Stage 14 Formation and Closure of Anterior Neuropore

9 Days, 13–20 Somites

Horizon XI
13–20 paired somites

This period could be extended to 22 somites, because the most advanced embryo, with a copulation age of 9 days 2 hours, had 23–24 pairs of somites. In the mouse, the formation of a new somite pair requires 1–2 hours. I have chosen an upper limit of 20 somites for this stage so that it will correspond to Streeter's horizon XI.

External Form

The turning of the embryo is now complete. Compared to human embryos, the mouse embryos of this group are strongly flexed in a dorsally convex direction (Figs. 99 and 108). Furthermore, there is a definite spiral torsion, the posterior end lying on the right side of the head. In rare cases, it may be found on the left side.

The external form is determined in this phase basically by the shape of the neural tube. Anteriorly, the medullary plate is about to close, whereas it is still a flat groove posteriorly. The first two branchial bars are clearly visible. The forelimb bud is not yet clearly delimited. At 15 somites, it appears as a condensation of the lateral plate material, without distinct boundaries (Fig. 100).

Length. The overall length varies considerably, mainly because of varying curvature of the body. If the amnion is cut, the embryo straightens a little. The length is measured from the crown to the curved posterior end in a straight line, and it varies from 1.2–2.5 mm. Externally, the roundish swellings of the uterus measure 3–5 mm. The spaces between the embryos are irregular in vivo, and if the mouse is killed, they are reduced by contraction of the uterus [26].

Circulatory System

In transparent fresh or formalin-fixed embryos, several vessels may be recognized by simple inspection (Fig. 108).

The broad *anterior cardinal vein* receives its blood from the close-meshed network of capillaries investing the neural tube (plexus perineuralis). It joins the posterior cardinal vein to form the *Ductus Cuvieri*.

The (paired) dorsal aorta may be seen immediately ventral to the row of somites. The endocardial tube of the bulbus cordis is very narrow and is separated by a considerable gap from the thick myoepicardium. Anteriorly it forms a right angle with the arterial trunk which is dilated at its end (aortic sac).

Development of the heart. The heart is now capable of maintaining some circulation of the blood. The atrium and ventricle are not yet paired. The shape of the arterial and venous parts of the heart, together with the connecting vessels, are shown in Figs. 109 and 110.

Placental circulation is just being established. The blood already circulates in the yolk sac. The paired dorsal aortae supply the yolk sac by means of a thick vitelline artery (Fig. 111).

This artery will originate later separately from the aorta by a new anastomosis [49], whereas the umbilical artery will remain in direct continuation of the aorta. This transformation is illustrated in Fig. 112.

Intestinal Tract

The original wide opening of the gut into the yolk sac is narrowed to a long, slender *vitelline duct*. As a consequence, fore- and hindgut are no longer represented by separate pockets, but form a continuous tube with blind ends. In the middle, it still opens into the yolk sac.

At 16 somites, the *oral plate* may rupture. At 21 somites only remnants of the membrane remain. The *cloacal membrane*, on the other hand, does not appear as a distinct membrane until the 16-somite stage, and it persists for a long time.

The *foregut* is now differentiating rapidly. The first two branchial clefts have formed (Fig. 113).

The floor of the foregut is thickened, while the dorsal epithelium remains thin. Ventrally, the *thyroid rudiment* evaginates, forming a small groove (Fig. 104). It lies just above the dilated aortic sac. Toward the end of this period, the *lung anlage* (rudiments of larynx-trachea-bronchi) appears as a ventral thickening of the epithelium. There is only a short distance between the lung rudiment and the hepatic diverticulum. This liver primordium develops earlier in mice than in humans and rapidly deepens. The stomach has not yet formed.

The coelom is a single cavity, the cavum pleuro-pericardiaco-peritoneale. In the vicinity of the umbilical ring it communicates with the exocoelom. The coelomic epithelium is thickened above the lung rudiments and in the region of the future stomach [83].

