

THE DEVELOPMENT OF THE NUCLEI PONTIS AND THE NUCLEUS ARCUATUS IN MAN

CHARLES R. ESSICK

From the Anatomical Laboratory of The Johns Hopkins University

TWELVE FIGURES

The following paper shows the origin of the gray matter found on the ventral surface of the adult rhombencephalon from the 'Rautenlippe' or rhombic lip, along its attachment to the medulla, and the path of migration of the cells formed here to the position characteristic of the fully developed brain. Almost all of the cells of the arcuate and pontine nuclei arise by karyokinetic division around the attachment of the roof of the fourth ventricle and then wander to their proper places. The pathway of this cellular migration proves to be a very superficial one, remarkable both for its constancy and its definite limits. The arcuate nucleus forms by a migration over the surface toward the ventral median fissure; the pontine nuclei choose a path which corresponds in every detail to the fibro-nuclear mass which I described for the adult as the corpus ponto-bulbare.

It is well known that all of the nuclear material in the central nervous system is derived from that portion of the ectoderm which closes in to form the neural tube; and our knowledge of the exact manner of this cellular distribution is due mainly to the researches of His. This author has pointed out that all of the nerve cells in the central nervous system first passed through the stage of neuroblasts and in their development are usually wont to leave the place of their origin so as to enter into the formation of the gray matter at a distance. The newly formed gray masses may: (1) remain in the neighborhood of the matrix, e.g., motor and arcuate cells of the spinal cord, or, (2) pierce the substance of the medullary tube in a radial direction and collect on the surface into an independent layer. Such wandering of cells from the

matrix of the original ventricular gray matter takes place in the formation of the cerebral cortex. (3) Subsequent to a definite bending in the medullary wall, newly formed gray masses may be transferred from the dorsal to the ventral portion of the brain by a migration of cells in a tangential direction, e.g., arcuate formations, olive and accessory olives, and a part of the nuclei lying in the pons. The third of these processes as described by His is responsible for the development of the gray matter constituting the arcuate nuclei of the medulla and the basilar nuclei of the pons.

The compact manner in which the neuroblasts arrange themselves in their migration to the pontine flexure, has attracted the attention of many observers both in macroscopic and microscopic preparations, nevertheless, with the exception of Streeter ('12) only a casual mention of it has been made by them. Blake ('00) in his description of the roof of the fourth ventricle noted cells which were transferred to the ectal surface of the oblongata by the formation of the secondary rhombic lip and he could trace them in many embryos as far cephalad as the trigeminal nerve. He ventured the suggestion that they might be connected with the ganglia of some of the cranial nerves. His ('04) has given a good illustration (fig. 103) of the rhombencephalon of a 5 cm. fetus and has shown the outlines of the thick mass of cells passing from the rhombic lip to the pontine formation. From its appearance with the naked eye, as well as in serial sections, he identifies it with the *Corpus Trapezoides*, which thus occupies a superficial position at this time (p. 163). Streeter ('07) in dissections of the Seventh nerve in pig embryos called attention to the presence of a ganglion mass connected with the pons ganglia which could be traced backward as a surface ridge between the facial and acoustic nerves, to end on the dorso-lateral surface of the restiform body. Since then he has suggested ('12) two possible origins for the pontine nuclei: the corpus ponto-bulbare and the mantle zone of the pontine region. Neuroblasts from the latter source emerge through the marginal zone as happens with the cortical cells of the cerebellum. Orzechowski ('08) in human foetus measuring 17 and 23 cm. has described ganglion masses connecting the rhombic lip, lateral recess wall and pons, which he considers the

embryonic corpus ponto-bulbare. He believes that the adult structure may contain portions of undeveloped embryonic tissue and be responsible for the frequent tumors in the cerebello-pontine angle. With the exception of the mention made by His of a migration of cells to form the arcuate nuclei nothing has been contributed to their development.

TABLE I

LENGTH (Crown-rump)	COLLECTION NUMBER	PLANE OF SECTION	THICKNESS
<i>mm.</i>			μ
20	128	Coronal	50
20	368	Sagittal	20
20	22	Transverse	50
23	382	Sagittal	50
24	405	Sagittal	40
30	227	Sagittal	50
30	75	Sagittal	50
30	86	Coronal	50
33	211	Sagittal	50-100
33	145	Sagittal	50-100
35	199	Sagittal	50
46	95	Sagittal	100
50	96	Sagittal	100
50	84	Transverse	50
50	184	Sagittal	50-100
80	172	Transverse	100
96	484	Transverse	40
113	490	Transverse	30
115	219	Sagittal	50-100
143	508	Transverse	40
188	509	Transverse	50
295	491	Transverse	40

In carrying out this investigation the writer was given abundant opportunity to examine the large collection of human embryos brought together in this laboratory by Professor Mall. Table I gives a list of the embryos used in this study.

Wax plate reconstructions were made after the method of Born. Human, pig and rabbit embryos, stained in toto with alum-cochineal, were prepared for dissection as described by Streeter ('04, p. 87). Whole brains stained in iron-haematoxylin also gave brilliant differentiation. The most instructive specimens of

the migrating strands of nuclear material were obtained in embryos stained in methylene blue. After previous hardening (10 per cent formalin is excellent), the brain is carefully taken out of the skull and all of the pia mater dissected off; very great care must be exercised in removing this vascular membrane in order that the tiny penetrating vessels do not tear the surface of the brain which then stains very deeply along the ruptured edges. The specimen is placed into an aqueous solution of methylene blue (saturate aqueous methylene blue and water equal parts) for thirty to sixty seconds, rinsed in water, and transferred to water for study. The whole brain is tinted blue but the most prominent parts take a more intense stain so that all of the fine surface irregularities are outlined in great detail. This brings out with remarkable clearness the anastomosing strands of cells converging into the pontine formation.

Confusion might arise out of the terms employed here so that a word may not be out of place concerning their meaning. Inasmuch as the flexures of the brain as well as the position of the head are not fixed, I have used the words 'cephalad' (forward, front, anteriorly, cerebrally), 'caudad' (backward, behind, spinalward), 'dorsal,' 'ventral,' 'lateral' and 'mesial' just as if the central nervous system were a simple straight tube placed in the head as the spinal cord is in the body. This, it seems to me, will facilitate the description of relations of parts which are constantly shifting their positions in relation to the body. In addition, I might state that I have used the term 'neuroblast' loosely, so as to include all undifferentiated cells which have not taken on a definite form.

In considering the development of these basilar masses it may be of advantage to review briefly some of the relations which exist in the rhombencephalon just before the cells, destined for the pontine and arcuate nuclei, set out from their germ centers. His ('91) has carefully reconstructed some of the intramedullary nuclei and nerve roots with their relations to the surface and brain flexures in an embryo of 22 mm. (figs. 5 and 17). He has called attention to the fact that at this time, towards the end of the second month, the formation of new neuroblasts has ceased in the medulla and it is only with difficulty that a mitotic figure is dis-

covered around the ventricular cavity where great numbers were present in the earlier stages. With the cessation of its activity, the epithelium lining the cavity of the fourth ventricle becomes very sharply marked off from the underlying nervous tissue and it would be expected that the various nuclear masses in the medulla have received their allotment of cells, further growth consisting of increase in size of individual elements and the addition of nerve fibers.

