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An electron microscopic study of the development of the ependyma of the central canal of the mouse spinal cord

R. R. STURROCK

Department of Anatomy, University of Dundee, Dundee DD1 4HN, Scotland

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INTRODUCTION

The morphology of the adult rodent ependyma in the lateral ventricle was the subject of an extensive study by Westergaard (1970) and the developing ependyma has been described in the lateral ventricle of mice by Hinds & Ruffett (1971) and Shoukimas & Hinds (1978). The morphology of the neonatal rabbit lateral ventricle has been examined by Stensaas & Gilson (1972). Also several investigations of the ependyma of the third (Colmant, 1967; Schachenmayr, 1967; Anand Kumar, 1968; Schwanitz, 1969; Bruni, 1974) and fourth ventricles (Leonhardt, 1967; Fleischhauer & Petrovicky, 1968; Scott *et al.* 1973; Kiss & Mitro, 1978) have been carried out to examine the morphology using a variety of techniques. The ependymal layer of the central canal of the mammalian spinal cord, however, has been largely neglected, especially in the adult. The only description of the ultrastructure of the ependymal cells in the adult mammal appears to be that of Kohno (1969) although Malinsky & Brichova (1967) have described the developing human spinal cord ependyma.

An examination of the central canal of the mouse spinal cord showed that for most of its extent it is lined by cells which appear to be unique to it. These cells exhibit features characteristic of both choroid plexus cells and astrocytes. The present study describes the development of this specialised ependymal epithelium.

MATERIALS AND METHODS

Mouse embryos aged 10, 11, 12, 13, 14, 15, 16, 17 and 18 days post-conception and mice aged 5, 15 and 150 days postnatum were used in the present study. Embryonic mice will be identified by the prefix E (e.g. E10 = 10 days post-conception), and postnatal mice will be identified by the prefix P (e.g. P5 = 5 days postnatum).

Pregnant mice were anaesthetised with Sagatal and the embryos were removed by Caesarean section. Embryos aged E10 and E11 were fixed by immersion in a solution of 2% glutaraldehyde and 2% paraformaldehyde in a 0·05 M-cacodylate buffer. From E12 to E18, the embryos were killed by perfusion-fixation with the same mixed aldehyde solution. The postnatal mice were anaesthetised with Sagatal and killed by perfusion-fixation with a mixture of 2% paraformaldehyde and 3% glutaraldehyde in a 0·165 M phosphate buffer. In postnatal animals, perfusion-fixation was always preceded by a brief flushing out of the vascular system with physiological saline.

The embryos were left overnight in fixative and then small segments of cord at the level of the upper limb buds and lower limb buds were dissected out, processed and embedded in Spurr's resin as described previously (Sturrock, 1979*a*). At E18, P5, P15 and P150, the spinal cords were dissected out and small blocks cut from the

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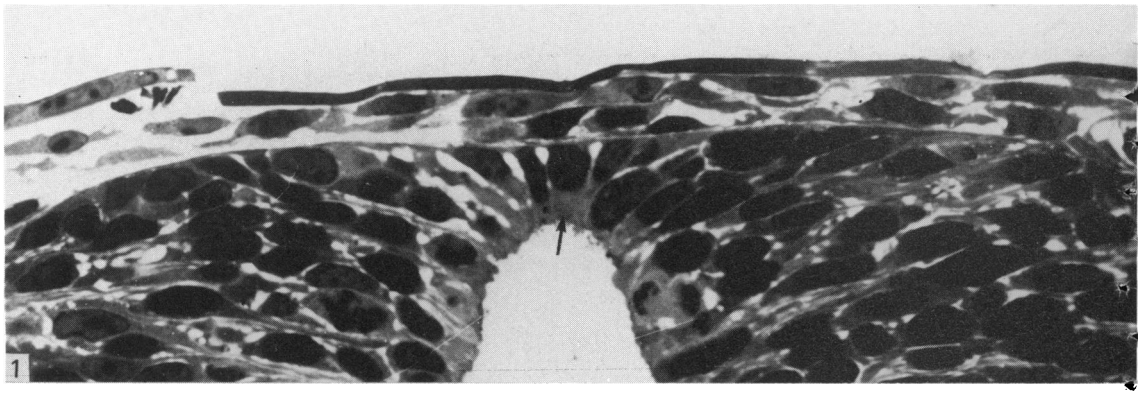

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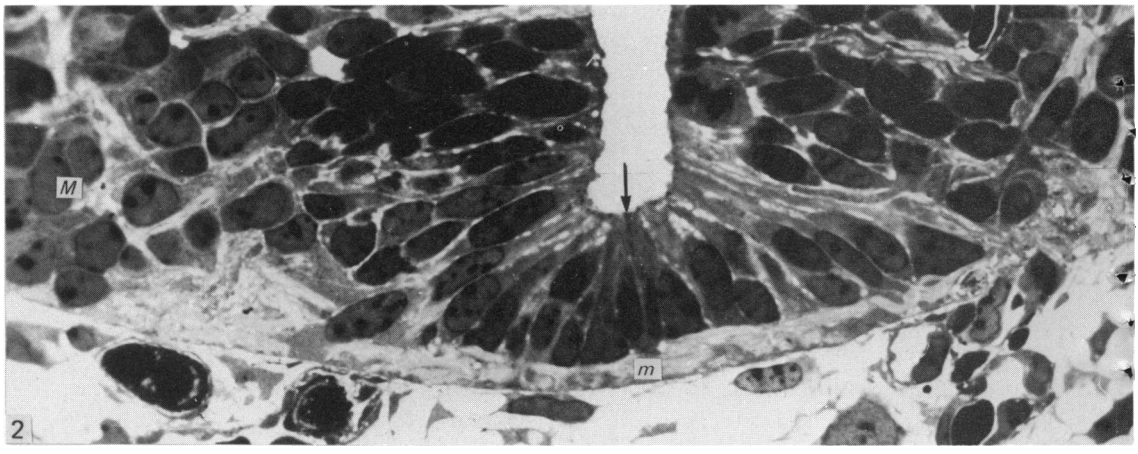

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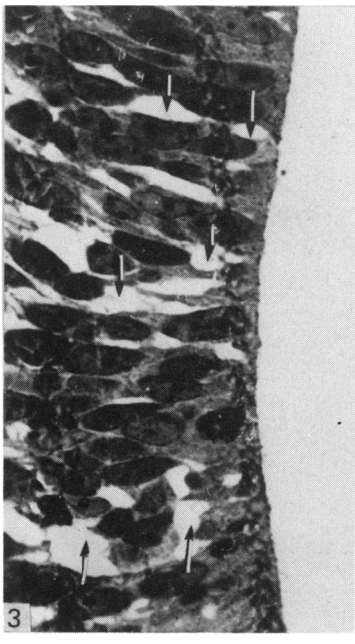

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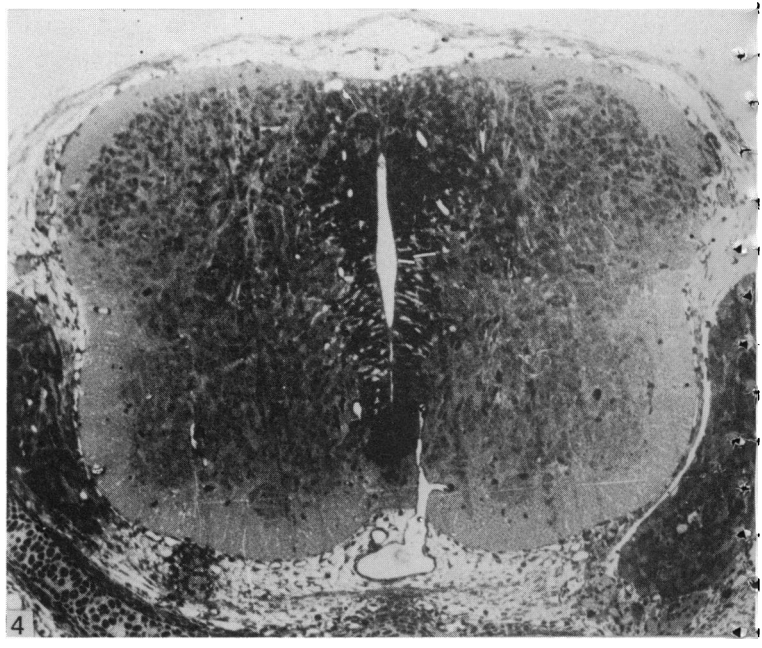

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cervical, thoracic, lumbar and sacral regions. These were processed in a similar fashion to the earlier embryonic spinal cords.