Urogenital System

In this period the somite stalks of the lower cervical and upper thoracic regions are composed of:

1. peritoneal funnel,
2. nephric vesicle; sometimes its lumen is still lacking,
3. nephric duct, as a rule still solid (Fig. 102).

At 15 somites, this typical organization can be observed at the level of the 10th to the 14th somite (KT 987). In another specimen, KT 986, with 16 somites, the pronephric duct has developed a distinct lumen at the level of the 15th somite. The anterior somites, however, are connected to the coelomic epithelium only by disorganized clusters of cells.

Germ cells [103] can be recognized in H.-E. sections for the first time at 15 somites, within the epithelium of the hind gut (KT 987).

Central Nervous System

The formation and closure of the anterior *neuropore* is a most remarkable event. It is accomplished at the same developmental phase as in humans, but seems to be displaced caudally. A comparison of Fig. 75 (7 somites) and 96 (9 somites) reveals an irregular closure of the brain folds. Initially, at 7 somites, they approach each other only posteriorly. Later, they also approach each anteriorly in the forebrain region, while there is still a wide gap in the

Figs. 98–107: Closure of anterior neuropore, 9 days, 15 and 21 somites

FIG. 98. Embryo in yolk sac (*D*) and amnion (*A*).
KT 988, 15 somites, 8 days 21 h. 7:1

FIG. 99. Dissection of embryo from membranes.
O = otic invagination.
KT 988, 15 somites. 11.5:1

FIG. 100. Oblique view of same embryo, showing anterior neuropore (*V.N.*).
Aa = forelimb. 10.5:1

FIG. 101. Cross section through embryo, 8 days 21 h, 15 somites.
Ao = aorta, *Au* = umbilical artery, *H* = hindgut.
KT 987/1. 180:1

FIG. 102. Detail of Fig. 101, with somite 14.
Ao = aorta; *Vn* = pronephric duct, without lumen here; *Pt* = peritoneal funnel. 350:1

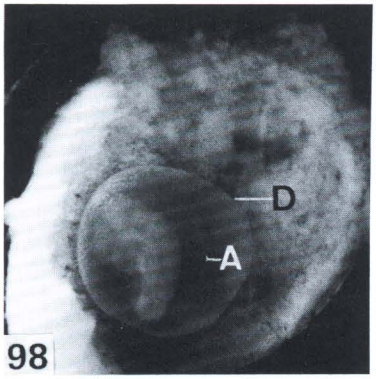
FIG. 103. Cross section through same embryo, 15 somites, at the level of the facial nerve (*N.VII*).
V.c. = vena capitis lateralis, *Ao* = aorta, *V* = foregut.
KT 987/1. 130:1

FIG. 104. Detail of thyreoidea-anlage (*Th*).
Tr = truncus arteriosus (aortic sac).

FIG. 105. Detail of Fig. 106, showing boundary zone of ectoplacental cone.
D = yolk sac, *R* = Reichert's membrane, *T* = trophoblastic giant cells, *All* = allantois. Zone of contact with embryonic vessels, *eG* = ectoplacental glycogen cells, *De* = decidua basalis. 100:1

FIG. 106. Cross section through uterus, with embryo and extraembryonic membranes.
A = amnion, *Au* = umbilical artery in umbilical cord.
KT 624, 9 days, 21 somites. 18:1

FIG. 107. Detail of Fig. 106, showing decidua capsularis and new uterine lumen (*below*).
T = trophoblastic giant cells, forming meshwork; *R* = Reichert's membrane and distal (parietal) layer of yolk sac; *D* = yolk sac, proximal (visceral) layer, with blood vessels. 100:1



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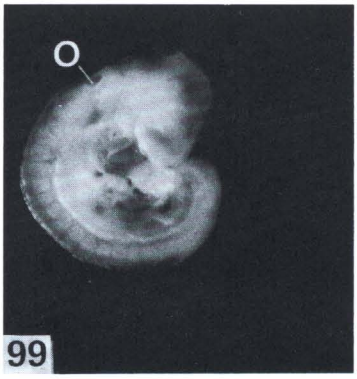