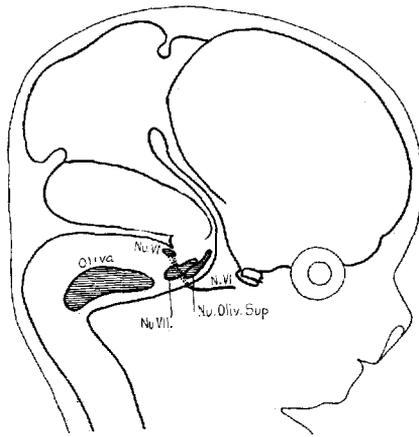


Fig. 1 Profile reconstruction of 22 mm. embryo. $\times 4.5$. Taken from His—*Entw. d. mensch. Rautenhirns*, fig. 5. I have drawn in the abducens nerve (N. VI) and its nucleus (Nu. VI).

If now one looks at fig. 1 (a profile drawing taken from His, ('91), to which I have added the sixth nucleus and its nerve) many differences from the adult are evident, the most striking perhaps being the great flexure in the pontine region—the cerebellar thickening almost touching the medulla. Just under the floor of the fourth ventricle appears the nucleus n. abducentis usually an elongated mass of cells lying immediately behind the ventricular furrow formed by the bend in the neural tube. From this nucleus the axones pass obliquely ventrally through the tegmentum to emerge just behind the summit of the pontine flexure in a series of rootlets which behave much as the hypoglossal nerve roots. They are quickly gathered together to form a single nerve trunk.

The facial nucleus has a remarkably constant form, the outline of which is similar both in sagittal and coronal sections. It might be compared to a pear, the smaller cephalic extremity tapering off bluntly. The nucleus preserves this constricted end in the adult as has been brought out in a model made by Mr. Weed in this laboratory. Cephalad through half of its extent it lies dorso-lateral and parallel to the superior olive and extending far in front of the outline of the nucleus n. abducentis. It will be observed that the relation to the superior olive is that of the adult yet one would miss the familiar appearance of the facial nucleus seen in transverse sections through the cephalic pole of the inferior olive. In other words, the caudal tip of the facial nucleus is distant a considerable interval from the cephalic tip of the inferior olive. The olivary complex, still very incompletely developed, is made up of an elongated mass of cells situated near the raphe. It shows a marked bend conforming to the flexure of the medulla in the neck region. Its cephalic pole, as projected on the lateral surface, falls behind the transverse level of the seventh nucleus. Of the greatest importance is the histological appearance of the rhombencephalon, the ventral surface of which is made up of the marginal veil ('Randschleier' of His) and in its nuclear free network run the fibers comprising the long association tracts. This layer, striking in sections on account of the dearth of nuclear material, forms a brilliant background which permits one to readily outline the nuclei wandering over its surface later.

From the embryological series of this laboratory definite evidences of the migration leading up to the formation of pontine nuclei appear in an embryo of 23 mm. (Mall Collection No. 382). Fig. 2 was drawn from a wax-plate reconstruction of the rhombencephalon of this embryo. Here the degree of medullary development corresponds pretty accurately to that of the 22 mm. embryo just described. The behavior of the cells lining the ventricular cavity deserves particular attention, inasmuch as they furnish the neuroblasts for the future pontine nuclei. The ependyma covering the floor of the fourth ventricle over the basal and alar plates has lost all signs of the great activity which it showed during the formation of the tegmental structures. The cells

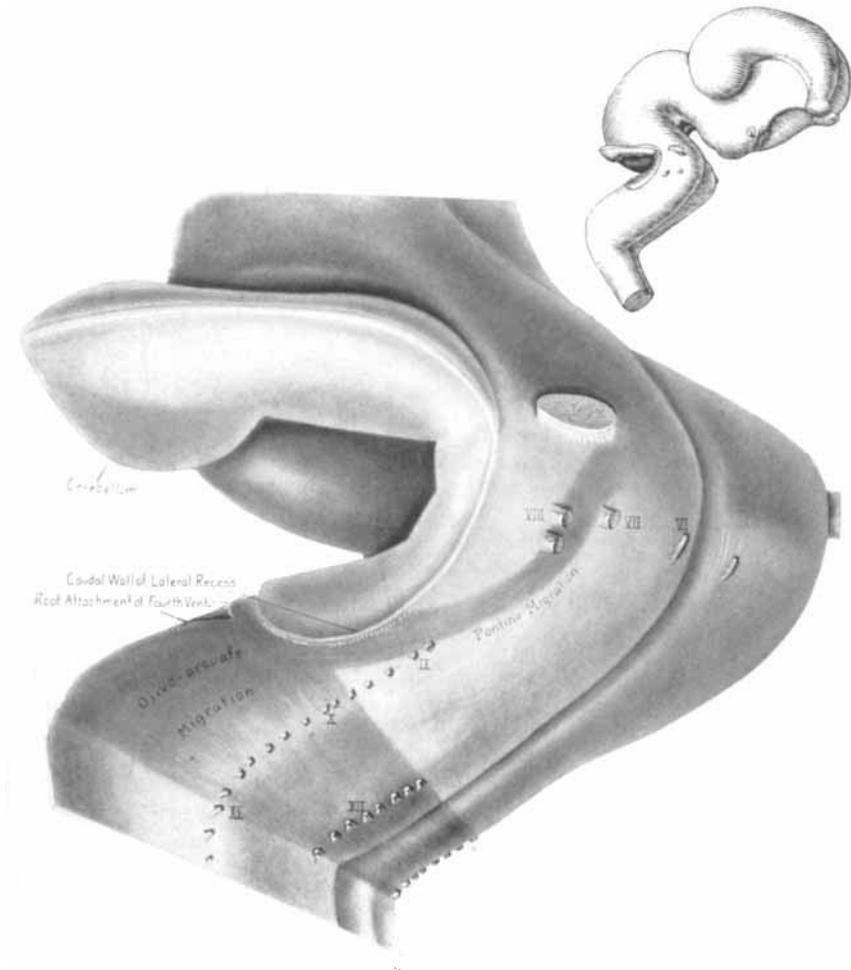


Fig. 2 Ventro-lateral view of a wax-plate model of the rhombencephalon of a 23 mm. embryo. $\times 18$. (No. 382).

composing it are sharply marked out into a definite lamina and only after searching through many microscopic fields is one able to detect evidences of cell division. In marked contrast to this inactive region, the lip-plate which makes up the caudal wall of the lateral recess and the roof of the fourth ventricle just behind it, is found busily engaged in producing new elements. The furrow formed by the attachment of the roof plate to the medulla, contains great numbers of karyokinetic figures in every high power field of the microscope and in this neighborhood the ependymal zone is not so sharply differentiated into such a thin layer as covers the medullary floor nearer the midline. Its cells are more closely packed, its nuclei take on a deeper stain, and the line of demarcation from the subjacent tissue is partly destroyed by the protoplasmic processes of the new neuroblasts which are beginning to push toward the surface of the brain. The exact manner of arriving at the surface is illustrated by a more fortunate section (fig. 3) through a slightly older embryo. Here the deeply staining cells, poor in protoplasmic envelope, may be seen to leave their position near the ventricular cavity, and to come together at the surface where they form a thin sheet of closely arranged cellular material. When once they have gained the surface of the brain they migrate toward the pontine flexure, always preserving their superficial position.

