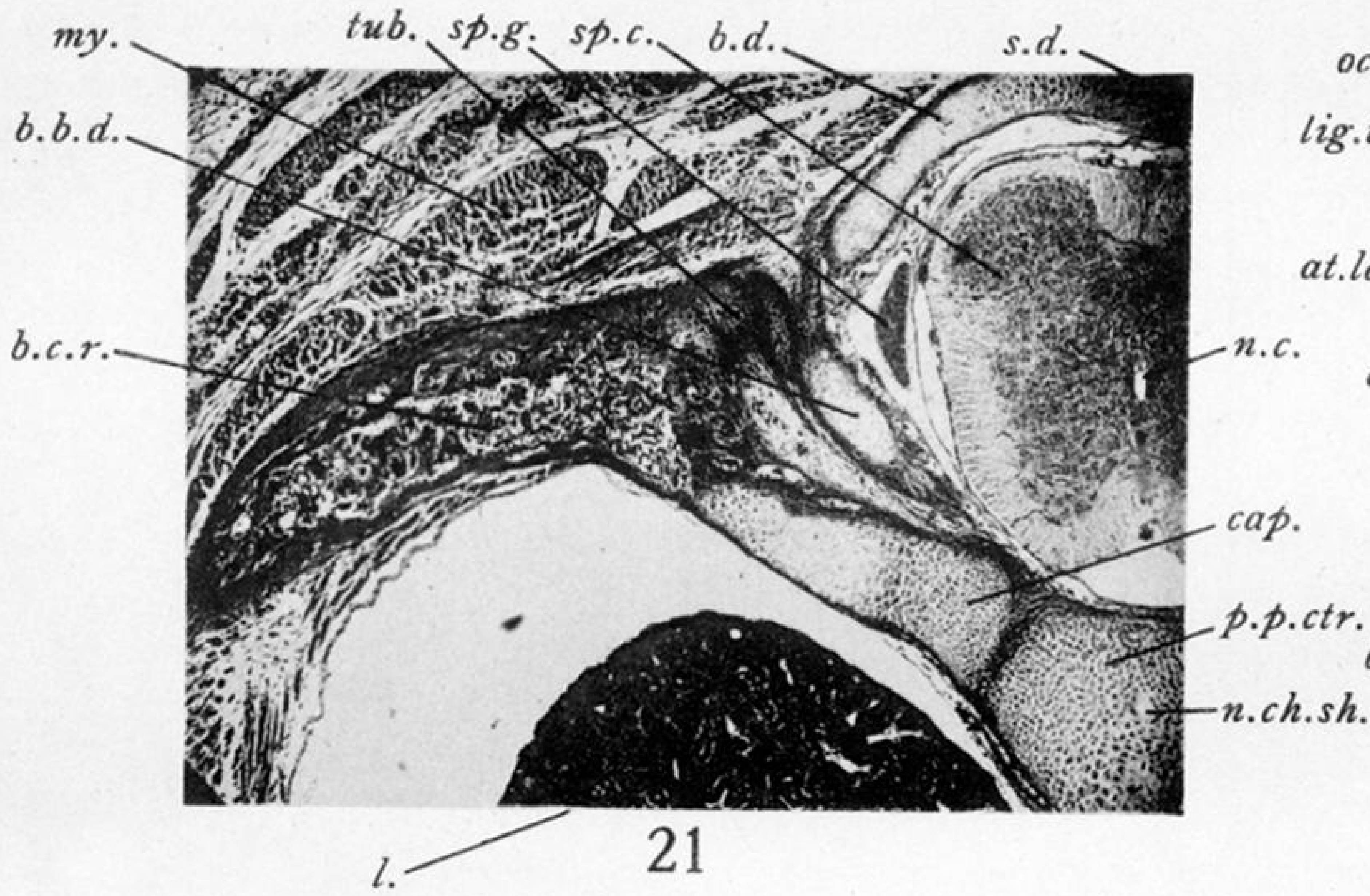