Semithin sections (1 μm) of spinal cord stained with 1 % toluidine blue were obtained at each age and the blocks were trimmed to include the complete central canal. Ultrathin sections stained with uranyl acetate and lead citrate were examined in an AE1 801 transmission electron microscope.

OBSERVATIONS

At E10 and E11, the spinal cord consisted of a thick pseudostratified columnar epithelium except in the region of the roof and floor plates which were made up of a simple columnar epithelium (Figs. 1, 2). A mantle layer was present ventrolaterally (Fig. 2) and a marginal layer was apparent laterally and ventrally. At E12 the marginal layer had extended around most of the cord but was still deficient over the roof plate. In the region of the floor plate the marginal layer appeared to be largely formed by transversely running axons (Fig. 26) which mingled with the dorsoventrally running ependymoglial fibres.

At E13 the ventrolateral part of the ependymal layer was much thinner, being only 2–3 nuclei deep in most places. Dorsolaterally the ependymal layer was 4–5 nuclei deep. The walls of the canal were in apposition both dorsally and ventrally, the canal being widest midway along its dorsoventral axis. By E14 the posterior columns had appeared. The ependymal layer was thickest in its mid-portion. The dorsal and ventral parts still retained their connections with the pial surface, but this was obvious only dorsally since the ventral fibres were obscured by the transversely running commissural fibres. In the mid-portion large extracellular spaces appeared between the ependymal cells (Fig. 3). Ventrally the lateral walls were coming to lie in apposition (Fig. 4).

By E15 the walls of the dorsal part of the canal had fused. The dorsal surface was formed by fine processes of very elongated columnar cells whose peripheral processes formed the dorsal median septum. The walls of the ventral part of the central canal were in apposition but not fused. The ventral slit-like part of the canal does not begin to fuse until E17. At E18 fibre bundles could be seen both ventrally and dorsally (Fig. 5). The ventral bundle soon became lost in the neuropil, but the dorsal bundle could be traced by way of the dorsal median septum to the pial surface.

At P5 the dorsal median septum was a prominent feature, but it was not possible, due to growth of the neuropil, to trace it to the central canal. At P15 and P150 the

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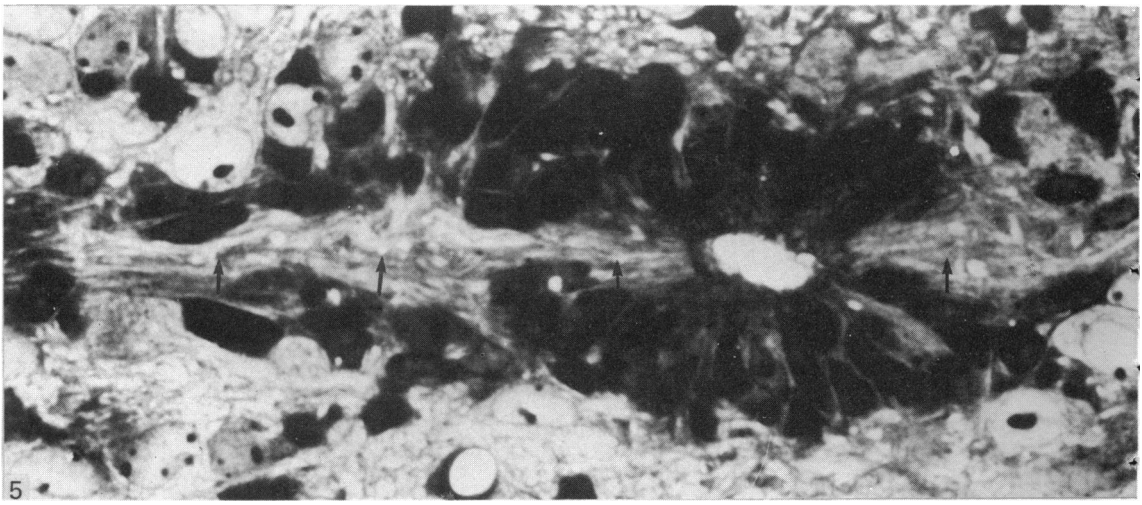
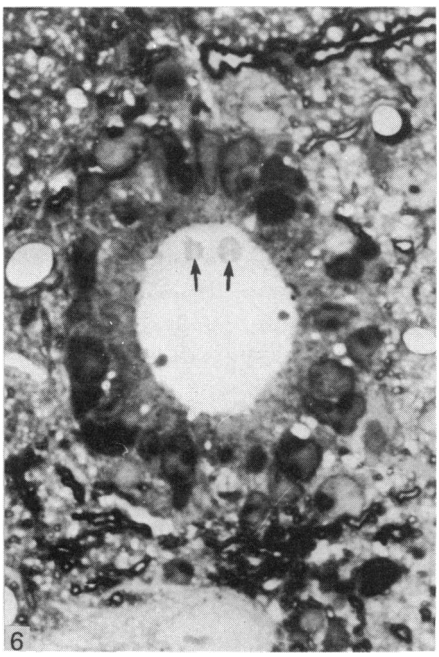
Fig. 1. 1 μm section, stained with toluidine blue, of an E11 mouse thoracic spinal cord. The arrow indicates the simple columnar epithelium of the roof plate. The ependymal layer ventrolateral to the roof plate is a pseudostratified columnar epithelium. There is no mantle or marginal layer dorsally at this age. × 800.

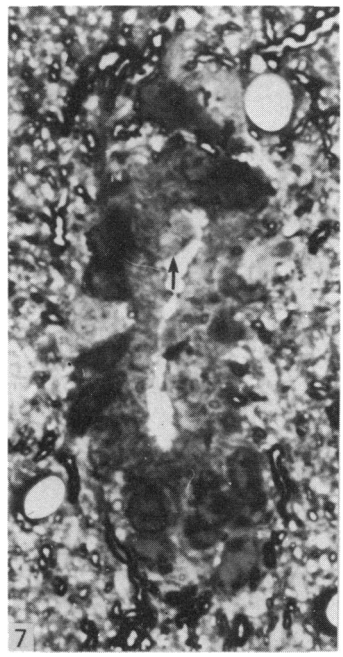
Fig. 2. 1 μm section, toluidine blue stained, of an E11 mouse embryo showing the floor plate region (arrow). This is part of the same section shown in Fig. 1. Note that ventrally the beginning of the marginal layer (*m*) is visible and a mantle layer (*M*) of differentiating neurons is also present. × 800.

Fig. 3. This shows the presence of large extracellular spaces in the E14 spinal cord ependyma (arrows). 1 μm section. × 800.

Fig. 4. At E14 the central canal walls are in apposition ventrally and are fairly narrow dorsally. The area indicated by the arrow is the region shown in high power in Fig. 3. Even at this magnification the extracellular spaces in the ependyma are a prominent feature at this age. 1 μm section. × 100.







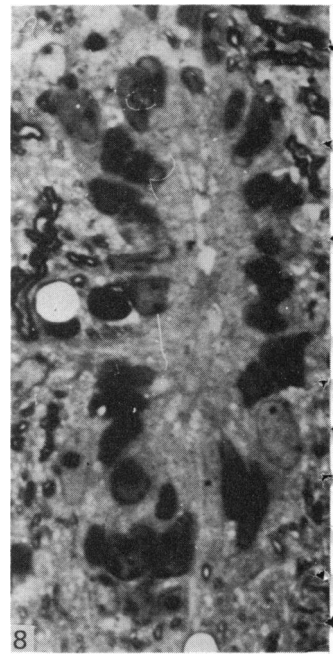


Fig. 5. Central canal and surrounding area at E18. The dorsal part is on the left. Ependymoglial fibres can be seen streaming in bundles both dorsally and ventrally (arrows). 1 μm section. $\times$1250.

Fig. 6. Adult upper cervical region at P150. The central canal in this region is circular and patent. The pale circular objects within the canal may be axon terminals. 1 μm section. $\times$1250.

Fig. 7. Central canal from thoracic cord at P150. The narrow slit-like canal has an obvious lumen but contains some darkly staining material (arrow). 1 μm section. $\times$1250.

Fig. 8. Central canal from sacral region at P150. The canal has only a very narrow lumen. 1 μm section. $\times$1250.