By referring to fig. 2 a very good idea can be obtained of the zone of proliferating cells and the area covered by the migrating neuroblasts that have gained the surface of the rhombencephalon. I shall omit the description of the arcuate formation for the present and consider only that narrow elongated column of cells which is seen to turn toward the pontine flexure. It is very easy to identify the densely-staining closely-arranged nerve cells in sections and I have imitated the appearance one gets specimens stained *in toto* by shading this column. The cells, that have left the ventricle, converge into a well-defined band which, as it curves around the restiform body, embraces the more anterior of the rootlets of the glossopharyngeal nerve and passes between the facial and acoustic nerves as far forward as the trigeminal nerve. At this stage the cellular sheet is very thin, being but 4-5 cells deep

between the seventh and eighth nerves and where it ends behind the fifth nerve being but a single cell in depth.

We have then a narrow well-defined band of neuroblasts derived from germinal centers situated along the attachment of the roof of fourth ventricle to the medulla, and moving over the surface of the brain toward the pontine flexure. The histological characteristics make it possible to trace them as far as the trigeminal nerve as a sheet which gradually thins out toward its advancing edge. One might well compare the process to ice growing over a pond, yet unlike the latter the new material is formed at the shores only and the whole sheet moves out over the surface its thin advancing edge to meet its fellow from the opposite side.

It should be noted that the degree of development of the rhombencephalon does not always correspond absolutely with the measurements of the human embryos given in this table. A priori, it would not be expected that at any given stage each organ would always be found to correspond to those of another embryo of like measurement, but in addition to the personal elements in measuring, the fluid in which they are measured often accounts for the difference of a few millimeters more or less. Embryo No. 405 measuring 24 mm. shows a younger stage from the standpoint of pontine development than No. 382 just described. In the former embryo we can see the same active participation of the ventricular epithelium in the production of new elements and the same distribution of karyokinetic figures, yet the front ranks of advancing neuroblasts have only reached the level of the facial and acoustic nerves. This would give us a stage slightly younger than No. 382 where the advancing edge has gained the transverse level of the trigeminal nerve. Furthermore another possible error is introduced by the measurements which I have made to show the growth of the pons and they must be interpreted freely since the plane of section is rarely perfect. Obliqueness of section therefore precludes accurate comparison yet the differences are great enough to draw general conclusions.

Nos. 227, 75, and 86 (measuring 30 mm.) furnish valuable steps leading up to the fusion of the columns of the advancing neuroblasts derived from the two halves of the brain. The fre-

quency and wide distribution of the karyokinetic figures occurring around the attachment of the ventricular roof as well as the caudal wall of the lateral recess, speak for the active participation that the rhombic lip is taking in the production of pontine nuclei and as a result the roof and recess wall are thickened perceptibly. The last fetus has been sectioned transversely through the medulla and fig. 3. shows well the large production of cells along the roof attachment. It is impossible to figure dividing cells at this magnification, yet in this one section which I have illustrated, I was able to count as many as 75 karyokinetic figures immediately beneath the membrana limitans interna. A very few could be

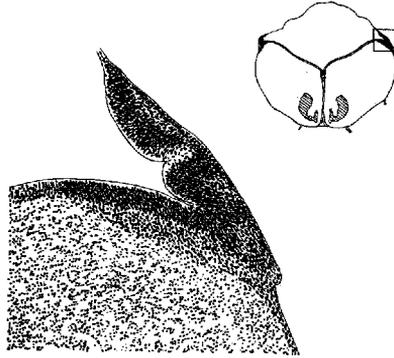
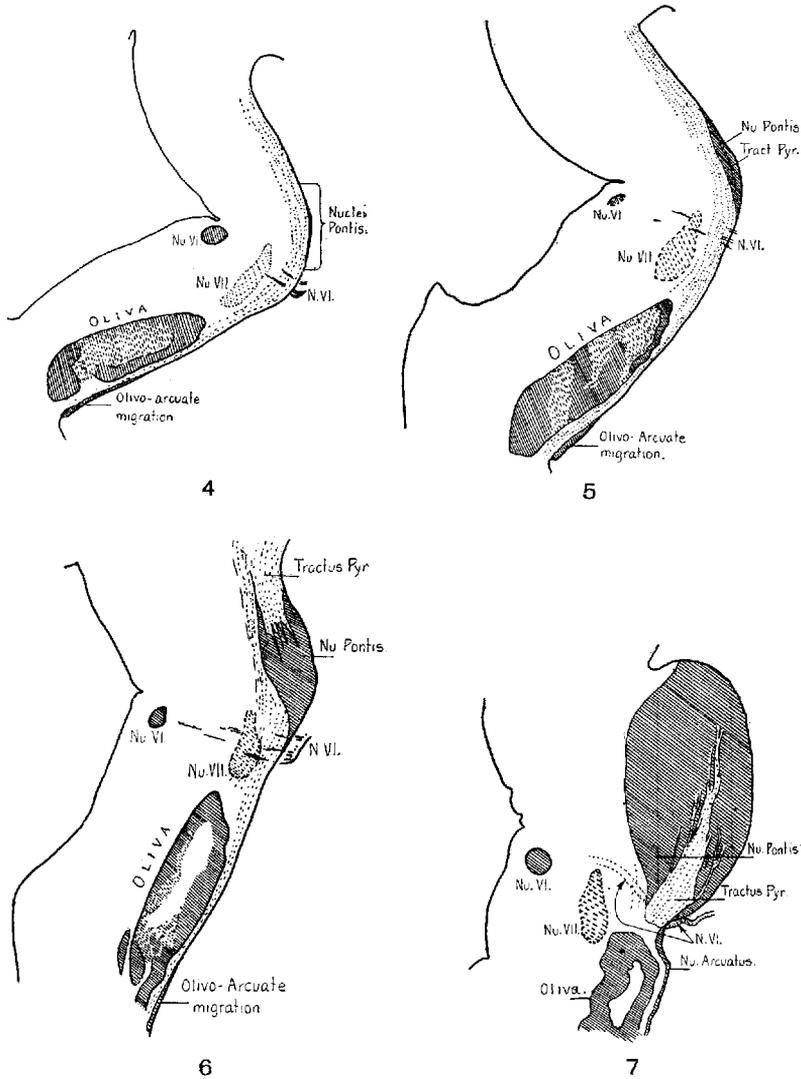


Fig. 3 Germinal centers for the basilar nuclei at the roof attachment of the fourth ventricle in a 30 mm. fetus. $\times 6.0$. (No. 86, slide 35, section 1).

made out where the cells converge at the surface, while only an occasional mitotic figure is met with among the cells turning around the restiform body. No evidences of indirect division could be found in the layer when it has arrived in front of the pontine flexure. We see, then, that the mitosis is confined sharply to the central canal. The increased production of new elements is also brought to one's attention by the increase in depth of the column passing between the facial and acoustic nerves to 10-15 superimposed neuroblasts; moreover those which had gained the trigeminal nerve in the 23 mm. embryo have moved forward and mesially in front of the pontine flexure.

The three specimens measuring 30 mm. give us the final steps in the completion of the first anlagen of the nuclei pontis. In No. 227 the cell lamina has not reached the ventral median fissure, it being possible to trace it from the border of the fourth ventricle forward to the fifth nerve where it curves mesially at almost a right angle toward its counterpart from the other side. The sagittal sections containing the sixth nerves mark the thin advancing edges of the cellular sheets approaching each other from the two sides of the brain. In No. 75 the most advanced cells have almost succeeded in gaining the midline, while in No. 86 the two columns have fused across the raphe. During their entire course from the rhombic lip on the dorsal surface to the raphe on the ventral surface, the wandering neuroblasts have kept a superficial position, only occasionally is there any tendency for any of the cells to penetrate into the clear, almost nuclear-free marginal veil. As they pass between the seventh and eighth nerves the cells are constricted into a narrow band 0.2 mm. wide, but on reaching the pontine flexure they spread out into a fan-shaped layer 0.6 mm. in caudocephalic extent. The lemnisci medialis and lateralis, which up to this stage had occupied a superficial position, are now covered over by a thin bridge of tissue and we can begin to speak of a tegmental and basilar part of the pons.

