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Morphometric Parameters of the Rat Choroid Plexus Blood Vessels

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Background: The choroid plexus is an epithelial-endothelial vascular structure within the ventricular system of the vertebrate brain. It consists of epithelial cells, fenestrated blood vessels, and the stroma, dependent on various physiological or pathological conditions.

Methods: The blood vessels divided in four subgroups of the choroid plexus of young (1 month) and adult (10, 13 and 22 months) rats were morphometrically studied. The investigations were performed on semithin sections examined with the light microscope using a square grid system calibrated for semiautomatic image analysis.

Results: No significant changes were observed in the luminal diameter and cross-sectional area of capillaries (vessels <15.0 μm in diameter) and large vessels (vessels of 15.5 – 30.0 μm and >30.0μm in diameter) in the young (mean luminal diameter of capillaries – 8.95±0.29 μm) and adult (mean luminal diameter of capillaries – 9.15±0.58 μm of 10 months, 9.08±0.26 μm of 13 months, and 9.25±0.29 μm of 22 months) rats.

Conclusion: These results indicate that the luminal diameter and cross-sectional area of the capillaries and large vessels of the plexus choroideus in the young and adult rat do not change. In young rats the blood vessels with luminal diameter >30 μm have not been found and this may be related with a larger number of the blood vessels with small luminal diameter, i.e. capillaries.

Key words: rat choroid plexus, blood vessels, morphometrical and ultrastructural study.

Introduction

The choroid plexuses are specialized highly vascular anatomycal structure which protrude into the lateral ventricle, as well as in the third ventricle and fourth ventricle. The surface of the choroid plexus consists of numerous villi each covered with single layer of epithelial cells surrounded by vascular connective tissue cells [6, 2]. These cells are generally considered to be modified ependymal cells with epithelial cell characteristics and referred to as choroidal epithelial cells.

The choroid plexus is mainly involved in the production of cerebrospinal fluid (CSF) by using the free access to the blood compartment of the leaky vessels. In order to separate blood and CSF compartments, choroid plexus epithelial cells and tanycytes of circumventricular organs constitute the blood–CSF–brain barrier [12]. As a secretory

source of vitamins, peptides and hormones for neurons, the choroid plexus provides substances for brain homeostasis [3]. Most blood vessels in the plexus choroideus are wide-calibre (approximately 15 μm) capillaries with thin fenestrated endothelial walls and bridging diaphragms overlying the fenestrations [5].

Purpose of the present study is an investigation of the morphometrical characteristics of the rat choroid plexus blood vessels.

Materials and Methods

Wistar rats aged 1 (n=5), 10 (n=5), 13 (n=5) and 22 (n=5) months were used in the present study. The animals were anesthetized with sodium pentobarbital (40 ml/kg, i.p.) and perfused intracardially with 2.5% glutaraldehyde and 2% paraformaldehyde in 0.1 M cacodilate buffer [4]. Extracted choroid plexuses were postfixes in 1% OsO_4, dehydrated through graded ethanol and embedded in Durcupan and examined with JEOL JEM 1200EX transmission electron microscope. The semithin sections (1μm) were stained with 1% toluidine blue for morphometric measurements and examined under Light microscope Carl Zeiss Jena.

Morphometric Analysis

Investigations were performed on semithin sections examined with the light microscope using a square grid system [11] calibrated for semiautomatic image analysis, described in our previous studies [6, 7].

In the present study the relative number of blood vessels and luminal diameter and cross-sectional area of the blood vessels divided in four subgroups were measured. The luminal diameter was measured as perpendicular distance across the maximum chord axis of each vessel.

Statistical Analysis

Results are reported as mean values ± SEM and as relative part in percentage, and statistically analyzed by Student's t-test using statistical package (STATISTICA, ver.6, Stat-Soft Inc., 2001), and differences were regarded as significant at $p<0.05$.

Results

Changes in luminal diameter, area of the luminal profile and relative part of the all blood vessels and vessels divided in four subgroups in young (1 month) and adult rats (10, 13 and 22 months) were determined. These findings are shown on Table 1 and in Figure 1. No significant changes were observed in the luminal diameter of capillaries (vessels <15.0 μm in diameter) and large vessels (vessels of 15.5 – 30.0 μm and >30.0 μm in diameter) in the young (mean luminal diameter of capillaries – 8.95±0.29 μm) and adult (mean luminal diameter of capillaries – 9.15±0.58 μm of 10 months, 9.08±0.26 μm of 13 months, and 9.25±0.29 μm of 22 months) rats. Blood vessels with luminal diameter >30 μm have not been found in young rats.