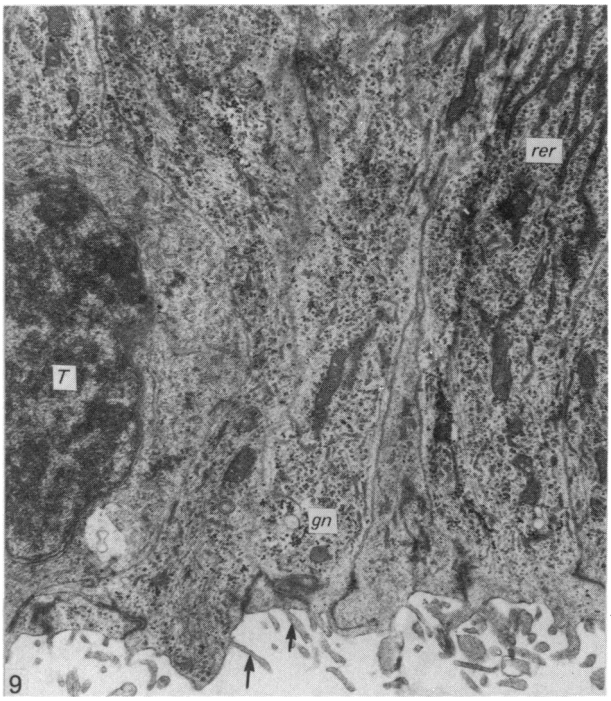
posterior columns had greatly increased in size as a result of myelination. The dorsal median septum was easily identifiable between the corticospinal tracts but was not evident in most regions between the gracile tracts. Postnatally the central canal in most regions was slit-like with very few spaces evident. In one adult animal the upper cervical canal was circular and patent, but more caudally had become slit-like (Figs. 6–8).

At E10 and E11 the ependymal cells resembled those described in other regions of the neural epithelium (e.g. Hinds & Ruffett, 1971). By E11, however, the floor plate showed some minor differences as a few scattered microvilli had begun to appear in this region. At E12 the floor plate differed markedly from other regions. The surface of the floor plate had numerous fine microvilli and the majority of cell processes contained glycogen granules (Fig. 9). Some cells contained long strands of rough endoplasmic reticulum filled with an amorphous material. In the ventral part of the lateral wall the cells appeared to be broader, with their nuclei closer to the central canal (Fig. 10). Few microvilli were present in the ventral half, but some glycogen-containing cells were observed (Fig. 11). In the dorsal half the cells were narrower and had numerous microvilli, as well as broader processes, but glycogen granules were not a feature of this region. The roof plate contained cilia as well as a few microvilli (Fig. 12). With increasing age the roof plate developed microvilli and glycogen-granules and by E15 glycogen granules and microvilli were found along the lateral walls (Fig. 13). The main difference in morphology at E15 between the roof and floor plates on the one hand and the lateral walls on the other was that the former consisted of long narrow columnar cells whereas the latter were made up of a low columnar epithelium.

At E16 the whole central canal was formed by ciliated columnar epithelial cells containing scattered glycogen granules. Cilia were occasionally seen (Fig. 14). At this stage the ependyma appeared to be differentiating into a fairly standard ependymal epithelium as described in other regions of the ventricular system but at E17 the features of the epithelium changed quite markedly. The villi, instead of being fairly narrow, scattered structures, became tangled and club-shaped and formed a matted surface similar to that seen on the surface of the mouse choroid plexus (Sturrock, 1979*a*). The canal itself appeared full of debris and contained interconnected spaces in which the remains of debris could be seen (Fig. 15). Ventrally, where the walls were fusing, an amorphous ground material studded with glycogen granules was present (Fig. 15). At E18 the microvilli layer became thicker and long cilia extended into the canal which now contained regions of amorphous ground substances of varying electron density studded with glycogen granules. The ependymal cells contained varying amounts of glycogen (Fig. 16). Empty membrane-bound spaces were also observed. Bulbous nerve endings were also seen in the canal (Fig. 16).

By P5 the ependymal cells contained thick bundles of microfilaments as well as glycogen granules (Fig. 17). Rarely these cells could be seen undergoing mitosis (Fig. 18). The canal was largely filled with areas of light and dark amorphous ground substance separated by membranes. The dark ground substance was much richer in glycogen than the light. Cilia were frequently embedded in this material (Fig. 19). Bulbous axon terminals could be traced from the canal (Fig. 20) particularly in the ventral and dorsal regions.

The ependymal cells in the adult frequently contained lipid droplets (Fig. 22). In the upper cervical region the ultrastructure of the ependyma resembled that of the



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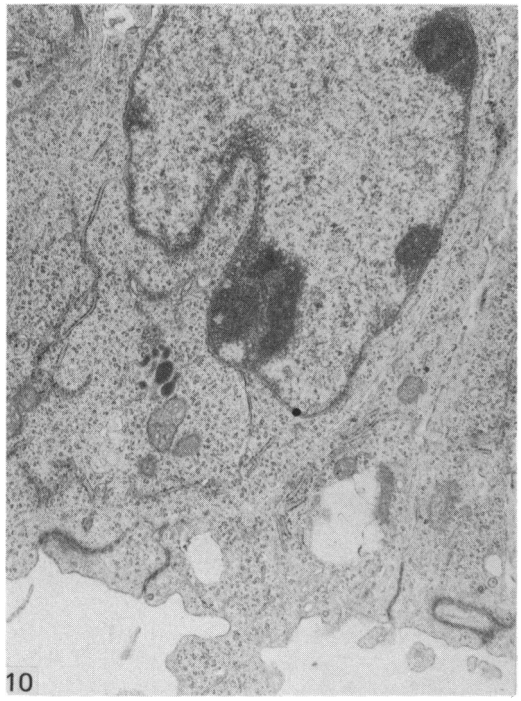
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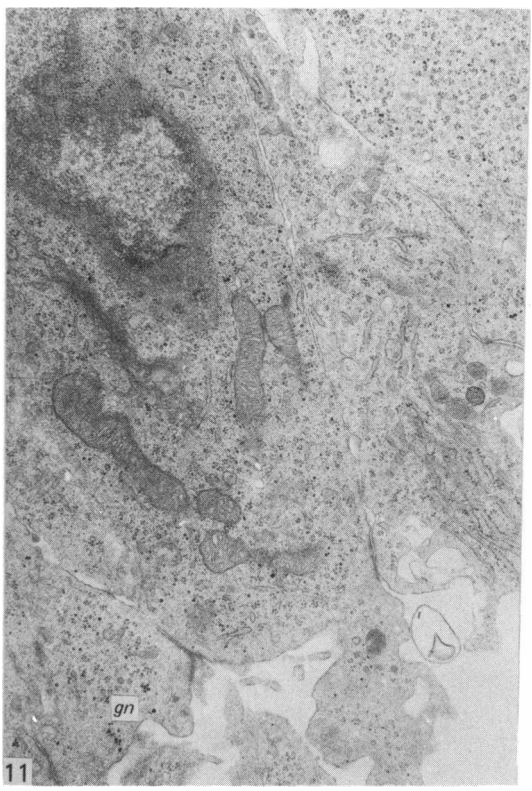

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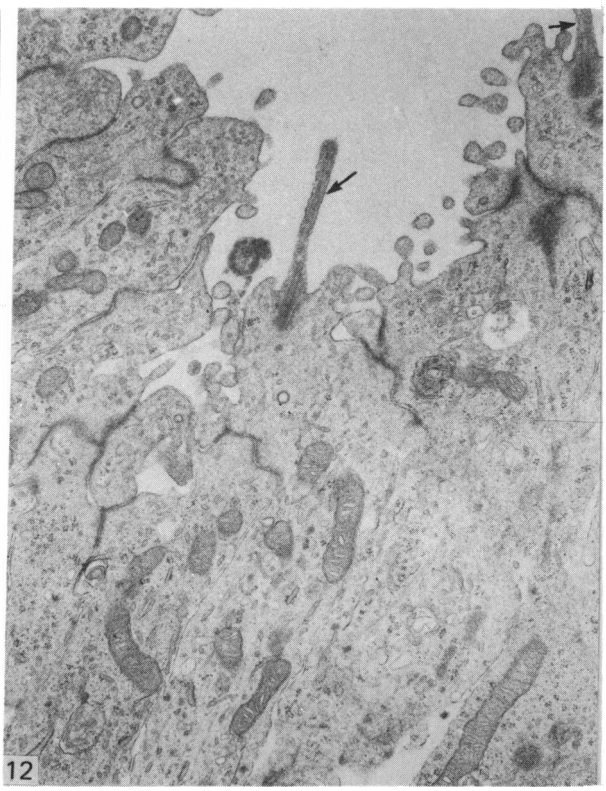
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lateral ventricle with small, narrow microvilli and cilia (Fig. 22). In this region the canal contained only an occasional very small clump of amorphous material. In most of the cervical canal the morphology was similar to that of the more caudal regions. The cells were columnar and similar to those found at P5 except for a reduction in the amount of glycogen, which was present only as sparsely scattered granules. The central canal was filled with similar material to that observed at P5 (Fig. 21). In regions where the canal was slit-like in shape the microvilli in the dorsal and ventral regions were up to 2 μm in length.