Nos. 211 and 145 (33 mm.) are cut sagittally and give us an opportunity to study the earliest pontine nuclei in their relation to the emergent nervus abducens. Fig. 4 is a camera lucida outline of the section through this nerve. The axones after leaving their nucleus take a ventro-cephalic course through the tegmentum and emerge from the neural tube just behind the most prominent part of the pontine flexure. The young pontine neuroblasts, on the other hand, lie wholly in front of this flexure, spread out into a sheet whose caudo-cephalic extent is 1.25 mm. and whose depth is 0.057 mm. at its thickest part, tapering down to the thickness of a single cell both caudally and cephalically. Between the most cephalic rootlets of the sixth nerve and the most caudal cells of the pontine nuclei is an appreciable interval (almost 0.5 mm.) so that one is at once reminded of the condition



Figs. 4 to 7 Camera lucida tracings of sagittal sections through the rhombencephalon of human embryos from Prof. Mall's collection. The nucleus facialis (Nu. VII) has been dotted in by profile reconstruction.

Fig. 4 33 mm. fetus. \times 9.5. (No. 145, sl. 19, sect. 3).

Fig. 5 35 mm. fetus. \times 9.5. (No. 199, sl. 37, sect. 1).

Fig. 6 50 mm. fetus. \times 7.2. (No. 96, slide 48).

Fig. 7 115 mm. fetus. \times 7.7. (No. 219, sl. 40, sect. 4).

seen in the adult lower animals where the abducens nerve emerges from the brain some distance behind the pontine formation.

At 35 mm. (No. 199) the increased cellular activity around the wall of the fourth ventricle is shown by the great numbers of dividing cells and the twofold increase in depth of the migrating column passing between the facial and acoustic nerves. Already the neuroblasts which are crowding from both sides toward the midline have piled up over the ventral surface of the brain, so that, near the raphe, they are now four times (0.22 mm.) as deep as the stage preceding. The free interval between the emergent abducens and the caudal edge of the pons is decreased to half what it was in the 33 mm. embryos. The important contribution which this embryo adds to the development of the basilar part of the pons, consists in a few strands of longitudinal fibers lying near the midline within the thin sheet of superficial neuroblasts newly descended from the lateral walls of the ventricle. Extending in a direction parallel to the axis of the central nervous system, these inconspicuous fiber bundles separate from the well developed bundle of axones comprising the lemniscus medialis at the level of the cephalic edge of the pontine sheet and plunge into the latter where they take up a middle position as far as its caudal edge. Here again they leave the nuclei pontis and join the fiber mass constituting the medial lemniscus. It is impossible to trace these isolated fibers except where they lie among the pontine nuclei, but, as we shall see when, by continual addition to their number, more of their course can be determined, these few strands represent the first beginnings of the longitudinal fibers which are recognized in the basilar part of the adult pons as the cortical projection system. They are represented in fig. 5 by two dotted lines traversing the pontine nuclei. Concerning the first appearance of the pyramidal tract there has been a general unanimity of opinion, the most important work being that of Flechsig's work on myelination time. Tiedemann ('16) thought he saw pyramids in fetus of the third month but he was evidently looking at the eminences formed by the inferior olives which at this time lie adjacent to the ventral median fissure and cause an elevation in the position occupied by the future pyramidal tract.

The latter subsequently forces its way between the olives and gradually displaces them laterally. Certainly at this time the number of fibers making up the pyramidal tract is insufficient to cause the surface markings on the medulla which we know as pyramids. Reasoning back from the interval of time—four months—between the appearance and myelination of other systems, Flechsig came to the conclusion that the pyramids must first be laid down between the middle and the end of the fifth month. In cross sections through the olive of an 80 mm.¹ (crown-rump) fetus he is unable to recognize any tissue which may be regarded as matrix for the pyramids but thinks they arise from fibers growing down from the cerebral cortex with remarkable rapidity in the second half of the fifth month. W. His ('04) has given a valuable table (p. 155) showing the various fiber systems which he was able to identify at each stage of embryonic growth. In a fetus of 83 mm. he was unable to find the pyramidal tract, but at a length of 120 mm. he saw evidences of its appearance together with cross pontine fibers. He gives us no statement as to the part of the brain in which he observed the pyramidal tract, merely noting its presence or absence in the various embryos in his collection. With this statement of the present knowledge of the cortical projection system, I shall omit until later the reasons for believing that the few axones isolated by the early pontine nuclei represent the anlage of the cortico-spinal tract in this 35 mm. fetus.

No. 95 (46 mm.) is the youngest stage in which I could determine cross fibers among the pontine nuclei. They are most conspicuous at the lateral borders where they gather together into compact strands to form the brachia pontis. Here the fibers have a superficial position, embracing laterally the corpora restiformia as the latter turn sharply into the cerebellar hemispheres. It is possible to trace the axones coming from the pontine nuclei for some distance into the cerebellum until their course parallels the

¹ The crown heel measurement which Flechsig used was 11 centimeters. For the sake of ready comparison with my study I have put his measurements into this form from the table given by Mall in *Handbuch der Entwicklungsgeschichte des Menschen-Keibel und Mall*, Leipzig, 1910, p. 205.

large mass of fibers coming up in the inferior peduncle. The trigeminal nerve in this stage sends its rootlets through these cross fibers in an oblique direction to reach its intramedullary nuclei. The cerebro-spinal neurones which were seen among the relatively thin sheet of nuclei covering the ventral surface of the pontine flexure in the stage just described (35 mm.), are increased enormously during the interval left in this series, and we have a striking similarity to the picture one gets in sagittal sections through the pontine region of the adult brain. The thickened layer of cells (now 0.642 mm.) are invaded by large anastomosing strands of fibers which collect at the cephalic and caudal border of the nuclear sheet into a solid bundle. This behavior is one of the peculiarities of the cortical projection system as it lies among the pontine nuclei where, as is well known, the otherwise compact fiber tract is broken up into smaller fasciculi by the cross fibers and nuclei of the pons. Caudad the reunited fibers fuse inseparably with the median lemniscus soon after leaving the nuclei pontis; cephalad I am unable to trace them beyond the cephalic flexure.

His has suggested that this interweaving of cross fibers of pons with pyramidal tract points to an alternating time of deposition of the component parts of the two systems—the development proceeding in a direction away from the central canal. This for the most part is true. We find the new cells which have migrated from the ventricular walls, spreading themselves over the surface of those already descended to the pontine flexure, and as the new axones come from the cortex they tend to grow among the younger nuclei, i.e., to grow nearer the surface. Thus each fasciculus when it enters the pontine nuclei, pushes along near the surface but it is soon deeply buried by new cells which are continually streaming down from the ventricle. As a result there is a separation of the pyramidal tract into a series of fasciculi which unite again at their exit from the caudal border of the pons. Some of the cells, however, after passing between the seventh and eighth nerves forsake their superficial position and plunge between the cross fibers of the pons. This is well illustrated in fig. 9 the more deeply staining young cells are seen forcing their way between

the transverse pontine fibers going into the brachium pontis. Moreover the fasciculi of the pyramidal tract keep on growing so that one must infer that some of the axones coming down from the cortex add themselves to the bundles more deeply placed in the pons.

