Scanning Electron Microscopy of *Plexus Choroideus* in Adult Sheep

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Abstract

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The outer morphology of the surface of the ependymal lining of the *plexus choroideus* was studied within the whole ventricular system of the brain in adult sheep using a scanning electron microscope. The more abundant occurrence of spherical secretory protrusions was found on the *plexus choroideus* (PCH) in the lateral cerebral ventricle. A frequent finding of the macrophage-like epiplexal cells was recorded on the PCH in the region of the lateral as well as the fourth ventricle. No differences were found in the structure of the PCH surface, which could be related to the sexual differentiation of the sheep. Our results are compared, above all, with those found in the adult goat and in the foetuses of both species of small ruminants.

Central nervous system, plexus choroideus, cerebral ventricles, adult sheep

Less attention has been paid to the study of the *plexus choroideus* as one of the structures of the ventricular system of the brain in different species of mammals using the scanning electron microscope than to the structure of the surface of the ependymal lining. In the prenatal but also in postnatal period, the surface of the PCH in laboratory rodents was observed by Yamadori (1972), Hosoya and Fujita (1973), Chamberlain (1974), Peters (1974), Mestres and Breipohl (1976), Peters and Swan (1979), in the dog (Allen 1975; Persky 1980), in the cat (Clementi and Marini 1972), in the rhesus macaque (Ling 1983), and in humans (Scott et al. 1972, 1974; Otani and Tanaka 1988, O'Rahilly and Müller 1990). In farm mammals the PCH was studied in the goat and sheep foetuses but also in the adult goat (Rajtová 1997, 2000). There is no record of the structure of the surface of epithelial cells that cover the PCH in adult sheep.

The aim of the work was to describe the epithelial surface of the PCH in the lateral, third and fourth cerebral ventricles in adult sheep with the aid of a scanning electron microscope.

Materials and Methods

Ten healthy adult sheep (5 ewes and 5 rams) were used. The animals were killed in March and October by exsanguination following thiopental anaesthesia. Immediately after exsanguination, the heads of the animals were perfused through the *a. carotis communis* with 0.2 mol/l phosphate buffer solution and prefixed with Karnovsky solution. The specimens of the individual parts of the *plexus choroideus* were postfixed in 3% glutaraldehyde in 0.2 mol/l cacodylate buffer. The temperature of the solutions was 4 °C, pH 7.4. The specimens were processed by the method of Murakami et al. (1977), dehydrated, dried by the critical point method and coated with gold in vacuum. Then they were studied in the Tesla BS 340 scanning electron microscope with image analysis.

Results

The *plexus choroideus ventriculi lateralis* forms low folds. The periphery of the ependymal cells is irregularly round, almost elongated. The apical membranes of most lining ependymal cells are convex, covered with thick, short, densely arranged microvillus-like structures. Among them on numerous sites are abundant, predominantly small and spherical,

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but also some elongated protrusions (Plate I, Fig. 1). On the surface of the ependymal cells or between the PCH folds, epiplexal cells frequently occurred. The bodies of some of them are small, irregularly round with uneven surfaces. They have 2 or 3 short processes, which are almost all formed by "ruffled membranes" (Plate I, Figs 2, 4). The peripheral margins of these membranes could pass into the filiform elongations. Other epiplexal cells have their bodies oval with a smooth surface. These form 3-4 processes, which are changed to "ruffled processes" either totally or partly (Figs 1, 3).

The surface of the *plexus choroideus ventriculi tertii* is folded almost as much as in the lateral ventricle. The apical membranes of the lining ependymal cells are markedly convex (Plate II, Figs 5, 8). They are covered with dense fibrous microvilli that are not regular in height. The presence of small spherical protrusions as well as epiplexal cells is not frequent in this region; these structures occur, as a rule, in the deeper sites between folds (Plate xy, Fig. 6). The epiplexal cells have small, irregularly round, or elongated bodies from which more protrusions originate. These form "ruffled membranes" either on the periphery or all over (Fig. 5). Beside them, longer, thin, filiform processes can also originate from the body (Plate II, Fig. 7).