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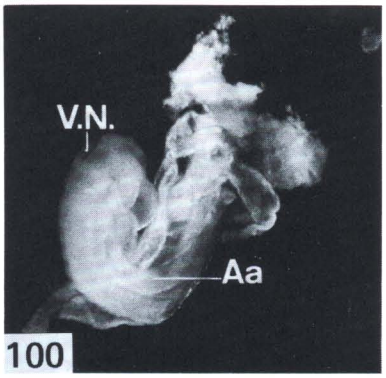

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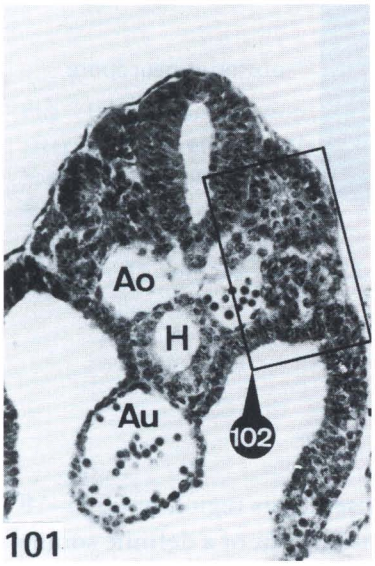

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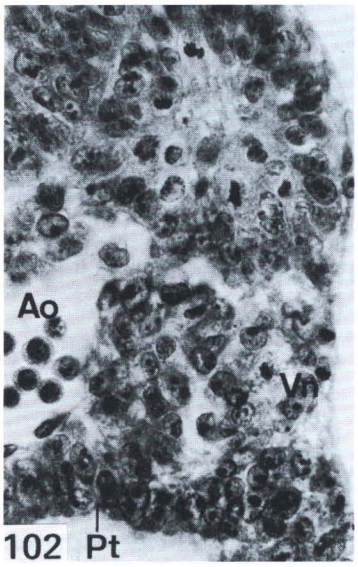

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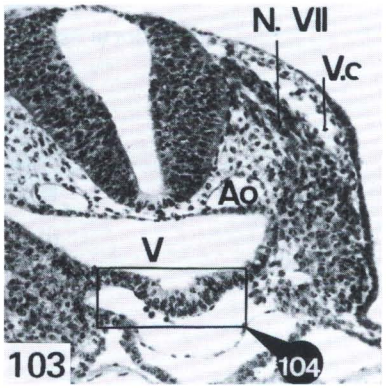

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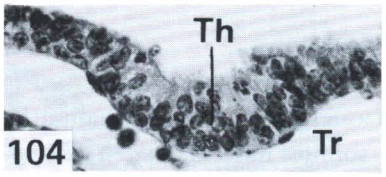

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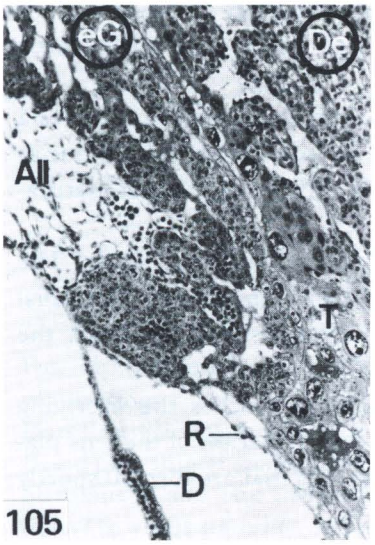

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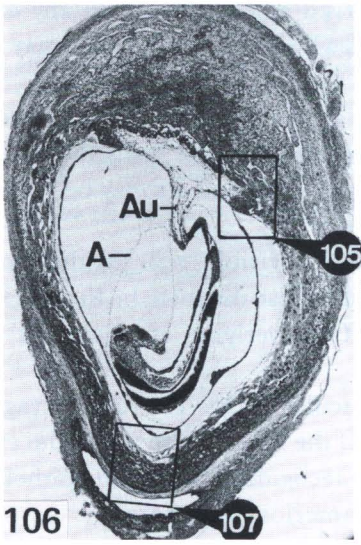