In view of the continuity of the cells of the floor and roof plates with the pia, it may be worth recording that the pial surface of the dorsal median septum and the similar region ventrally exhibited certain features which were not found elsewhere around the periphery of the cord. In younger embryos the surface was extremely convoluted with frond-like cytoplasmic processes (Figs. 23–25) similar to those described by Gamble (1971) in the filum terminale of the human fetus. Areas extremely rich in glycogen were occasionally present in the subpial cytoplasmic processes (Figs. 23, 24). The fimbria-like processes were still present dorsally at P5 and the fact that each process was covered with basement membrane suggests these are not artefacts (Fig. 25).

The peripheral processes in the roof and floor regions in the mid-line were rich in glycogen as early as E12 (Fig. 26) and the development of these processes up to adult life is shown in Figures 26–29. In the adult the processes forming the dorsal median septum were extremely rich in microfibrils (Fig. 29) in the region of the cortico-spinal tracts but were found only intermittently dorsal to this.

DISCUSSION

The distribution of ependymoglial fibres in the embryonic chick spinal cord was described by Ramon y Cajal (1890) and his diagram of newborn mouse spinal cord (1952) shows the persistence of ependymoglial cells particularly in the dorsal median septum and in the mid-line ventrally.

The present study casts some light on differentiation of the ependymoglial cells since they appear to form virtually all of the roof and floor plates. The floor plate differentiates before the roof plate and at E11 already has peripheral processes which pass through the developing anterior white commissure to the pial surface, while at this stage the roof plate consists of a simple columnar epithelium from the canal surface to the pial surface. At this stage the floor plate cells, unlike other central

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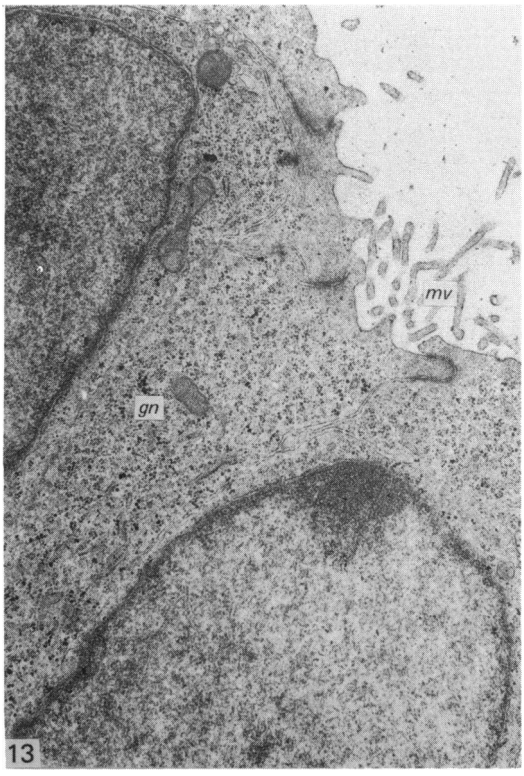
Fig. 9. Electron micrograph of the ependymal epithelium of the floor plate at E12. Most of the cell processes contain glycogen (*gn*), and the rough endoplasmic reticulum (*rer*) of the glycogen-containing cells is arranged in long strands and is filled with an amorphous material. Microvilli (arrows) are common. A telophase nucleus (*T*) is visible. × 12600.

Fig. 10. Electron micrograph showing that the ventral part of the lateral walls of the central canal at E12 is formed of cells whose nucleus is closer to the apical surface than is the case in the floor plate. These cells contain less rough endoplasmic reticulum and no glycogen granules but numerous free ribosomes are present. Microvilli are sparse. × 12600.

Fig. 11. Electron micrograph showing that some glycogen-containing cells were present in the ventral part of the lateral wall (*gn*) but these cells were in the minority at E12. × 12600.

Fig. 12. At E12 the roof plate is very narrow. This electron micrograph shows that the processes reaching the apical surface do not contain glycogen but cilia (arrows) are common. Microvilli are broad and more frequent than in the ventral part of the lateral wall. × 12600.



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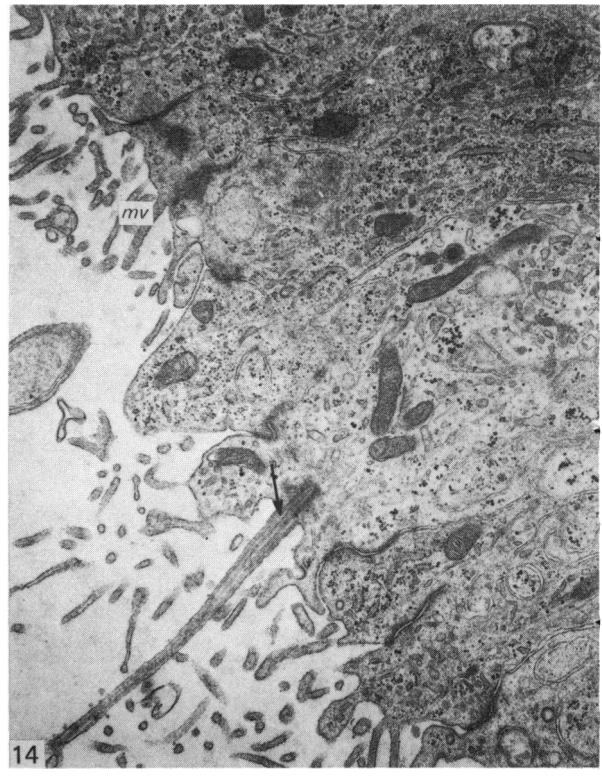

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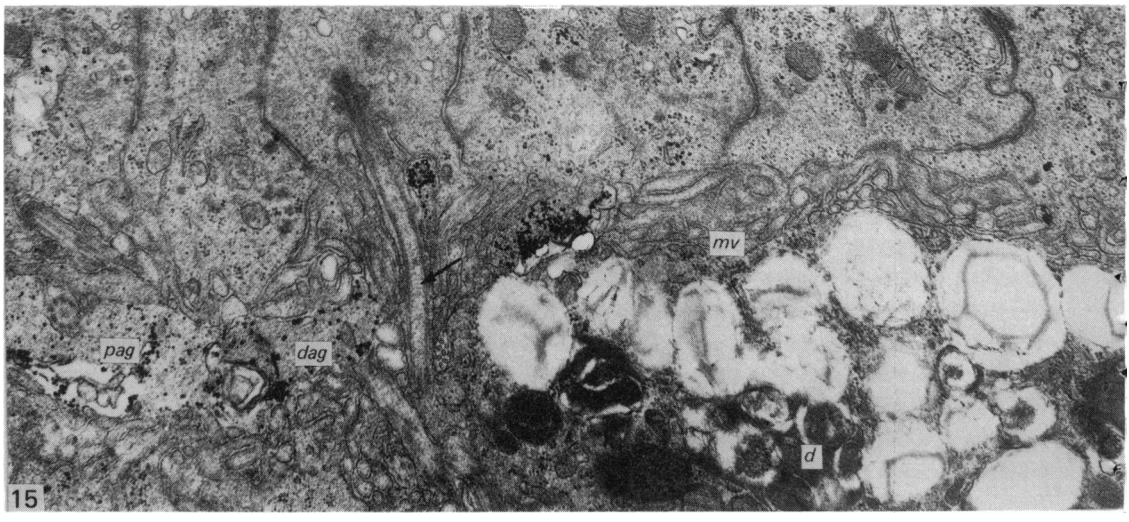

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canal ependymal cells, have a few narrow microvilli. By E12 the floor plate cells have undergone a considerable degree of differentiation and contain long strands of rough endoplasmic reticulum and numerous glycogen granules. In other parts of the central canal there are few glycogen-containing cells and the majority of ependymal cells are undifferentiated cells with few organelles except for clusters of free ribosomes. By E13 roof and floor plates are made up of glycogen-containing ependymoglial cells and by E16 the whole canal is lined with glycogen-containing cells with relatively numerous organelles. It seems safe to conclude that the glycogen-containing cells are ependymoglial cells and their peripheral processes resemble those tentatively described as ependymoglial guide fibres in the forebrain (Sturrock & Smart, 1980). They are also similar to those previously described in the embryonic mouse spinal cord (Henrikson & Vaughn, 1974).