The surface of the *plexus choroideus ventriculi quarti* in adult sheep is especially folded, and forms numerous small glomoid structures (Plate III, Fig. 9). The apical membranes of the ependymal cells are convex, and covered with low, thin, densely arranged microvilli that are at some sites replaced with thick, dense, microvillus-like structures. Some small round almost elongated protrusions occur sporadically between microvilli (Plate III, Figs 9, 10, 12). The epiplexal cells are dispersed over many sites, above all, in the gaps between folds (Figs 9, 11, 12). Their bodies are predominantly oval almost tapering with 2-3 processes which either whole or partly form "ruffled membranes" on the periphery.

In all the sheep examined, no typical cilia were found on the surface of the entire PCH; nor were differences between ewes and rams recorded.

Discussion

If the results in the adult sheep presently studied are compared with those in adult goats (Rajtová 2000) some common, but also different characteristics may be stated. The apical membranes of the ependymal cells of the PCH in both species of small ruminants are markedly convex and the PCH *ventriculi quarti* surface is the most folded, and therefore it is very fragile. In adult sheep, as in goats, almost the entire PCH surface is covered with thin microvilli or microvillus-like thicker structures. The typical cilia are absent. The small spherical or elongated protrusions of secretion in the adult sheep studied occurred abundantly on the PCH surface in the lateral and fourth cerebral ventricle, while in the region of the third ventricle their presence was rarer. Unlike in sheep, in adult goats the secretory protrusions were recorded only in the *ventriculus lateralis* (Rajtová 2000).

There was an abundant occurrence of epiplexal cells in goats in the third cerebral ventricle, above all, in the region of *foramen interventriculare* (Rajtová 2000); in adult sheep their presence was substantially lower. Their occurrence was recorded, especially in the fourth but also in the lateral cerebral ventricle.

Some authors, engaging in the description of supraependymal or epiplexal cells, have considered them as macrophages capable of movement and phagocytosis (Kitamura 1973; Hosoya and Fujita 1973; Chamberlain 1974; Bleier et al. 1975; Sturrock,1983, 1988), or as the structures derived from the macrophages circulating in the blood (Carpenter et al. 1970; Ling 1983; Boya et al. 1986; Maxwell and McGadey 1988; Lu et al. 1993; Ling and Wong 1993; Ling et al. 1998).

The outer morphology of the PCH surface in adult sheep differs more substantially from that in the sheep foetuses (Rajtová 1997) than from that in the adult goat. In the sheep and

goat foetuses, on the PCH besides microvilli, there are also typical cilia scattered or arranged in tufts. Unlike in the adult sheep (the same pertains to adult goats, Rajtová 2000) the occurrence of spherical secretory protrusions in the period of the prenatal life of sheep is very abundant, which would indicate a much more active prenatal than postnatal secretory activity of PCH. The presence of epiplexal cells on the foetal PCH of both species of small ruminants was limited only to the third and partially the lateral ventricle (Rajtová 1997), in adult sheep they were found in a greater number in the *ventriculus quartus*.

Even if we have recorded the presence of EPC on the PCH surface, and although their outer morphology is the same as that of some cells considered by other authors as macrophages on the PCH surface (Carpenter et al. 1970, Maxwell and McGadey 1988; Ling and Wong 1993; Ling et al. 1998) we have not made a statement about their origin, because our study in the scanning electron microscope was not supplemented by a confirmation in the transmission electron microscope or histochemically. The outer morphology of epiplexal cells in adult sheep and goats (Rajtová 2000), as well as in the foetal material from both species of small ruminants (Rajtová 1997) is the same as their supraependymal cells under the physiological conditions (Rajtová 1988, 1990; Rajtová and Kačmarik 1998). Similarly according to Tseng et al. (1983) the epiplexal cells resemble the supraependymal and ameboid cells in the *cavum septum pellucidum*.

In adult sheep, as has been found in adult goats, it is also valid for their prenatal period (Rajtová 1997, 2000). No differences were recorded which would be connected with the sexual differentiation as was found in case of the ependymal lining of the cerebral ventricles of the brain.

Plexus choroideus dospelej ovce: štúdium v riadkovacom elektrónovom mikroskope

Práca sa zaoberá štúdiom povrchu epiteliálnej výstielky plexus choroideus (PCH) v celom ventrikulárnom systéme mozgu u dospelej ovce pomocou riadkovacieho elektrónového mikroskopu. Na plexus choroideus (PCH) *laterálnej komory* boli nájdené hojné sférické protrúzie. Častý výskyt makrofágom podobných epiplexových buniek bol zaznamenaný na PCH v oblasti laterálnej, ale aj štvrtej komory. Neboli zistené rozdiely v stavbe povrchu PCH, ktoré by súviseli so sexuálnou diferenciáciou oviec. V práci sa porovnávajú výsledky získané z dospelých oviec predovšetkým s údajmi u dospelých kôz ale aj plodov u obidvoch druhov malých prežúvavcov.