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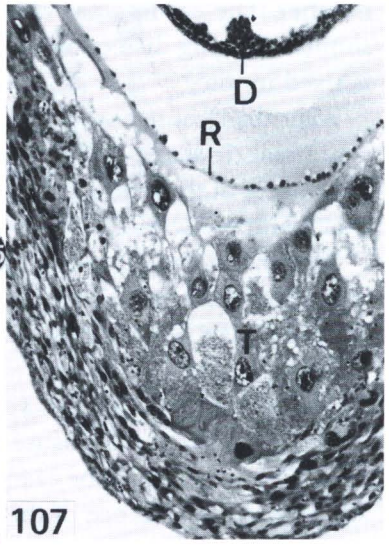

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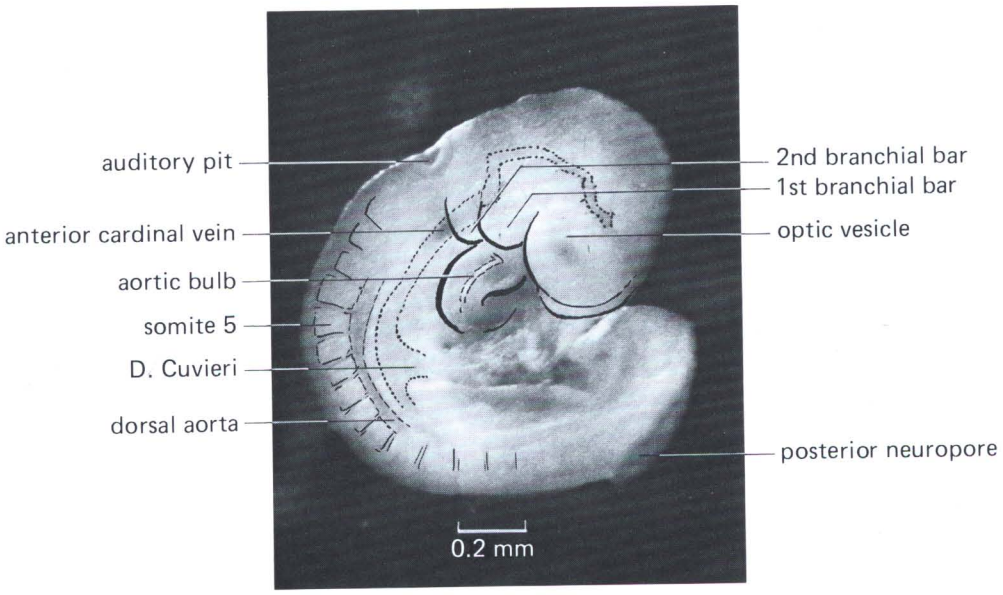

FIG. 108. Embryo, formalin fixed.
KT 937a, 14 somites, 9 days

anterior rhombencephalon (Figs. 97 and 100). They do not close in this region until 15–18 somites have formed. The closure does not coincide with the attainment of a definite somite number. The earliest was observed at 15 somites. In any case, the folds are closed at 19 somites. During dissection, a newly closed neural tube may open again, even if the uterus is fixed in toto.

After the closure of the anterior neuropore, the floor of the rhombencephalon acquires a characteristic shape. Six *rhombomeres* [162] are formed, which will disappear later. Prior to the closure of the neural tube, the rhombomeres may be seen in different phases of maturation (compare with Fig. 82).

The *cranial neural crest* [161] is visible *prior* to the closure of the brain folds. The trigeminal- and facialis crests are most distinct. In Fig. 103, the rudiment of the facialis-ganglion may be recognized. The overlying epidermis is thickened and represents the facialis-placode.