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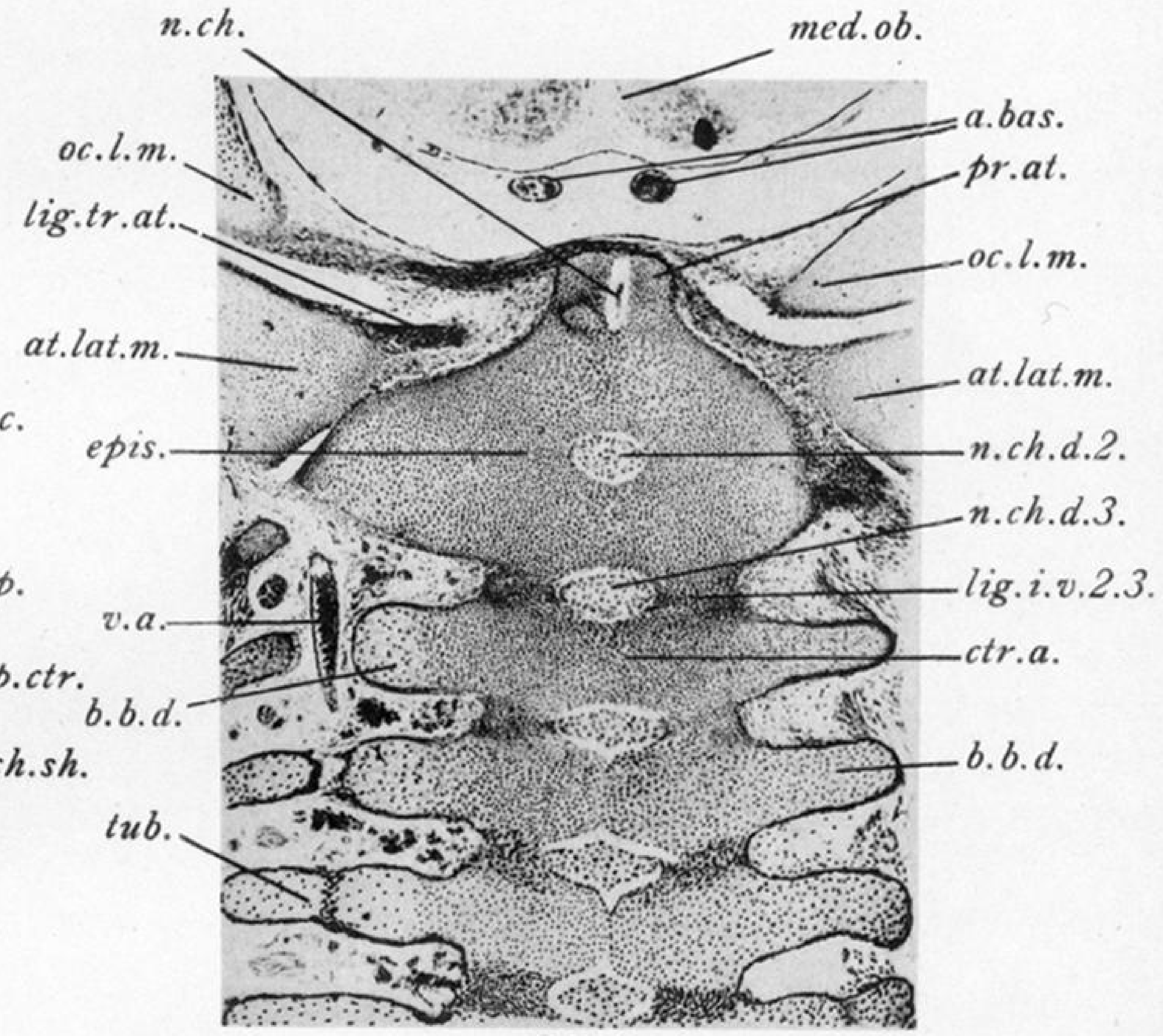