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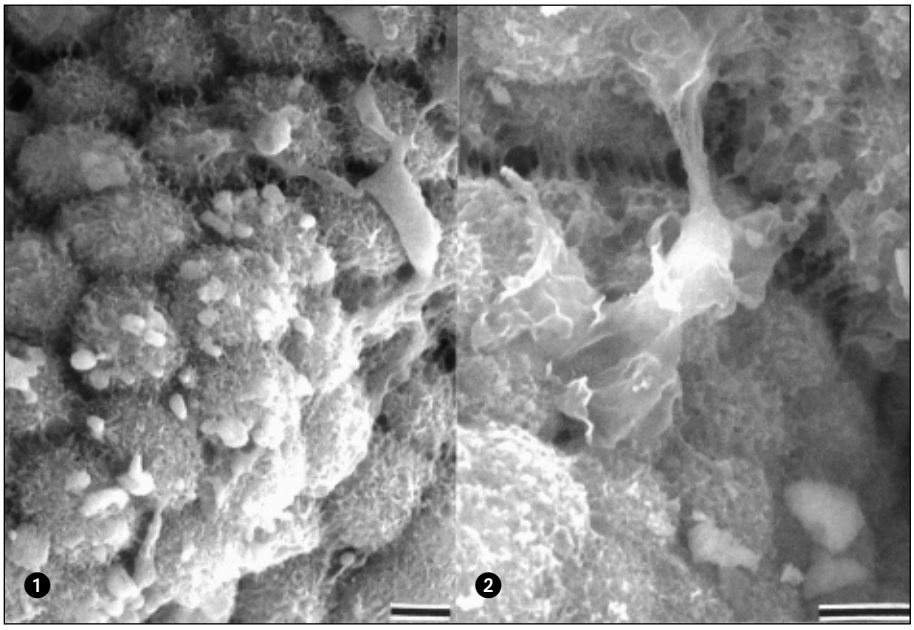
Plate I
Rajtová V.: Scanning ... pp. 279-282



Figs 1-4. Details of the *plexus choroideus ventriculi lateralis*. The surface of apical membranes is more or less covered by thicker microvillus-like structures. Small spherical to prolongated protrusions are abundant. The occurrence of the epiplexal cells is frequent. Bars: Fig. 1 = 5 μm, Figs 2 - 4=5 μm.