The *eye anlage* is in the vesicular stage. The optic evagination reaches the overlying epidermis prior to the closure of the neural tube and induces the formation of the lens placode. This placode can be easily recognized by the tall epithelial cells that are formed shortly after the disappearance of the anterior neuropore.

The *olfactory placode* appears shortly before the lens placode, as a striking thickening of the epithelium, while the brain tube is still open. At first it is adjacent to the forebrain. Now it begins to be separated from it by invading mesenchyme.

The *otic plate* is transforming into a deep open groove. The invagination proceeds more or less regularly with increasing somite numbers.

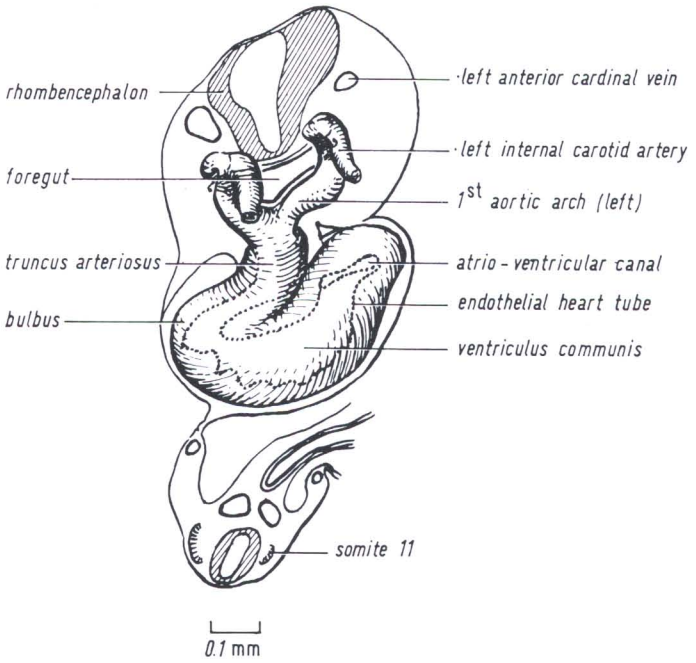

FIG. 109. Reconstruction of the arterial region of the heart.
Ventral view, based on a frontal section through somite 11 and anterior rhombencephalon.
KT 987, 16 somites, 8 days 21 h

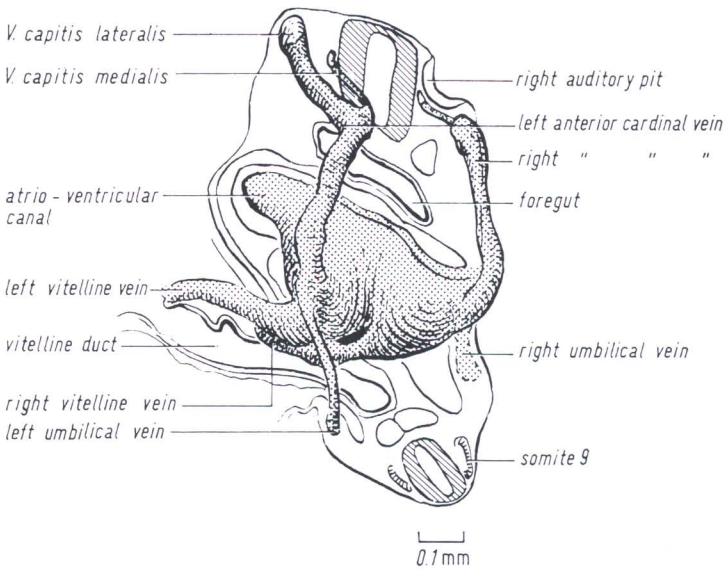

FIG. 110. Reconstruction of the venous sinus.
Dorsal view, from left side. Based on a frontal section, level of somite 9 and otic invagination. The venus sinus lies dorsal to this plane.
KT 987, 16 somites, 8 days 21 h