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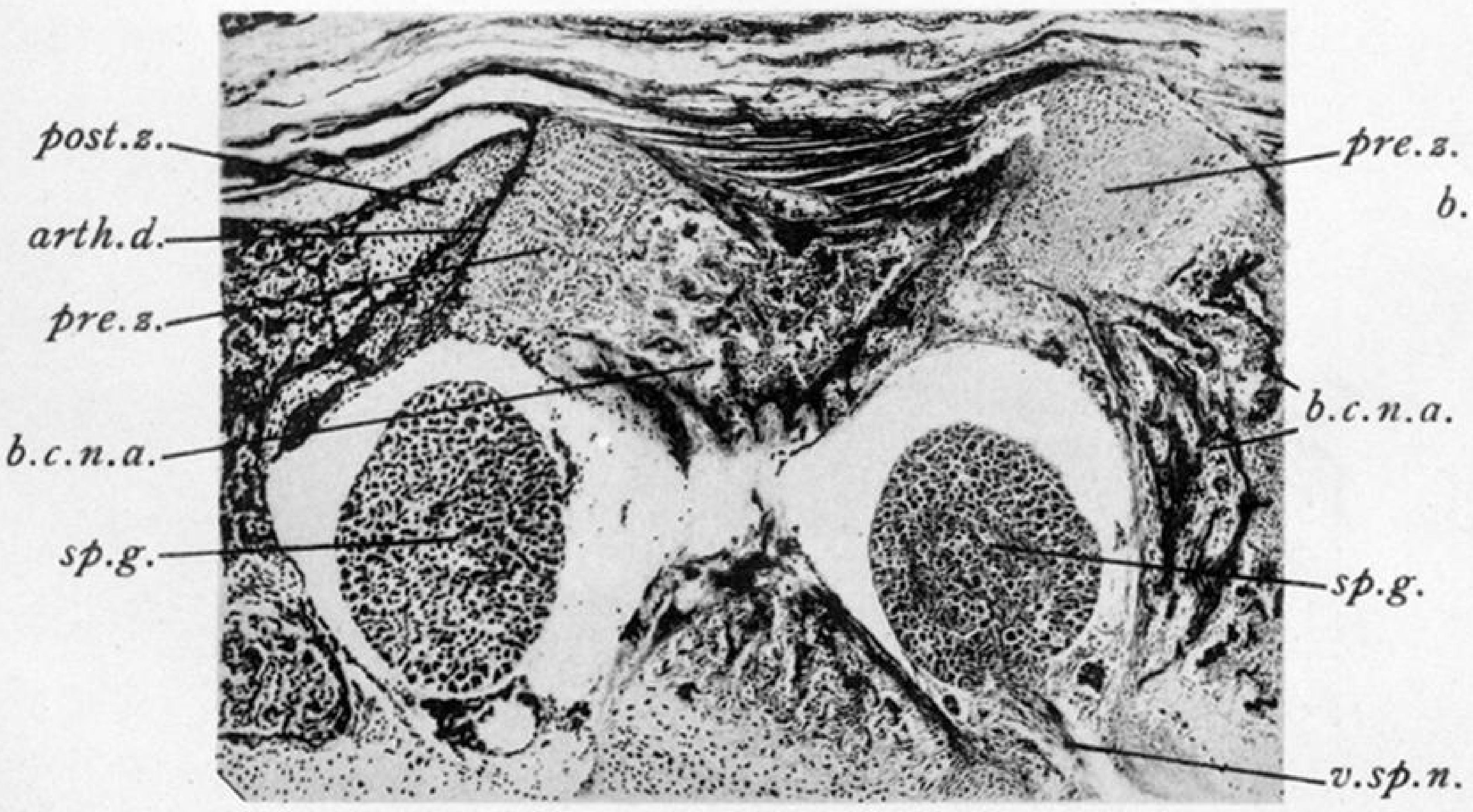