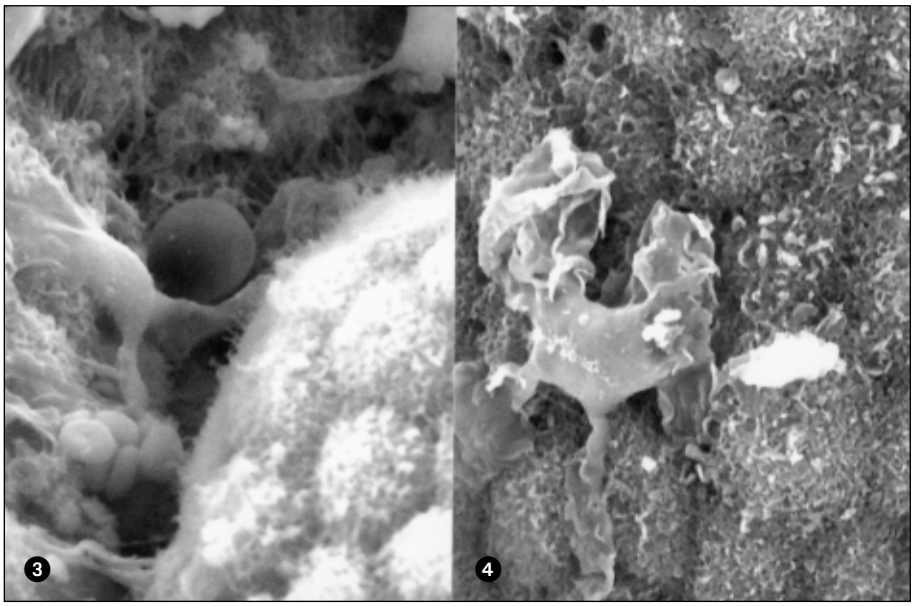
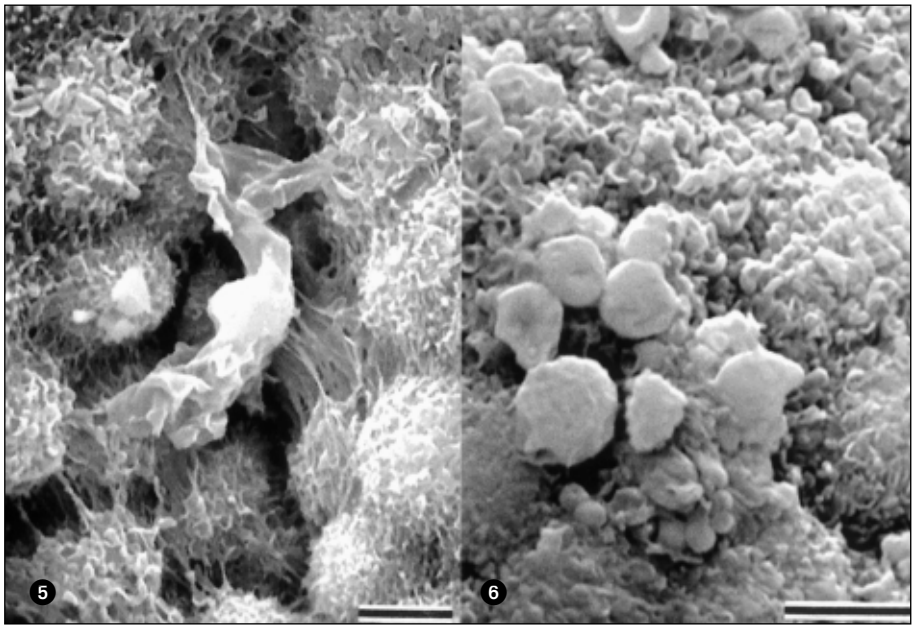


Plate II



Figs 5 – 8. The surface of the convex apical membranes of the *plexus choroideus ventriculi tertii* cover filiform and thick, microvillus-like structures. The occurrence of small spherical protrusions is rare. Isolated epiplexal cells have rounded or elongated bodies with filiform processes (arrows), and the processes which form “ruffled membranes”. Bars: Figs 5, 8 = 5 μm, Figs 6, 7 = 5 μm.