During the period of growth between 35 mm. and 46 mm. enormous numbers of neuroblasts have come down from the lateral boundaries of the fourth ventricle. These new cells together with their processes sent out transversely and the cortical axones threading their way among them have increased the thickness of the basilar portion to 0.642 mm. There is a tendency for the cells from both sides to crowd toward the midline, thus giving rise to the typical crescentic shape of the pontine nuclei which one obtains in transverse sections through the pons. The increase in thickness is also accompanied by an increase in caudo-cephalic extent. The latter, however, does not proceed with the same proportional rate as the former, so that the sagittal sections of pontine nuclei are becoming more and more oval. In spreading caudad the interval between the nuclei pontis and the abducens rootlets has been gradually reduced until in this fetus the more cephalic axones are surrounded by pontine nerve cells.

In fetus of 50 mm. (Nos. 84, 96, 184) the number of cerebrospinal neurones have increased to such an extent that it is now possible to follow them with sufficient accuracy to be certain that we are dealing with the axones of the cortical projection system. The fibers splitting up among the pontine nuclei already form comparatively large bundles (fig. 6) which are collected together into a solid fasciculus at the cephalic end of the pons. Here they come into close relationship with the lemniscus medialis, but it is not impossible to trace the large fiber mass into the internal capsule. Traced cerebrally the *crura* gradually diverge from the midline and turning around the cephalic flexure they lie ventral and lateral to the nucleus hypothalamicus, while the medial lemniscus has a more dorso-lateral position with regard to this nucleus. The fibers making up the pyramidal tract can be traced definitely into the internal capsule. Spinal-ward I have been unable to differentiate the projection system from the medial lem-

niscus soon after it has left the caudal border of the pons. There is then no question but that we have been dealing with the beginnings of the pyramidal tract as early as 35 mm.; its behavior among the pontine nuclei making identification certain. Former observers have confined most of their attention to the medulla oblongata where it is hopeless to try to pick out the few strands of fibers when they first grow down from the cortex. The increase in number of these axones is so gradual that it is only in the older fetus where enough fibers are collected to form the surface marking on the medulla which we can recognize as pyramids. Flechsig is sure that there are no pyramids at 80 mm. and probably "the pyramidal tract is completely lacking." To harmonize the system with other observed facts he assumes that they must grow down rather rapidly from the cortex when once they start, since their myelinization occurs after birth and the usual interval between the formation of a nerve fiber and its acquirement of a myelin sheath is about four months. This of necessity would have the pyramidal tract appear about the middle to the end of the fifth month or 14 to 16 cm. To this one must answer that a myelin sheath does not appear on every axone of this system simultaneously; it begins rather on isolated fibers and is first complete at the age of two years.

In these fetus of the eleventh week the basilar part of the pons has reached a thickness of 0.7 mm. (fig. 6). The abducens nerve rootlets are almost entirely surrounded by nuclear material after they leave the tegmentum, only the caudal two or three fasciculi being free. Great numbers of neuroblasts are encountered passing between the seventh and eighth nerves, forming a stream 0.16 mm. deep, while the germ centers at the ventricular margin are busily producing new cells. A fortunate sagittal section through No. 96 has been illustrated to show the participation which the greatly thickened lateral recess wall takes in contributing cells to the pons. For purposes of orientation a wax-plate reconstruction was made with the section drawn on its cut surface (fig. 8). As the cerebellum in its growth crowds against the medulla, this caudal wall is flattened out and becomes part of the mesial wall of the recess. A separation of the cells coming from

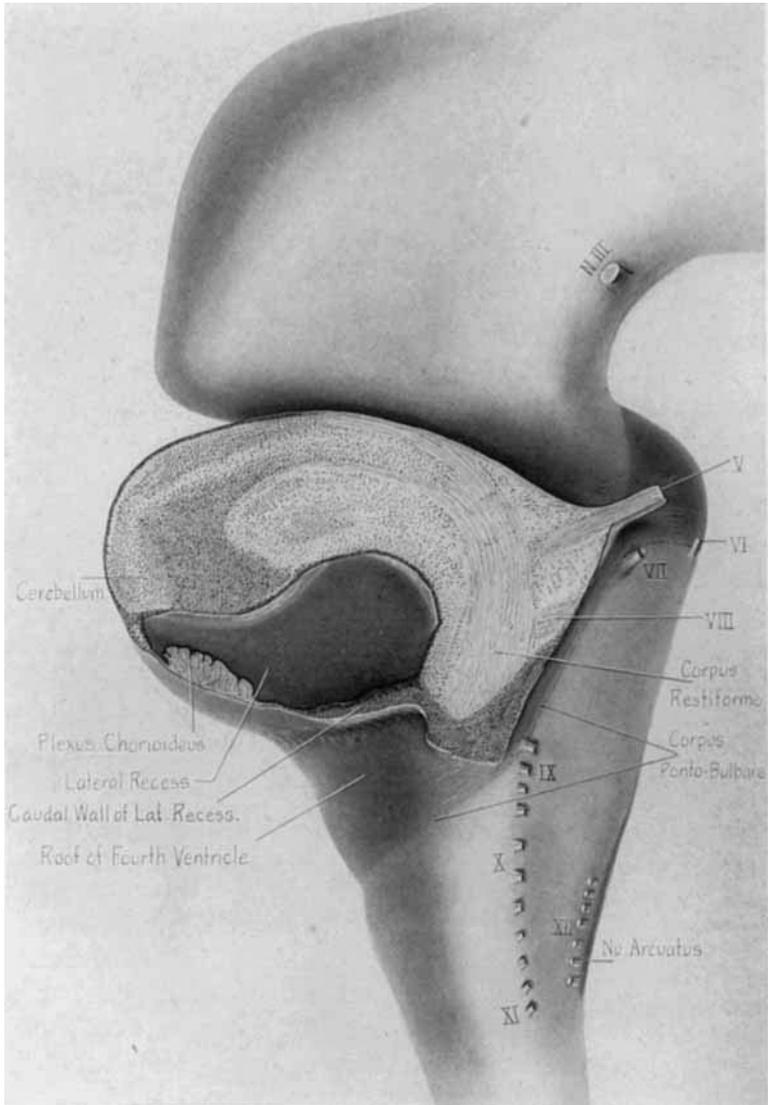


Fig. 8 Sagittal section through the cerebellum and lateral recess of a 50 mm. fetus from a wax-plate reconstruction. $\times 15$. (No. 96).

the recess wall from those of the ventricular roof is purely arbitrary, since the two origins are really continuous with each other. This association in the adult was first pointed out by Orzechowski ('08, p. 41). For embryological reasons the nuclear thickenings of the lateral recess wall with their accompanying fibers occurring in the adult should be included in the structure which I have termed the corpus ponto-bulbare. At this stage the cavity of the fourth ventricle shows a peculiar tendency to form small outpouchings along the attachment of the roof at the place where the pontine nuclei are being formed. From two to four such recesses can be made out extending laterally for a considerable distance from the main ventricular cavity and causing the external surface to be thrown up into ridges. In section they may be round or slit-like and are lined with deeply staining cells, great numbers of which are found in process of karyokinetic division. The production of neuroblasts at this stage is enormous and these lateral extensions from the ventricle furnish a greater expansion of ependymal surface and thus increase the germ layer where cell division can take place.

