Freeze-fracture replicas were prepared from sheep foetal choroid plexuses at 40, 45, 60 and 125 d of gestation using methods described previously[14]. Figure 1*a* shows a replica of a fracture through the apical region of the choroid plexus epithelium of an early sheep foetus (40 d gestation); it shows the cell membrane faces of adjoining cells. Below the level of the microvilli are seen the linear ridges and complementary linear grooves which constitute the tight junction. Figure 1*b* shows a replica at 125 d gestation; its appearance is not obviously different from that at 40 d. Similar results were obtained at 45 and 60 d gestation. To test whether permeability of non-electrolytes and establishment of ionic gradients might correlate with strand number and/or junctional depth, detailed studies of a large number of tight junctions at different foetal ages have been carried out (K.M. and N.R.S., unpublished). Measurements of the minimum number of strands and the mean junctional depth were made as described by Claude and Goodenough[6]. Freeze-fracture replicas of more than 400 areas of lateral–apical cell membrane have been examined. Even in the youngest foetuses the choroid plexus intercellular tight junctions contained several continuous strands in all cases. At 40 d gestation the minimum number of strands was 3.64 ± 1.12 and the mean junctional depth was 0.34 ± 0.17 μm (s.d., $n=121$). At 125 d gestation the number of fibrils (3.45 ± 1.13; s.d., $n=49$) was not significantly different, but the junctional depth (0.28 ± 0.14 μm; s.d., $n=49$) was significantly less. It has been shown that at these gestational ages, the thin section electron-microscopic appearance of the tight junctions was similar (C. A. N. Evans, D.H.M., K.M., J. M. Reynolds, M. L. Reynolds and N.R.S., unpublished).

Studies of the penetration of materials from blood into brain and cerebrospinal fluid (CSF) in sheep foetuses of the same gestational ages as those examined in the present study have shown considerable changes in the permeability characteristics of the system during foetal development. For example, lipid-insoluble molecules of small molecular weight (erythritol, sucrose and inulin) as well as albumin penetrate from blood into CSF at a rate and to an extent which may suggest unrestricted passive diffusion at 60 d; in contrast, there is considerable restriction by 125 d, at which stage the mean pore radius was estimated to be about 1 nm (unpublished). Foetal development of CSF–plasma electrolyte gradients has been discussed previously[11] and the gradients for individual ions were shown to appear at very different stages in the order magnesium, chloride, potassium, calcium. The concentration of protein in early foetal CSF is very high, for example 500 mg per 100 ml in 55-d sheep foetuses. At least part of this high level seems to be caused by a mechanism which transports specific proteins (for example, α-foetoprotein and transferrin) from blood into CSF early in gestation but not later when the CSF concentration of protein has fallen to near adult levels (50 mg per 100 ml)[13].

Since tight junction strand number as revealed by freeze fracture does not change significantly during a major part of gestation, this cannot account for the observed alterations in the permeability properties. From the wide variety of epithelia studied with the freeze-fracture technique it is clear that there are important exceptions to the proposed correlation of permeability with fibril (strand) number (ref. 10 and refs therein). Additional evidence against this correlation comes from recent experiments[16] which showed that in the toad bladder treated with hypertonic lysine there was a marked decrease in transepithelial resistance, but no significant change in either fibril number or mean depth of the tight junctions.

It may be that some other feature of tight junctions not revealed by electron microscopy could account for some of these changes in permeability. Both the molecular structure of the junctional membrane and the biochemical composition of the material enclosed by the junction fibrils might be important for passive ion transport. Nevertheless, one type of extracellular pathway through tight junctions could not account for all our permeability data for the blood–CSF barrier. For example, as mentioned above, there is penetration of albumin from blood into CSF in the 60-d foetus. Such a large pathway, were it present within choroid plexus tight junctions, would be well within the resolution of our electron-microscopical observations. It seems more likely that protein and non-electrolyte penetration and perhaps some type of ion transport involves a quite different route, that is, a transcellular one, consisting of a tubulovesicular system. An intracellular system of tubules and cisternae of endoplasmic reticulum has recently been implicated in transcellular transport of protein[14] and non-electrolytes (C. A. N. Evans, D. H. M., K.M., J. M. Reynolds, M. L. Reynolds and N.R.S., unpublished) in foetal choroid plexus as well as in active sodium transport in frog skin[15].

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K. MØLLGÅRD

Institute of Medical Anatomy A, University of Copenhagen, Universitetsparken 1, 2100 Copenhagen, Denmark

D. H. MALINOWSKA
N. R. SAUNDERS

Department of Physiology, University College London, Gower Street, London WC1E 6BT, UK

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Errata

The order of the authors of the article "Release and metabolism of substance P in rat hypothalamus" (*Nature*, **264**, 81; 1976) should have been T. Jessell, L. L. Iversen and I. Kanazawa and not as printed.

In the article "Stratospheric aerosols and climatic change" (*Nature*, **263**, 551; 1976) the order of the authors' names was changed and should have read J. B. Pollack, O. B. Toon, A. Summers, B. Baldwin, C. Sagan and W. Van Camp.

In the article "Polymers for the sustained release of proteins and other macromolecules" by R. Langer and J. Folkman (*Nature*, **263**, 797; 1976) the units on the abscissa in Fig. 1 should be hours.