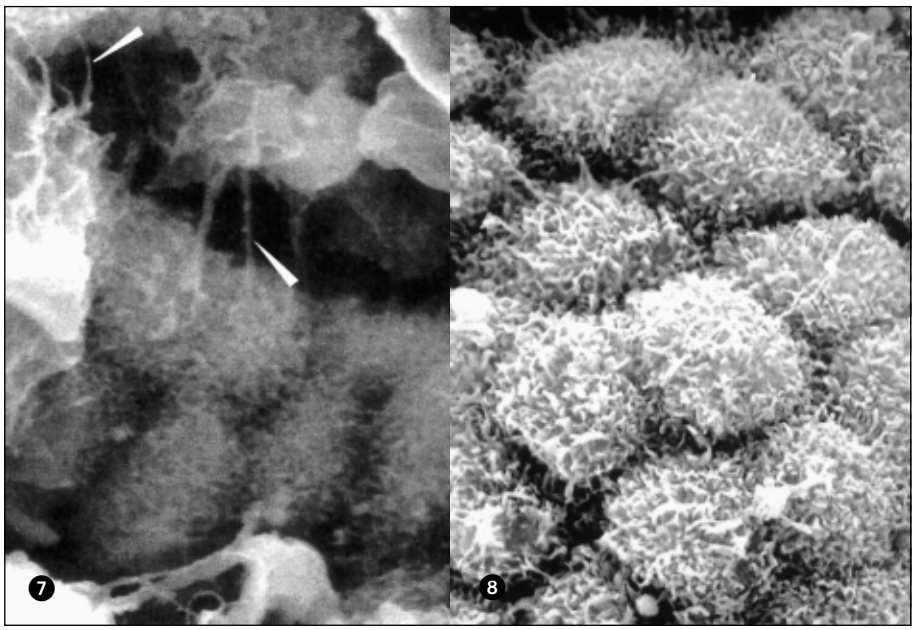
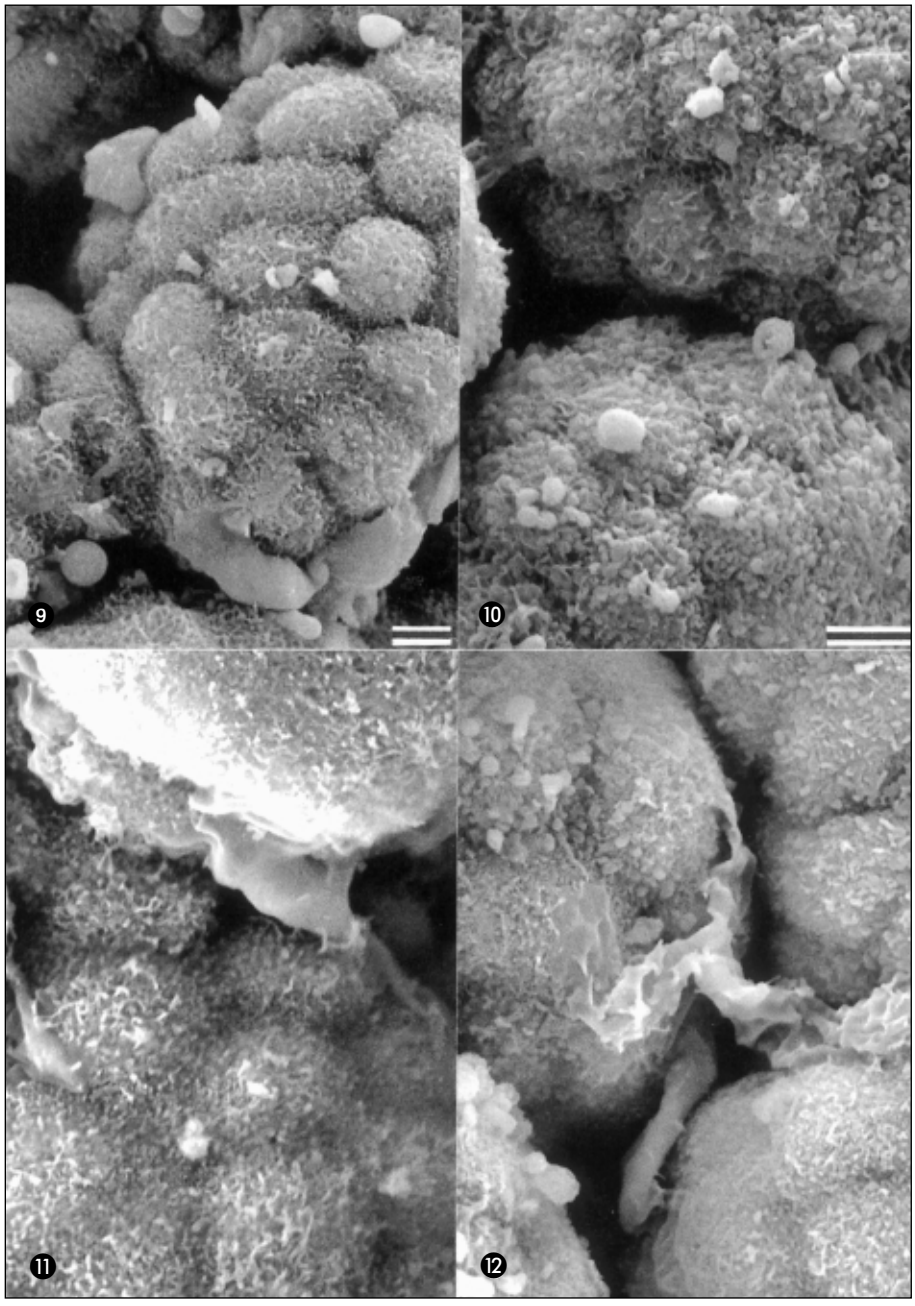


Plate III



Figs 9 – 12. The surface of the plexus choroideus ventriculi quarti consists of the glomoid formations. The convex apical membranes, apart from except the microvilli and microvillus-like structures, cover small abundant spherical protrusions. The epiplexal cells have two or more processes forming the “ruffled membranes”. Bars: Fig. 9 = 5 μm, Figs 10 – 12 = 5 μm