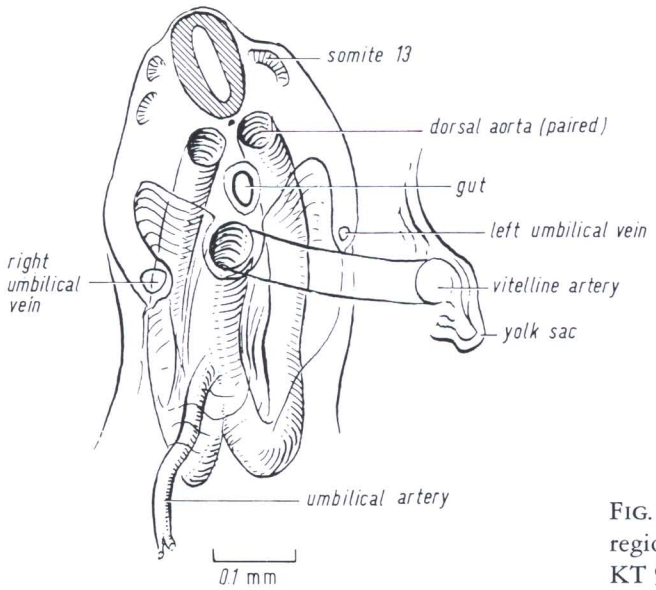

FIG. 111. Vessels of the posterior body region.
KT 987, 16 somites, 8 days 21 h

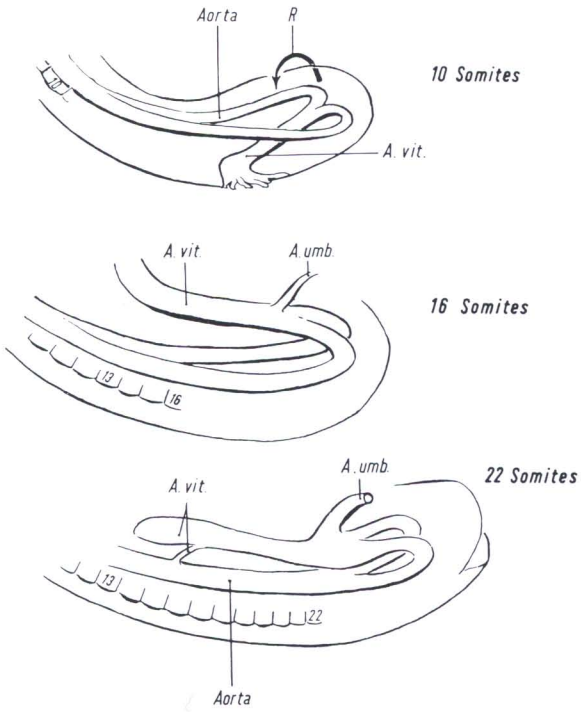

FIG. 112. Development of the posterior arterial stems. At 22 somites, the vitelline artery takes a new origin, by means of an anastomosis. The connection with the umbilical artery is lost later. The *arrow* (*R*) indicates the direction of the embryonic rotation.