The distribution of ependymoglial cells and undifferentiated ependymal cells corresponds with the distribution of mitotic activity as described by Smart (1972) with the majority of undifferentiated cells being present in sites of higher mitotic activity.

Since the roof plate cells at E11 are identical to undifferentiated ependymal cells found along the lateral walls it seems certain that undifferentiated cells give rise to ependymoglial cells as well as neurons. Only a small proportion of undifferentiated cells eventually differentiate into ependymoglial cells or true ependymal cells and this is one of the mechanisms whereby the central canal reduces in size during development. There is also some degree of fusion of the walls both dorsally, which occurs first, and ventrally, which occurs later.

The transient presence of large extracellular spaces between the ependymal cells at E14 is worthy of comment. By E14 ependymal mitosis has largely tapered off (Smart, 1972) and the undifferentiated ependymal cells are gradually becoming fewer and fewer. At the same time the pattern of vascularisation of the cord is undergoing a major change (Sturrock, unpublished) with a regression of ependymal blood vessels and the onset of a rapid increase in the vascularity of the grey matter. Large ependymal extracellular spaces have been observed in the neonatal rabbit lateral ventricle (Stensaas & Gilson, 1972) and in the mouse lateral ventricle from E17 until shortly after birth (Sturrock & Smart, 1980). It is almost certain that these spaces are artefactual (Sturrock & Smart, 1980) but they probably do represent some change, perhaps in permeability of the ependyma. In the mouse forebrain, ependymal mitosis has declined greatly by E17 (Sturrock, 1979*c*) and the large embryonic ependymal blood supply is also undergoing regression. At E14 the mouse spinal cord ependyma is therefore at a similar stage of development as the perinatal mouse fore-

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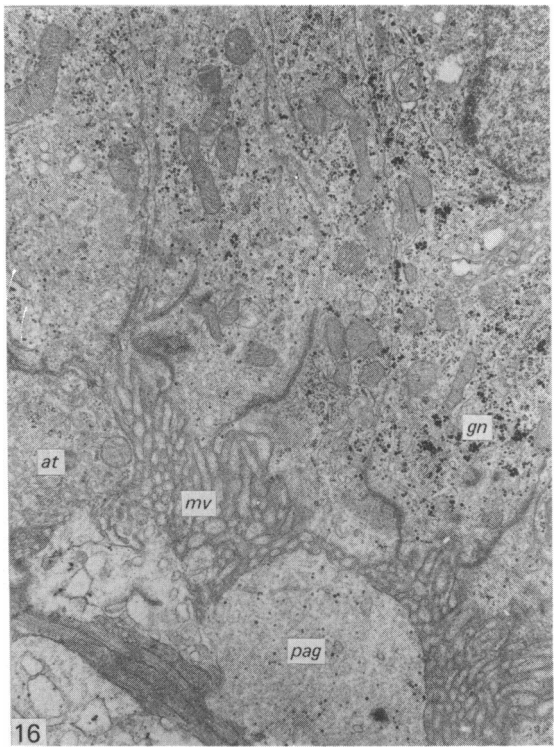
Fig. 13. Electron micrograph from cord at E15. All the cells lining the canal contained glycogen (*gn*). The lateral walls were largely composed of a simple columnar/cuboidal epithelium. Microvilli (*mv*) were common. × 12600.

Fig. 14. Electron micrograph from cord at E16 where microvilli (*mv*) appeared to have increased in number. In the roof plate cilia (arrow) were often present and usually the cells with a cilium had a paler cytoplasm and were particularly rich in glycogen. × 12600.

Fig. 15. Electron micrograph from cord at E17. The microvilli had changed in character. They were broad and tangled (*mv*) and covered the whole canal surface of the ependyma. Cilia (arrow) were much more frequently observed than in younger animals. The canal was filled with debris (*d*) and vacuolar spaces, probably artefactual. Glycogen-containing material was present in the canal in two forms, one with a pale amorphous ground substance (*pag*) with sparse glycogen, and the other with a dark amorphous ground substance (*dag*). These two types of material were found throughout almost the whole central canal from E17 onwards. × 18900.



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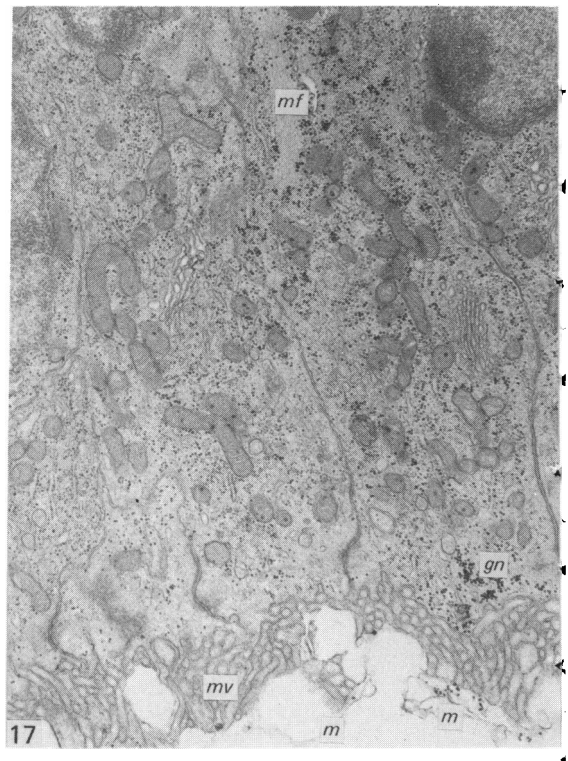

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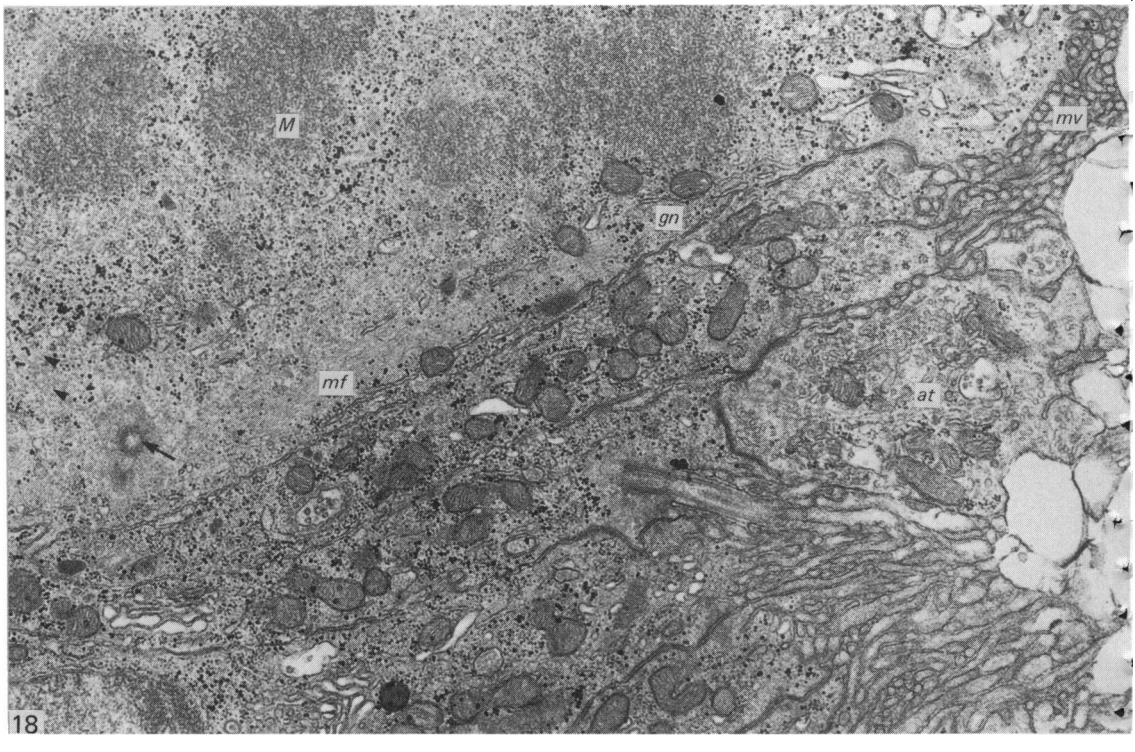

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brain, with the difference that in the spinal cord the ependymal extracellular spaces are present for only one day in contrast to the forebrain where the extracellular spaces are present for several days. This is probably a reflection of the much more rapid development of the spinal cord.