The surface of the choroid plexus consists of small villi each covered with a single layer of large cuboidal epithelial cells, electron-dense epithelial cytoplasm, well diffe-

Table 1. Morphometric data of choroid plexus blood vessels of young and adult rats (luminal diameter in μm and cross-sectional area in μm^2)

Blood vessels Diameter	1 month		10 months		13 months		22 months	
	Luminal diameter ± SEM	Luminal area ± SEM	Luminal diameter ± SEM	Luminal area ± SEM	Luminal diameter ± SEM	Luminal area ± SEM	Luminal diameter ± SEM	Luminal area ± SEM
5.0 – 7.5 μm	6.54±0.29	72.82±3.74	6.77±0.80	76.48±4.08	7.06±0.22	74.95±4.37	7.18±0.25	79.85±3.88
8.0 – 15.0 μm	11.35±0.28	177.27±6.95	11.53±0.37	192.14±8.24	11.10±0.30	188.77±9.69	11.32±0.33	190.39±8.25
15.5 – 30.0 μm	19.03±1.07	412.15±53.10	35.52±3.34	437.45±34.78	18.79±0.69	364.52±35.99	19.73±0.74	403.36±32.66
>30 μm	-	-	35.52±3.34	741.11±40.68	32.18±1.98	774.08±27.99	37.54±2.45	774.56±18.35
Number of measurements	324	324	280	280	262	262	350	350

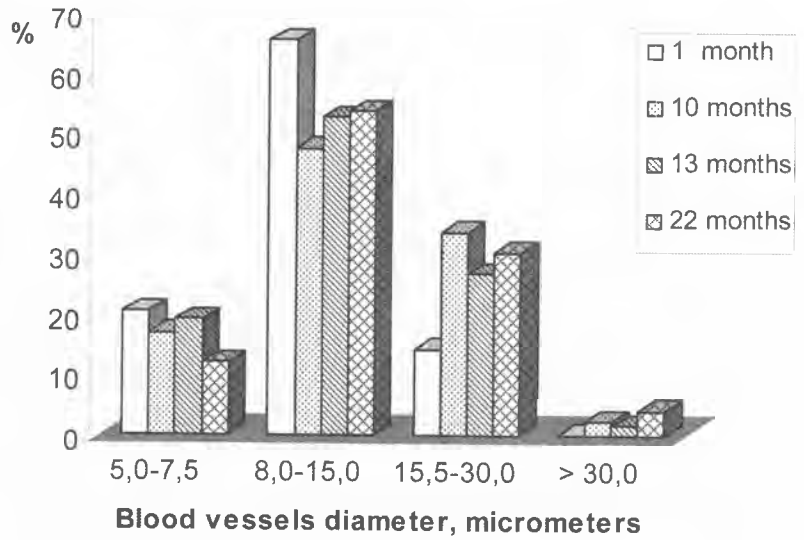

Fig. 1. Comparison of morphometric data of choroid plexus blood vessels in young and adult rats (relative part in %)

rentiated connective tissue elements and capillary with many fenestrations (1-month rat). The microvilli of the epithelial cells of the rat choroid plexus aged 10 and 13 months are fine, slender and dense, the nuclei of the epithelial cells are rounded, basally located and have relatively homogenous chromatin (Fig. 2). The cytoplasm of the dark epithelial cells contains more polyribosomes and endoplasmic reticulum. In the epithelial cytoplasm are seen many lipid droplets, imbibing mitochondria, dense bodies and electron-dense lysosome-like bodies with lamellar structures. These changes (22 months rats) might indicate a gradual change in function or at least a decrease in efficiency of the choroid plexus and may be evidence of slow degeneration of the rat choroid plexus.

Discussion

Up to present time little quantitative information has been available regarding the vessels of the brain, in particular, regarding the vessels of the rat choroid plexus. The capillaries of the plexuses had a large diameter and sinusoidal dilations, and showed the presence of occasional short, blind sprouts indicative of angiogenesis. Short anastomoses between arterioles supplying the plexuses and venules draining them were only rarely observed [13].