In the older fetus the system of ventricular outpouchings becomes more complicated and secondary processes are formed which are distinctly tubular. The size of the lumen varies, being sometimes less than the width of a single nucleus. It is always lined with a simple layer of cells which are definitely ependymal and as long as pontine nerve cells are being formed these tubules can be made out with a little difficulty among the closely packed neuroblasts but always the center of mitotic activity. With the emigration of the last of the new elements the ventricular prolongations stand out with much greater clearness. This is particularly well shown in the five and eight months fetus—in the latter, one is struck by the greater number of such tubules both in the roof attachment and the caudal wall of the lateral recess.

At the beginning of the fourth month, as shown by No. 172 (80 mm.), the evidences of marked cellular activity, i.e., extensive mitosis and deeper staining are still present around the roof attachment of the fourth ventricle and the caudal wall of the lateral recess. This fetus does not illustrate any new principle

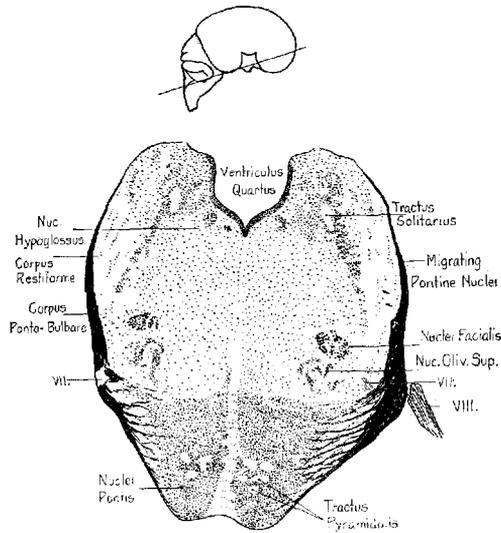


Fig. 9 Oblique section through the rhombencephalon of an 80 mm. fetus. $\times 7.5$ (No. 172, sl. 200, sect. 2).

but furnishes a beautiful single section which includes all of the relations of the cells wandering to the pontine nuclei. Owing to the cervical flexure a cut which sections the spinal cord transversely passes tangentially through the medulla and pons (fig. 9). The great thickness at which these sections were cut gives the migrating column of nuclei a very deep red stain, almost black, and I have represented it as black; the plane of section falls behind the roof thickening which marks the true germinal centers, but it does show with remarkable clearness the whole path of the neuroblasts starting from the ventricular edge, encircling the corpus restiforme, passing between the facial and acoustic nerves, to take their place among the cross fibers of the pons.

During the period extending through fetus 508 (143 mm.) there is a continued addition of cells to the basilar part of the pons. In a fetus of the thirteenth week, 96 mm. (No. 484), the maximum production of new cells has been reached. At this time the ventricular edges on both sides of the brain are full of karyokinetic figures and extending from these places are two thick columns of

closely packed young cells which pour into the pons between the seventh and eighth nerves. The cells which had already descended from the rhombic lip show no tendency to assume the ganglionic form. They are still rather closely arranged, the cytoplasm scanty and clear, the nucleus small and containing one or two chromatin condensations but no real nucleolus. Two different reactions toward the haematoxylin can be made out among the nuclei of these cells—the one quite densely staining and usually smaller nucleus, the other slightly larger and more vesicular. The latter form the larger proportion of nuclei. From the extensive cross-fiber system already present one must assume that great numbers of these pontine cells have sent out nerve processes and have taken up their final position. The newly added cells tend to remain superficially yet a considerable portion push in between the transverse fibers and cells already fixed. Although the stain of No. 490 (113 mm.) does not permit of good histological study, yet it may be readily seen that the pontine nuclei are still receiving great numbers of new elements from the rhombic lip. At 143 mm. (No. 508) the production of neuroblasts destined for the basilar parts of the hind brain has diminished very appreciably. The mitoses around the ventricular margin are fewer and the ependymal lining has begun to be separated quite sharply from the underlying nervous tissue. Many of the cells in the path of migration have larger and clearer protoplasmic bodies, apparently unwilling to complete their journey to the pontine region. Others appear as all of those in earlier stages with elongated almost naked nucleus pushing on toward the pons before assuming the ganglionic form. Of the cells which have long since gained their permanent position in the pons, many can now be recognized as ganglion cells. The nucleus is very much larger, although rarely showing a distinct nucleolus—the cytoplasm, paler than the framework in which it is embedded, is also increased in amount. The greater number of cells have grown slightly—possessing a smaller, more densely staining nucleus surrounded by a clear protoplasmic envelope.

During the interval between 143 mm. (No. 508) and 188 mm. (No. 509) the formation of neuroblasts ceases entirely and the

further development of the pons consists of an addition of axones (with their later myelinization) and the maturity of the nerve cells. The rhombic lip has given up all signs of activity in the next stage of which I had access to serial sections. In No. 509 (188 mm.) the lining of the central canal is uniformly at rest in the medullary region and is now as sharply demarcated around the roof attachment as that covering the basal and alar plates. The roof thickening now reminds one of those sections through the adult medulla which pass through the ponticulus of Henle. Comparatively few of the cells have not descended into the pontine region but are taking on the characters of adult ganglion cells along the path where the pontine cells migrated at an earlier period. Just how many cells fail to move into the pontine region but take up their position around the restiform body varies in the different brains. This helps us to understand the wide differences which were noted in the size of the fully developed corpus pontobulbare. With the disappearance of the closely packed nuclei around the attachment of the rhombic lip and the consequent clearing up of the roof thickening, the ventricular outpouchings stand out with great clearness. They are not very unlike tubular glands with a single cell lining them and show a tendency to branch frequently.

The increase in the number and size of the bundles of cross fibers in the pons gives the basilar part a greater thickness in the adult where one finds the fiber material outweighing the nuclear material. In the first beginnings on the contrary just the reverse holds true, and the nuclei pontis alone form the protuberances in the pontine region. The individual ganglion cells, although larger than the preceding stages, are still immature. The protoplasmic bodies do not accept the counter stain and few of the nuclei possess a well formed nucleolus.

By the eighth month (No. 491) the protoplasm of the ganglionic cell is no longer clear but takes up the counter stain. Many of them now look like the ganglion cells of the adult except for their smaller size. The entire migratory path is strewn with cells. Passing between the facial and acoustic nerves the column can be traced around the restiform body into the roof of the fourth ven-

tricle and the mesial wall of the lateral recess. A great many of the cells can be recognized as ganglion cells of the corpus pontobulbare, but as a whole the appearance is one of immaturity. Here and there can be found a cell whose protoplasm stains but as a rule the large vesicular nuclei are surrounded by a colorless zone. The migration of nuclear material in the medulla has ceased entirely in this stage and one has to expect only the maturity of those elements already present.