Between E16 and E17 the canal becomes filled with glycogen-containing material and fusion of the ventral walls occurs, reducing the canal to its final size. The glycogen-rich amorphous material which fills most of the central canal from E17 is identical to the material found between the walls of the mouse lateral ventricle immediately prior to their fusion (Sturrock, 1979*b*). Although this material was present in the ventral part of the canal before fusion, it was absent before and during fusion of the dorsal walls, so the earlier suggestion (Sturrock, 1979*b*) that this material might act as a form of glue is unlikely, particularly since there is no evidence of any further fusion of the walls of the central canal after E17 despite the canal being full of the glycogen-containing material. As in the lateral ventricle, the material in the canal exists in two forms; one with a pale ground substance and relatively few glycogen granules; the other with a dark ground substance, rich in glycogen. Each type is separated from the other by a membrane. The function of this material is unknown. Its presence in the central canal and in the narrow space between the fusing walls of the lateral ventricle could be the result of this substance simply being a CSF precipitate which tends to accumulate in parts of the ventricular system where circulation of CSF is sluggish. Conversely it might be a secretion of the choroid plexus which is rich in glycogen, especially during development (Goldmann, 1913; Kappers, 1958; Tennyson & Pappas, 1962; Sturrock, 1979*a*). The glycogen-rich material is found in all parts of the central canal lined by ependymal cells with a choroid plexus-like microvillous surface.

Club-shaped microvilli first appear in the canal surface of the ependyma at E17, the same age at which glycogen-rich material is first found filling the central canal. Similar club-shaped microvilli were described in the human fetal spinal cord by Gamble (1969). The only region from which the glycogen-rich material was absent was the upper cervical region and there the ependymal cells do not possess club-shaped microvilli. The relationship between the material and the microvilli is unknown. It may be that the ependymal cells differentiate into choroid plexus-like cells in order to secrete the material or the microvilli may differentiate as a response to being in contact with the material. The former explanation seems the more likely since the ependymal cells are rich in glycogen.

If club-shaped microvilli are one characteristic feature of adult central canal ependyma, the other is the presence of masses of microfibrils such as are normally found in astrocytes. The microfibrils were first observed at P5 and at this stage the

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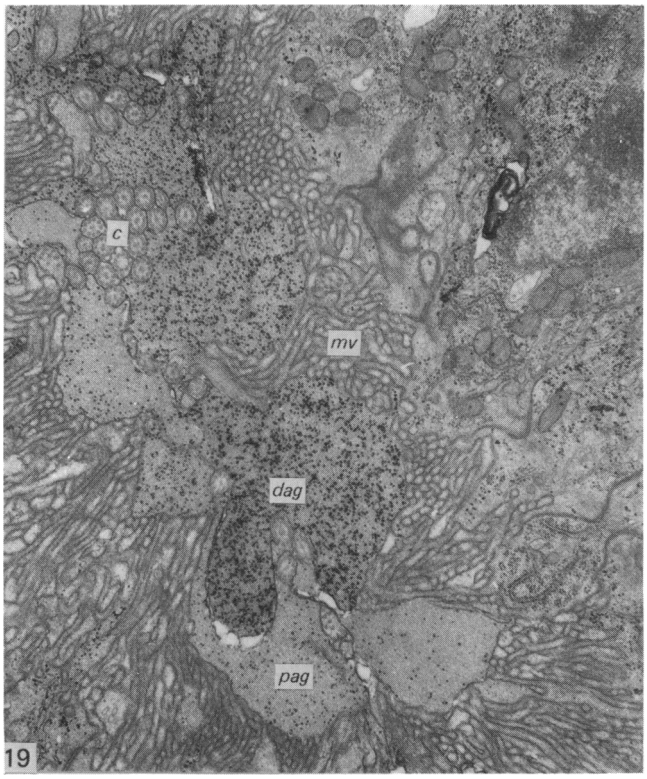
Fig. 16. Electron micrograph from cord at E18. The ependymal cells are rich in glycogen (*gn*). The matted microvilli (*mv*) line the canal which is full of amorphous ground substance (*pag*). An axon terminal (*at*) can be seen in this section. × 12600.

Fig. 17. Electron micrograph from cord at P5. As well as glycogen (*gn*), the ependymal cells contain bundles of microfilaments (*mf*). Microvilli (*mv*) line the canal. In this section the canal appears empty, but fine membranes (*m*) can be seen which suggests that the amorphous material has been lost during processing. × 12600.

Fig. 18. Electron micrograph from cord at P5. Mitotic figures are rare after E15 but a fully differentiated cell containing microfibrils (*mf*), glycogen (*gn*) and microvilli (*mv*) was observed undergoing mitosis. A centriole (arrow) was present. × 10000.



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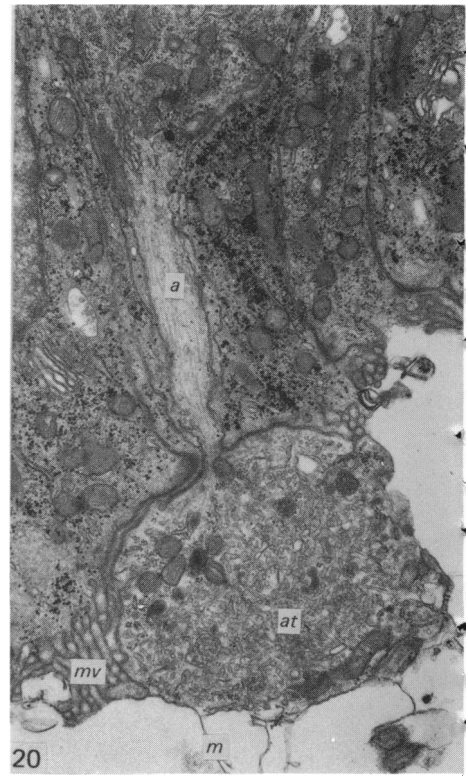

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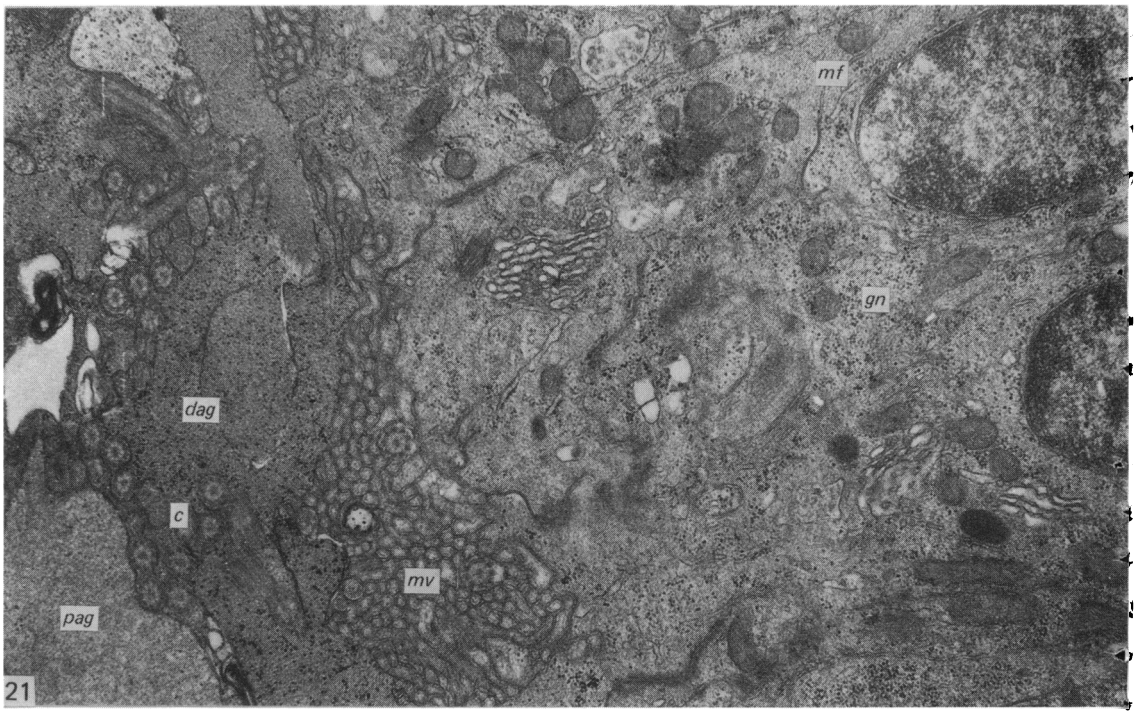

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central canal ependymal cells are capable of undergoing mitosis. The microfibrils are present in very large amounts, probably even exceeding the amounts usually found in mouse fibrous astrocytes. During development of the central canal ependymal cells, glycogen is present in very large quantities before and immediately after birth but by P150 only sparsely scattered glycogen granules are present. This is similar to the distribution of glycogen granules in mouse forebrain astrocytes during development (Sturrock, 1974; 1976), as well as in mouse choroid plexus cells (Sturrock, 1979*a*).