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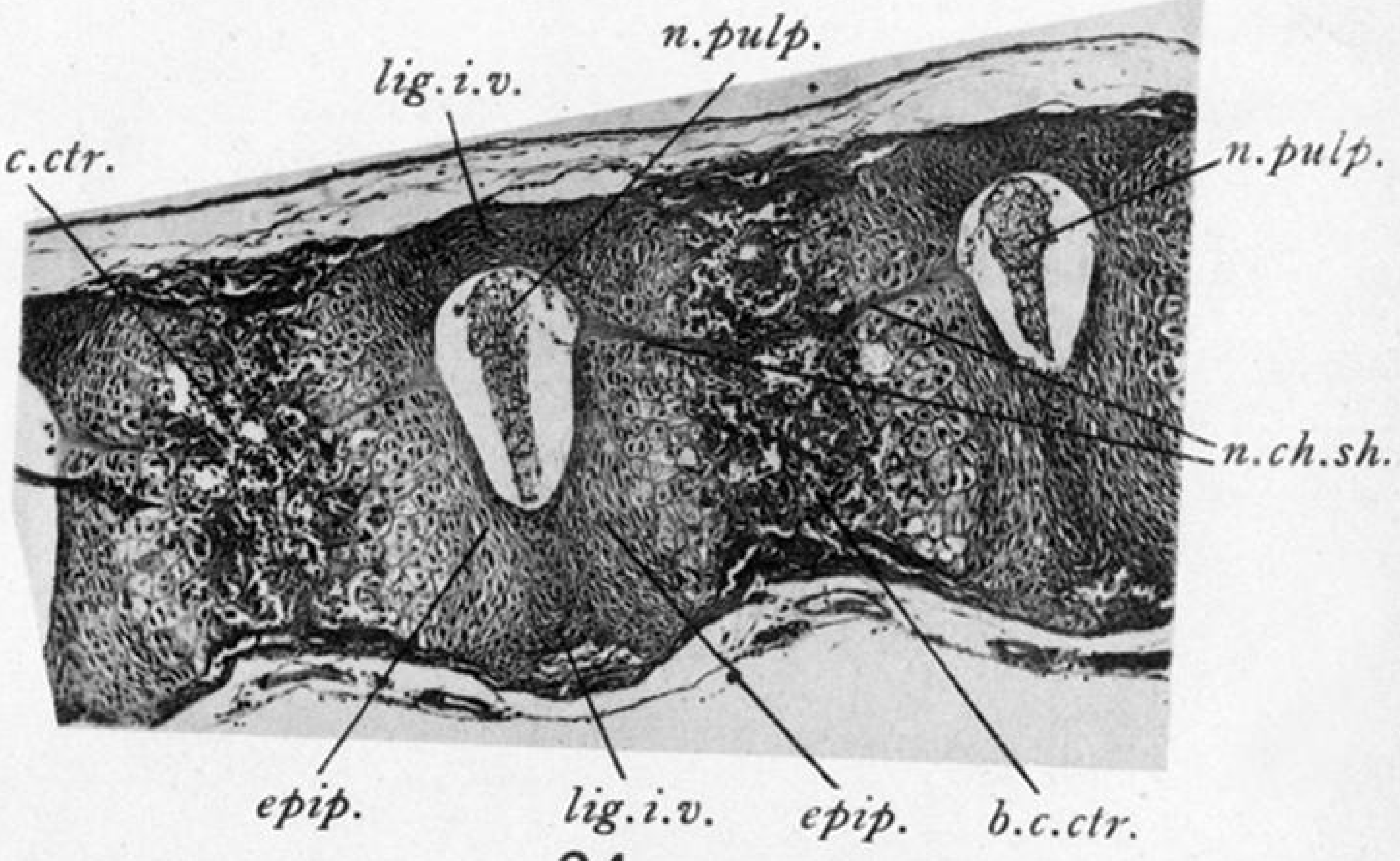

24

PLATE 14.

FIG. 19.—Section, from a transverse series, passing through the anterior portion of the atlas basidorsal of a 12·5 mm. embryo. On the right is seen the tissue enclosing the loop of the vertebral artery and also the condylar part of the occipital rudiment. × 45.

FIG. 20.—Sagittal section through the anterior cervical region of a 12·5 mm. embryo. Again, note the notochordal dilatation. × 65.

FIG. 21.—Transverse section through the thoracic region of a 14·0 mm. embryo. Showing the bone centre in the angle of a rib. × 45.

FIG. 22.—Frontal section through the cervical region of a 14·0 mm. embryo. × 34.

FIG. 23.—Parasagittal section through the base of a neural arch in the newly born animal (23·0 mm.). × 44.

FIG. 24.—Sagittal section through the thoracic centra of the newly born animal, showing the nucleus pulposus and bone centres. Note the unbroken notochordal sheath remnant. × 58.