Throughout the description I have disregarded a very important factor in development which transforms the hind brain of an embryo of the second month into the adult form: I refer to the obliteration of the pontine flexure. The maximum flexure in the neural tube occurs about the time of the appearance of the first nerve cells on the ventral surface and then diminishes gradually so that at birth there is still an indication of it on the ventricular floor in front of the emmentia abducentis by a furrow running transversely. The relations of the tegmental nuclei to one another as well as to the olivary complex are distorted by the extreme flexion of the brain in the pontine region. The reduction of this may be regarded as taking place around the nucleus n. abducentis as an axis, since it is situated immediately beneath the ventricular floor just behind the bend in the brain. The other nuclei, superior olive, facial nucleus and olivary complex are distributed around its circumference and are consequently separated from one another. A glance at fig. 1 will show this arrangement. Figs. 4, 5, 6, and 7, are camera lucida drawings of sections through the nucleus n. abducentis with its emergent root bundles, which were selected from such sagittal series as illustrated the change in position of the nuclear masses during the obliteration of the pontine flexure. The nucleus facialis is projected into the section as indicated by the broken lines. As the neural tube unbends, the olivary complex and pontine nuclei are gradually pushed toward one another until the cephalic tip of the former comes to be covered by the latter. The abducens nerve which in younger stages (figs. 4, 5 and 6) pursues a straight course within the medulla is bent by this process, so that it takes a caudal direction in order to reach the surface of the brain (fig. 7). The facial nu-

cleus at first separated from the olivary body by a considerable interval comes to lie in the same transverse section as the latter, while the pontine nuclei cover up the cephalic two-thirds of this nucleus. We have, then, in addition to the mere increase in size of the pontine and olivary nuclei an actual alteration of their positions as a result of the straightening out of the neural tube. As a consequence nuclear masses which were separated from one another, are crowded together and the course of the sixth cranial nerve altered.

Having considered the origin of the main mass of nuclei pontis, the possibility of cells from other sources must not be overlooked. In the region of the pontine flexure near the raphe one can make out at an early period collections of cells extending from the ventricular floor to the pontine nuclei with which they are connected. They occupy the position which is held by the nuclei reticularis tegmenti pontis (Flechsig) in the adult. Long before any cells appeared superficially on the pontine flexure the karyokinetic figures had disappeared in the ependymal sheet near the raphe, so that it is highly improbable that the nuclei pontis depends on this portion of the neural tube for many of its elements. In addition these cells of the nuclei reticularis tegmenti pontis are evident long before the pontine nuclei appear and never have the characteristic appearance of young wandering neuroblasts during pontine development. In some of the older embryos a thin sheet (one to two cells deep) are migrating from the wall of the lateral recess in front of the dorsal cochlear nuclei but the layer is narrow and composed of comparatively few cells. These cells join the pontine nuclei behind the trigeminal nerve. It is hardly necessary to exclude other sources if one considers seriously the great production of new cells around the rhombic lip. This begins at 23 mm. and continues incessantly until the fetus has passed 143 mm. in crown-rump measurement. Couple with this extensive period the short time in which any mitotic division is complete and the great numbers met with in every section and it will not take a great stretch of imagination to account for all of the cells in the nuclei pontis.

ARCUATE NUCLEI

Examination of different adult brains in microscopical sections reveals a great variation in the amount of nuclear material which goes to make up the basilar portions of the brain stem. This is especially true of the arcuate nuclei where small, more or less isolated patches of nuclear material may often be scattered along the ventral and lateral surfaces of the medulla as far as the restiform body. The arcuate nucleus proper, the most constant of these masses, lies near the ventral median fissure superficial to the pyramidal tract, extending from a point caudal to the olive up to and fusing with the pontine nuclei. At its caudal extremity, under the olive, this mass is always of greater dimensions and tapers off somewhat as it is followed toward the pons—in some brains disappearing here and there for a few sections, in others forming a continuous narrow strip under the whole length of the medulla. The arcuate nuclei proper, as well as these superficial isolated masses lying more laterally, will be shown to have a common origin and at one time to be actually continuous with one another. The principles governing their development are identical with those which we have studied in connection with the pontine formation. The same germ centers around the attachment of the rhombic lip contribute cells which migrate superficially over the medulla in front of the cervical flexure.

The formation of the arcuate nucleus, unfortunately, is not so simple as that of the pontine nuclei, but is complicated by the simultaneous development of the olivary complex. As His has shown, the latter begins as a migration from the alar plate of the rhombencephalon early in the second month. Toward the end of this month the olive can be outlined readily although it has only a small fraction of the cells which it contains in the adult. At this time one can make out in embryos of about 20 mm. large elongated nuclei, almost devoid of a protoplasmic body leaving the ventricular margin along the attachment of the rhombic lip. They are arranged in strands of a single cell in depth and two or three in width, streaming over the surface of the medulla just under the external limiting membrane. This migration is directed

toward the portion of the medulla which is under the partially formed olivary complex and recalls the undifferentiated wandering cells seen in connection with the nuclei pontis. A great many leave the surface at various points and plunge into the depth to join the neuroblasts already massed up in the olivary nuclei. A broad sheet, however, remains superficially and can be traced from the roof attachment ventrally. In some brains (Nos. 368 and 453) this sheet has moved among the vagus rootlets and advanced almost to the emerging hypoglossal roots. In another (No. 22) the migrating cells cover the entire ventral surface of the medulla just in front of the cervical flexure having met, across the raphe, those moving down from the opposite side. In other words, there exists at this period a band of superficial undifferentiated cells uniting the roof attachment on both sides which is not unlike the early pontine bridge in fetus of 30 mm. The former begins just in front of the cervical flexure and subtends one-half to two-thirds of the olivary complex (fig. 2), but unlike the latter many cells leave it everywhere and make their way into the substance of the medulla to form gray matter in the interior.

It is striking (1) that the neuroblasts of the developing arcuate nuclei imitating the pontine formation, pay no attention to the raphe but cross it in an uninterrupted sheet; (2) that they appear before the anlage of the nuclei pontis, and if one turns to mammalian embryos (I have studied pig and rabbit) which are slightly larger than 20 mm. in crown-rump measurement, (3) that a well-developed arcuate formation exists just as in the human material.

Turning to the adult brain we find each arcuate nucleus a discrete mass which is separated from its counterpart by the raphe. Moreover the arcuate nucleus is peculiar to man so that from a phylogenetic standpoint we should expect to find it developing later than the pontine nuclei inasmuch as it is last to be acquired. Furthermore, very soon after its formation in pigs and rabbits one looks for it in vain. At 51 mm. only comparatively few cells can be found, while the superficial layer of migrating cells has disappeared completely from the subolivary region of a fetal pig of 60 mm. In man, on the other hand, when once there is a collection of cells over the medulla in the subolivary region (as in

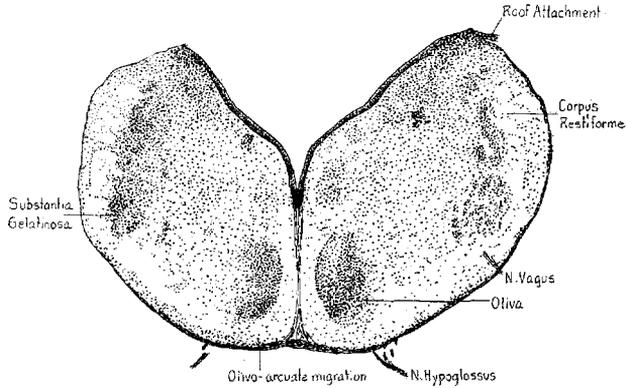
No. 22) all of the later stages invariably show nuclear material in the position which we know will be occupied by arcuate nuclei. There appears but one rational explanation which will harmonize all of these apparently jarring facts which we have determined. In human embryos at the beginning of the second month there is an intramedullary migration of cells from the rhomboid lip to make up the olive, toward the end of the month the path of migration becomes more and more superficial until many of the cells actually cross the raphe before plunging into the medulla. In the lower mammals the comparatively simple olivary complex soon acquires its allotment of cells and when production of olivary neuroblasts ceases in the roof attachment, those on the surface soon find their way into the interior. In man, on the contrary, before the olive has received all of its cells and while the migration from the rhombic lip is still proceeding actively, neuroblasts which cannot be differentiated from those destined for the olive, begin to wander over the surface among the vagus roots. These elements stop on the ventral surface near the raphe and constitute the anlage of the arcuate nucleus. Stated differently, we are probably not dealing with arcuate formation in human embryos of 20 mm. where a cell lamina lies on the surface of the medulla in the place where we know the arcuate nucleus ought to be.