Statistical evaluation of data showed that the diameter and cross-sectional area of capillaries (vessels <15.0 μm in diameter) and that of large vessels (vessels of 15.5 – 30.0 μm and >30.0μm in diameter) were statistically unchanged in the young and adult rats. The mean luminal diameter of blood vessels of 15.5-30.0 μm was 19.75±0.78 μm in the 10, 13 and 22 months rats. The mean cross-sectional area of capillaries (vessels <15.0 μm in diameter) in the young rats was 125.04±4.99 μm^2 and the mean cross-sectional area of capillaries in adult rats was 133.76±6.41 μm^2. Blood vessels with luminal diameter >30 μm have not been found in young rats and this may be related

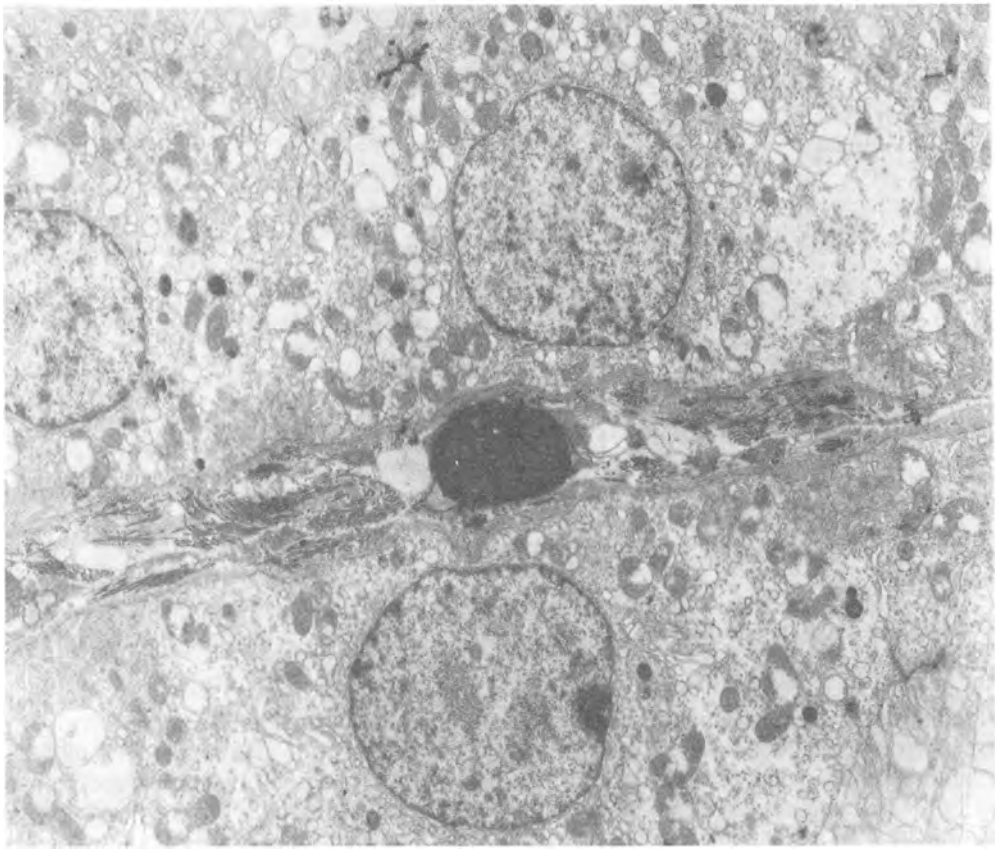

Fig. 2. Rat choroid plexus aged 13 months. The epithelial cells with a round nucleus, numerous microvilli, scanty connective tissue and blood vessel at the basal part of the epithelial cells. × 3 000

with a larger number of the blood vessels with small luminal diameter, i.e. capillaries (85.80%) and blood vessels of 15.5 – 30.0 μm in diameter (14.20%).

The choroid plexus epithelium constitutes a physical barrier between blood and cerebrospinal fluid (the blood–CSF barrier – BCSFB) by virtue of the complexity of the tight junctions between adjacent epithelial cells. The age-related changes (22-month rats) might indicate a gradual change in function or at least a decrease in efficiency of the choroid plexus and may be evidence of slow degeneration of the rat choroid plexus [7, 9, 10].

The investigation of the age-related changes in choroid plexus indicate that normal ageing processes alter protein content in the CSF, CSF secretion and integrity of the BCSFB, which could impact on CSF homeostasis and turnover [1, 8].

Conclusion

These results indicate that the luminal diameter and cross-sectional area of the capillaries and large vessels of the plexus choroideus in the young and adult rat do not change. In young rats the blood vessels with luminal diameter >30 μm have not been found and

may be related with a larger number of the blood vessels with small luminal diameter, i.e. capillaries (85.80%). In adult rats the relative part of the capillaries is mean 67.25%. The ageing of choroid plexus in rats is morphologically characterized by atrophy of the epithelial cells and these functional changes could result in decreased choroid plexus functions of secretion, absorption and transport of substances and could play a part in normal homeostatic functions in CNS.

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