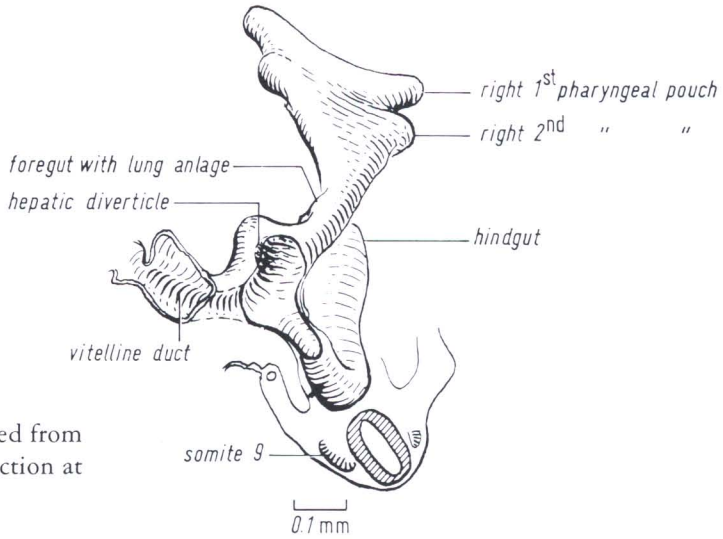

FIG. 113. Anlage of the gut, viewed from dorsal and left, starting from a section at the level of the 9th somite.
8 days 21 h, 16 somites

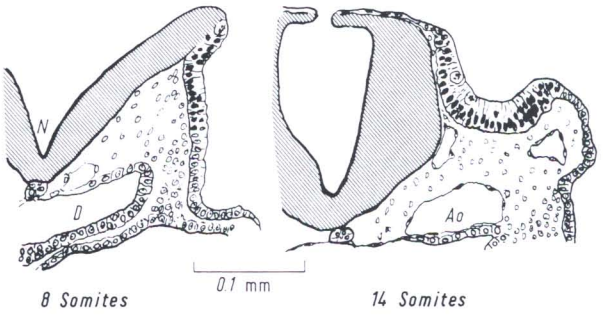

FIG. 114. Development of the otic invagination. Cross sections. The neural tube remains open here for a longer time than in human embryos. *D* = gut-anlage, *Ao* = aorta, *N* = neural groove.

Endometrium and Placenta

The new uterine lumen (Fig. 84) is now continuous (Fig. 106). The adjacent decidua capsularis is poorly vascularized and is composed of relatively small cells. It borders a zone of trophoblastic cells that form a perivitelline meshwork invaded by maternal blood (Fig. 107). Near this zone, the deciduous cells are often multinucleate and are darkly stained.

The *placenta* is developing in the region where the allantois reaches the ectoplacental plate ("chorionic plate"). The ectoplacental cavity disappears by fusion of the two ectoplacental laminae. The fusion begins in the middle and reaches the margins by 8 days (Fig. 77). Toward the maternal tissue, the boundary zone is organized into two different regions:

1. Toward the embryo, there is a rather solid wall of cells, into which the large allantoic vessels enter. They contain some nucleated embryonic erythrocytes (Fig. 105).
   Toward the decidua numerous clefts are developing so that the cell mass is split into anastomosing strands. The clefts are filled with maternal blood. In this way, the *labyrinth* of the placenta is formed.
2. Further toward the decidua basalis there is a zone of ectoplacental glycogen cells that are joined, more peripherally, by single trophoblastic giant cells, in continuation of the peri-

vitelline meshwork (Fig. 105). In contrast to the placental labyrinth, only *maternal blood* is circulating in this junctional zone [38] (trophospongium or reticular zone).

The yolk sac is provided now with a well formed vascular net, and it functions as additional "placenta" (yolk-sac placenta). In the course of dissection, the visceral layer of the yolk sac is immediately exposed after cutting the uterine muscular wall and decidua. There is a cleft between the proximal and distal layer, and the distal layer adheres to the decidua.

Material	Age	Embryos
KT 624–26	9 days	1 with 5 somites
		1 with 11 somites
		1 with 16 somites
		2 with 20 somites
		1 with 21 somites
		1 resorption
KT 985-88	8 days 21 h	1 with 8 somites, anterior neuropore open
		1 with 13 somites, anterior neuropore open
		2 with 14 somites, anterior neuropore open
		1 with 15 somites, anterior neuropore open
		2 with 16 somites, anterior neuropore open
		1 resorption
KT 935–37	9 days 3 h	1 with 15 somites
		2 with 17 somites, anterior neuropore closed
		1 with 20 somites, anterior neuropore closed
		3 with 22 somites, anterior neuropore closed
		1 with 23 somites, anterior neuropore closed
		1 with 24 somites, anterior neuropore closed