Just when the arcuate neuroblasts begin to descend from the rhombic lip can only be conjectured; this uncertainty has led me to call it 'olivo-arcuate migration.' Probably at 30 mm., as exemplified by No. 86, most of the thick superficial sheet of cells in the arcuate region represents a migration of olivary elements (fig. 10). Here the deeply staining nuclei form a continuous lamina over the ventral surface, the caudo-cephalic extent of which corresponds to the spinal one-half of the olive. Even older embryos present this pons-like structure as figs. 4, 5, and 6 illustrate. First in a fetus of 80 mm. (No. 172) does one meet with any large number of superficial neuroblasts under the cerebral one-half of the olive. Here almost the entire surface of the medulla is the seat of cellular migration. From the cervical flexure almost to the pontine nuclei the pyramidal tract is covered

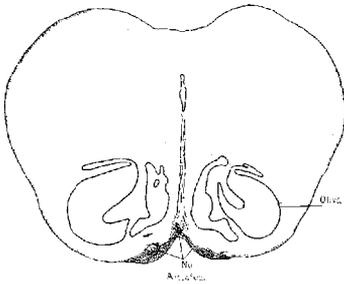
by a superficial sheet of cells many of which are pushing their way into the medulla near the raphe. The wandering of cells to the region of the cerebral half of the olive fills up, in this embryo, the gap between the pons and the band (olivo arcuate migration, (fig. 2) which was first completed in embryos of 20 mm.

The addition of new elements is even more marked in the 96 mm. fetus (No. 484) where great numbers of moving cells are directed toward the ventral portion of the medulla immediately behind the pons. Here the cells are leaving not only the rhombic lip to pursue a course similar to the earliest olivo-arcuate migration but also from the ventral edge of the thick column of migrating pontine nuclei. All along the corpus ponto-bulbare of this fetus neuroblasts can be seen to leave its ventral edge and migrate directly toward the ventral median fissure. In the adult it is well known that the arcuate nuclei fuse across the midline as one nears the pons, although at the caudal end of the olive they present two discrete swellings which lie some distance from the midline. Two mechanical factors are concerned in breaking up this uninterrupted sheet of nuclear material which is so striking in the younger fetus (fig. 10). These are the formation of the external arcuate fibers and the growth of the pyramidal tract. Already in this fetus a considerable number of arcuate axones are crossing in the raphe, the main mass of nuclei, however, still lie on either side of the midline (fig. 11). It remains for the constant interstitial addition of pyramidal axones to bring about the further separation of the arcuate nuclei. It is apparent that the cortical projection system must occupy a very inconspicuous part of the cross sectional area of the medulla of this embryo, when one considers the superficial position of the olivary complex and their proximity to the ventral medial fissure. Compare with this the fig. 12 which is a camera lucida tracing of No. 508 (143 mm). The level of this section was made to correspond with that of No. 484 by choosing both about one-tenth of the distance from the caudal to the cephalic pole of the olive. The extensive addition to the pyramidal tract has pushed the olives apart as well as the arcuates. The latter, remaining superficial to the rapidly growing nerve system, have been drawn away from one another.

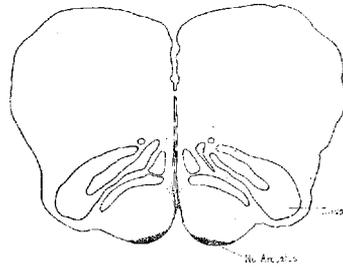
Along the raphe can be found a few cells, the remains of the connecting bridge, and these persist in this position even in the adult. Farther laterally one may often find small isolated masses at almost any point along the periphery of the medulla, the number



10



11



12

Fig. 10 Cross section through the lower olivary region of a 30 mm. fetus. \times 16.5. (No. 86, sl. 28, sect. 13).

Fig. 11 Camera lucida tracing of a cross section through the lower olivary region of a 96 mm. fetus. \times 8.2. (No. 484, sl. 3, row 2, sect. 7).

Fig. 12 Camera lucida tracing of a cross section through the lower olivary region of a 143 mm. fetus. \times 7.3. (No. 508, sl. 3, row 4, sect. 2).

and amount varying with different brains. These represent portions of the basilar nuclei which have not descended to the position of the arcuate nuclei proper. It will be remembered that in No. 508 many of the pontine nuclei are assuming ganglionic

form. This is not true in the arcuate formation where the nuclei are still very densely staining and there is very little protoplasm in the bodies. In No. 509 (188 mm.), however, the protoplasmic body is represented by a clear unstained area around a pale vesicular nucleus. Here it is possible to speak of young ganglion cells with certainty although most of the elements are still undifferentiated.

To conclude, then, we have in the rhombic lip or 'Rautenlippe' of His a common ancestor for the olive, pontine nuclei and arcuate nuclei—the nuclei pontis being formed by a migration through a restricted pathway, the corpus ponto-bulbare; the nuclei arcuati along with part of the olive by a superficial migration over the ventral surface of the medulla.

BIBLIOGRAPHY

- BLAKE, J. A. 1900 The roof and lateral recesses of the fourth ventricle considered morphologically and embryologically. *Jour. Comp. Neur.*, vol. 10.
- ESSICK, C. R. 1907 The corpus ponto-bulbare—a hitherto undescribed nuclear mass in the human hind brain. *Amer. Jour. Anat.*, vol. 7.
1909 On the embryology of the corpus ponto-bulbare and its relation to the development of the pons. *Anat. Rec.*, vol. 3.
- FLECHSIG, P. 1876 *Die Leitungsbahnen in Gehirn und Rückenmark des Menschen.* Leipzig.
- HIS, W. 1891 *Die Entwicklung des menschlichen Rautenhirns vom Ende des ersten bis zum Beginn des dritten Monats.* Leipzig.
1904 *Die Entwicklung des menschlichen Gehirns während der ersten Monate.* Leipzig.
- ORZECOWSKI, K. 1908 Ein Fall von Missbildung des Lateralrecessus. Ein Beitrag zur Onkologie des Kleinhirnbrückenwinkels. *Arbeiten aus dem Neurol. Instit. Wien*, Bd. 14.
- STREETER, G. L. 1904 The development of the cranial and spinal nerves in the occipital region of the human embryo. *Amer. Jour. Anat.*, vol. 4.
1907 On the development of the membranous labyrinth and the acoustic and facial nerves in the human embryo. *Amer. Jour. Anat.*, vol. 6.
1912 The development of the nervous system. *Manual of Human Embryology*, Keibel and Mall, vol. 2.
- TIEDEMANN, F. 1816 *Anatomie und Bildungsgeschichte des Gehirns in Foetus des Menschen.* Nürnberg.