Until the spinal cord has become myelinated, the pial surface formed by ependymoglial processes from the roof and floor plates differs from the pial surface elsewhere due to the presence of frond-like processes. At first it seemed that these might be artefacts, perhaps caused by pulling off the large vessels in these regions during processing; but since the basement membrane surrounds each process, this seems unlikely. In embryos the processes in these areas are occasionally rich in glycogen. The significance of the differences found in these areas is not known but, in the urodele, ependymoglial processes also end in a similar irregular fashion (Schonbach, 1969).

It is not possible to state from the present study whether ependymoglial cells in the mouse spinal cord retain contact with the pial surface throughout life. The dorsal median system appears incomplete between the two gracile fasciculi. This is most probably due to stretching of the septum with growth during myelination, leading to the septum becoming narrower, and in some places, absent. Astrocytes undoubtedly contribute to the formation of the dorsal median septum but the quantity of microfibrils in the ependymal cells suggests that at least some true ependymoglial cells remain. The postnatal ependymal cells are rich in microfibrils, much more so than fibrous astrocytes in the mouse.

The presence of bulbous axon terminals has been noted in the lateral ventricle (Westergaard, 1970), in the third ventricle (Scott, Kozlowski & Sheridan, 1974) and in the fourth ventricle (Leonhardt, 1967) and they have been considered to be neurosecretory terminals. It is perhaps surprising that there should be neurosecretory endings in regions of the central canal where the lumen is filled with electron-dense material. This leads to the question of how much, if any, CSF circulation occurs in the central canal once it has become filled with this material and if CSF circulation does not occur how much diffusion can occur through the amorphous material?

In conclusion it seems that early development and differentiation of central canal ependyma are similar to that in other parts of the ventricular system although ependymoglial cells are more prominent. From E17, however, the majority of central

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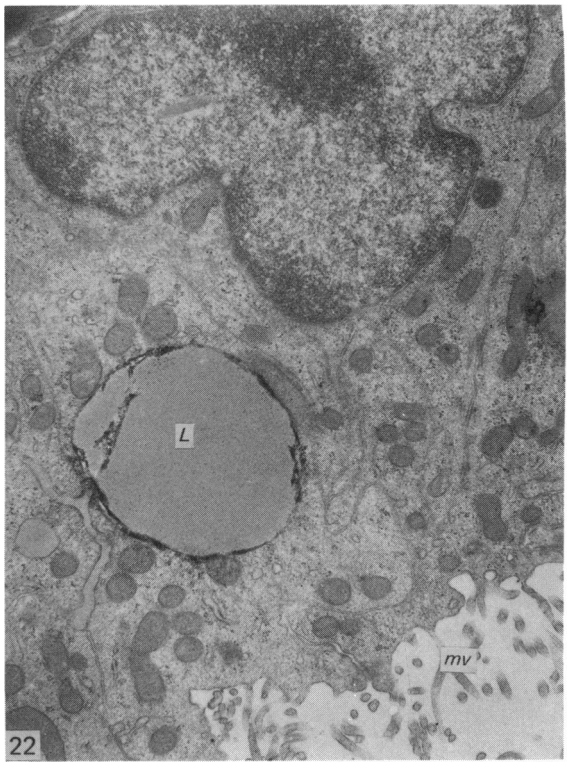
Fig. 19. Electron micrograph from cervical cord at P5. This section shows the central canal filled with pale (*pag*) and dark (*dag*) amorphous material. Microvilli (*mv*) are numerous and bundles of cilia (*c*) can be seen × 8000.

Fig. 20. Electron micrograph from cord at P5. An axon (*a*) can be seen passing between ependymal cells to end in a large axon terminal (*at*) within the central canal. The axon terminal contains mitochondria and numerous vesicles. It is attached to the ependymal cells by junctional complexes. *mv*, microvilli. × 12600.

Fig. 21. Electron micrograph from cord at P150. The ependymal cells contain microfilaments (*mf*) but glycogen (*gn*) granules are much sparser than at P5. Microvilli (*mv*) are similar to those seen from E17 onwards and bundles of cilia (*c*) lie amongst membrane-bound masses of pale (*pag*) and dark (*dag*) material. Sacral cord. × 12600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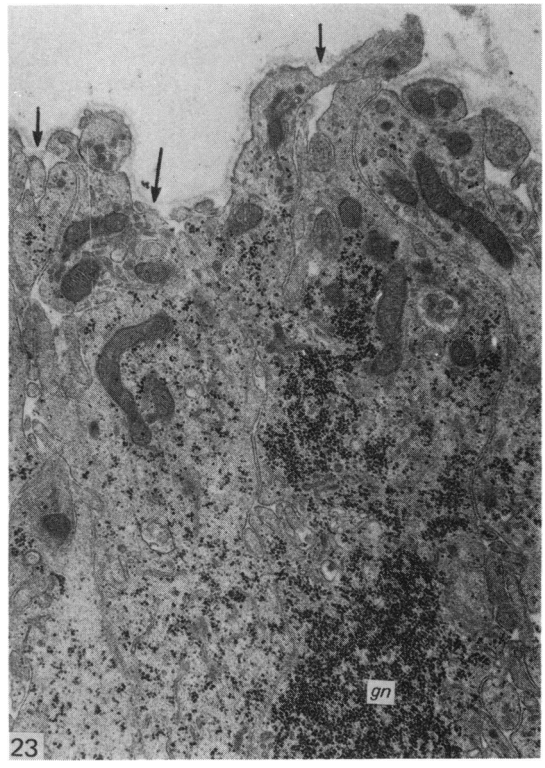
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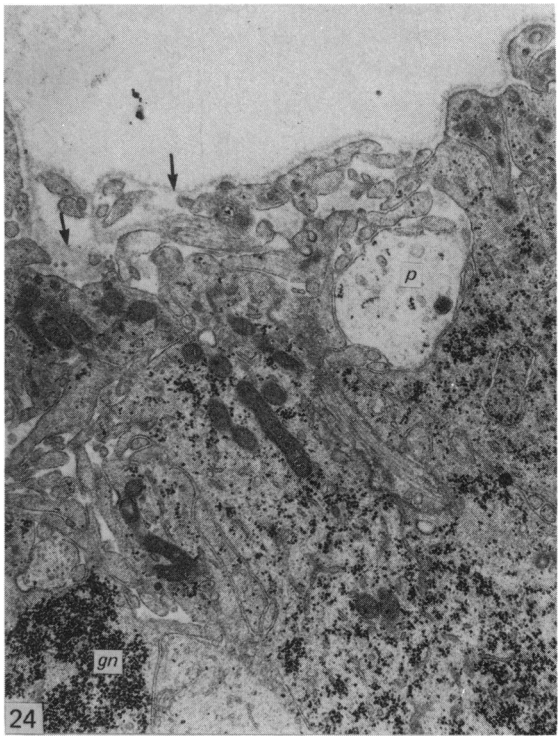
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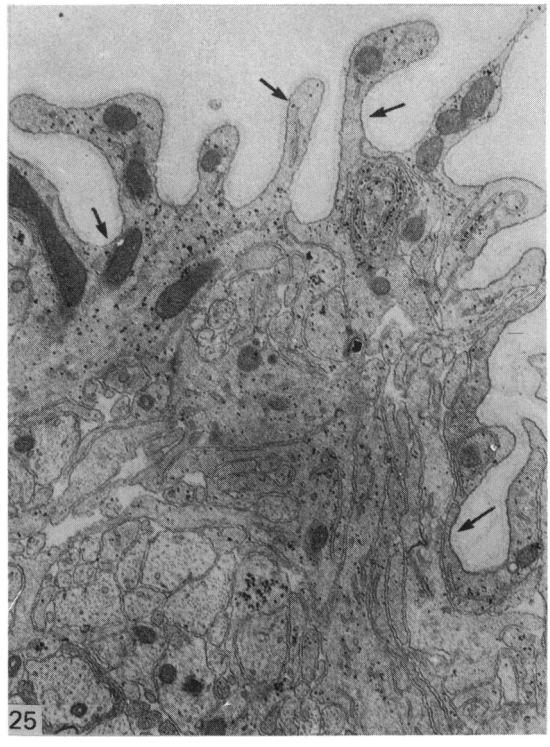
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canal ependymal cells develop characteristics which seem to be unique to the central canal.

SUMMARY

The central canal of the adult mouse spinal cord is lined for most of its extent by ependymal cells which are rich in microfilaments and whose apical surface is covered with matted, broad microvilli. The canal itself is filled with amorphous material containing glycogen granules. Two forms of this material are present, a dark form rich in glycogen, and a light form containing a few glycogen granules. Each type appears to be surrounded by a membrane. The upper cervical region, however, has a large empty lumen and the ependymal cells in this region have only scattered, narrow microvilli.

During development, the floor and roof plates are at first composed largely of ependymoglial cells, unlike the lateral walls, where undifferentiated neuroepithelial cells predominate. By E15 few undifferentiated neuroepithelial cells remain. At E17 the morphology of the ependymal cells changes. Their apical surface becomes covered with matted, club-shaped microvilli and the central canal is filled with glycogen-containing material. By P5 microfibrils are present in large bundles in the ependymal cells.

The piaglial surface opposite the roof and floor plates has finger-like projections unique to these regions and these persist at the surface of the dorsal median septum until myelination is well advanced after P5. The fibres forming the dorsal median septum are at first pale processes containing scattered glycogen granules and microtubules. By P5 microfibrils are present and at P150 the processes are packed with masses of microfibrils.

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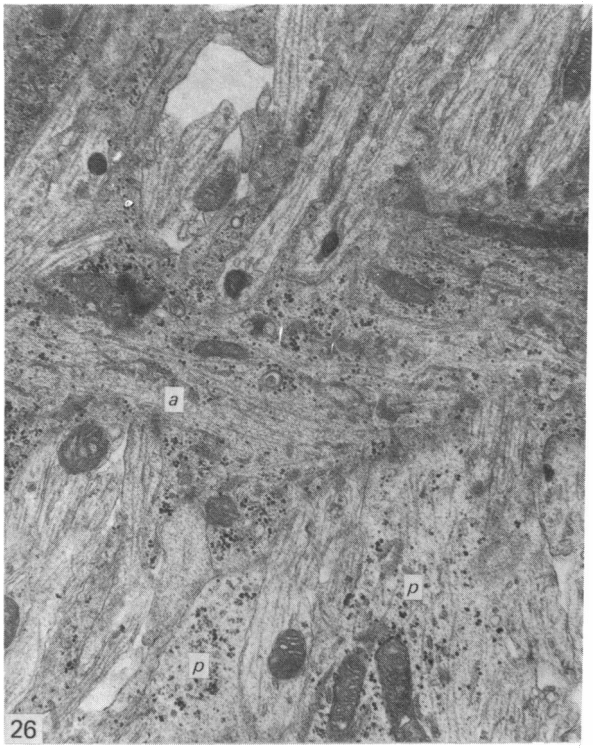
Fig. 22. Electron micrograph from upper cervical cord at P150. The ependymal cells contain microfibrils and sparse glycogen but where there is a large lumen (see Fig. 6) the canal surface of the ependymal cells has scattered narrow microvilli (*mv*) in contrast to other regions. At P150 lipid droplets (*L*) are found in the cytoplasm of ependymal cells throughout the cord. × 12600.

Fig. 23. Electron micrograph showing ventral surface of spinal cord at E16. The peripheral processes of the ependymoglial cells forming the floor plate are very rich in glycogen (*gn*) just below the surface. The surface is formed by frond-like processes covered with basement membrane (arrows). × 12600.

Fig. 24. Electron micrograph showing dorsal surface of spinal cord at E16. The appearance of the ependymoglial cells forming the dorsal median septum at this age is similar to that seen at the ventral surface (Fig. 23). Masses of glycogen (*gn*) are found and a basement membrane (arrows) can be seen. A very pale process (*p*) is present. × 12600.

Fig. 25. Electron micrograph showing dorsal surface of spinal cord at P5. At this age the surface of the dorsal median septum is characterised by finger-like projections each covered by basement membrane (arrows). × 12600.



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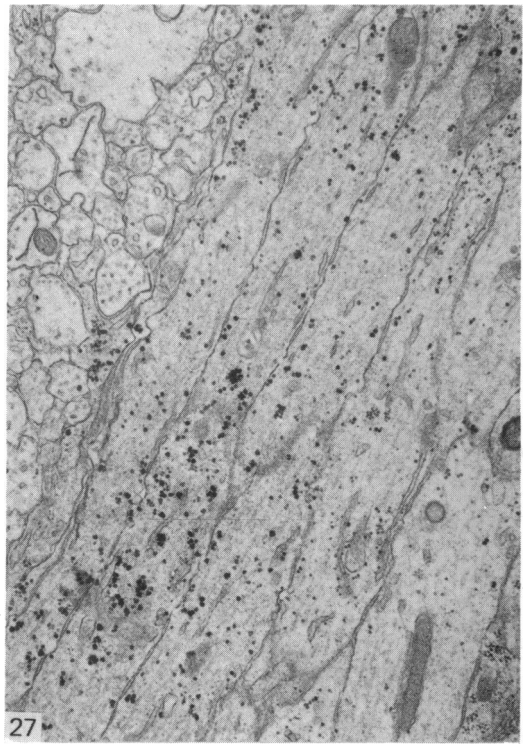
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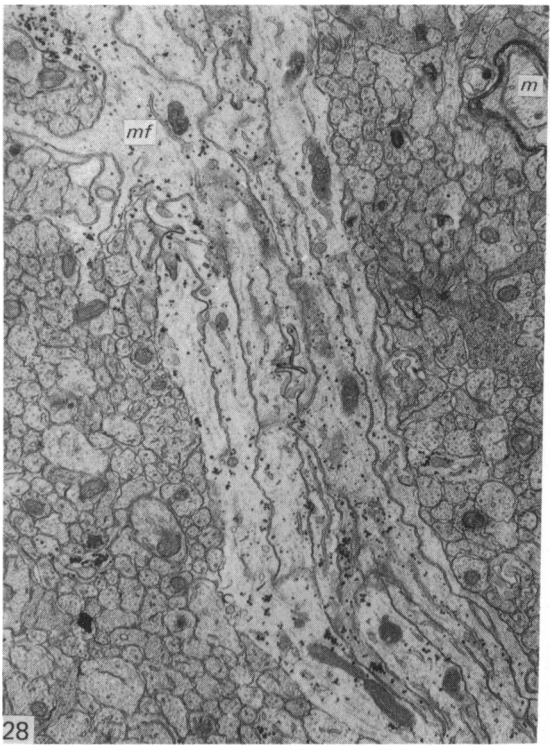

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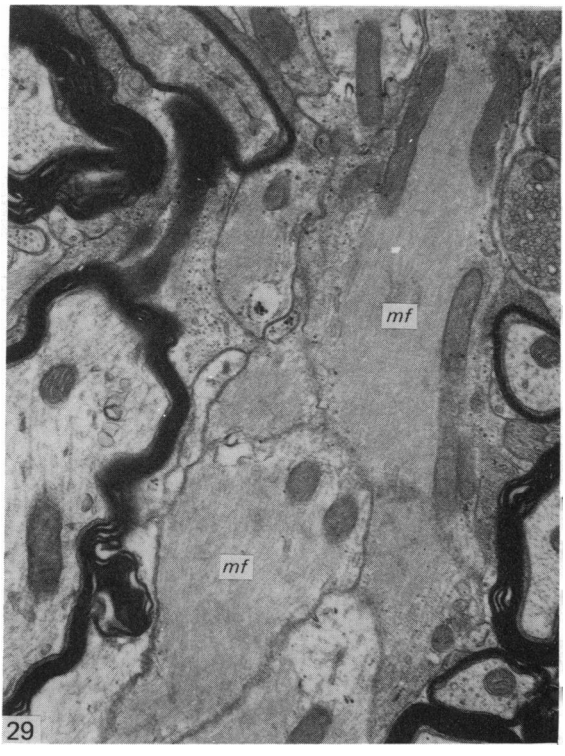

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Fig. 26. Electron micrograph of cord showing pale glycogen-containing peripheral processes (*p*) of floor plate ependymoglial cells becoming entwined with axons (*a*) of the developing anterior white commissure. At E12. ×18900.

Figs. 27–29. These electron micrographs show the development of the dorsal median septum in the region of the corticospinal tract. At E18 (Fig. 27; ×20000) the processes contain microtubules and scattered glycogen granules as well as a few mitochondria and strands of rough endoplasmic reticulum. At P5 (Fig. 28; ×12600) microfibrils (*mf*) are present in the pale-staining septal fibres. A few myelinated axons (*m*) are present in the corticospinal tract. By P150 the dorsal median septum is complete only in the region of the corticospinal tract. The fibres forming it are particularly rich in microfibrils (Fig. 29; ×20000).

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