

Neurodegenerative Changes in Aging Rats

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The choroid plexus in the brain ventricles consists of epithelial cells, fenestrated blood vessels, and the stroma, dependent on various physiological conditions. Changes of the rat choroid plexus, characterized by reduction of the capillaries (20%) and atrophy of epithelial cells, are evidence of aging degeneration processes. GD1a gangliosides could be used as biomarkers of the neurodegeneration. Serum IgG anti-GD1a titer was determined in young and aging rats to establish the existence of neurodegeneration process. Serum IgG anti-GM1 titer correlated with demyelination and serum IgG anti-GM3 titer correlated with loss of integrity of the blood brain barrier (BBB). Our morphological and immunological studies demonstrated that changes in the structure of the plexus choroideus, neurodegeneration, demyelination and damage of the integrity of the BBB are available in adult rats.

Key words: aging rats, choroid plexus blood vessels, morphometric analysis, serum IgG anti-ganglioside antibodies, neurodegeneration.

Introduction

The choroid plexuses are specialized highly vascular anatomical structure which protrude into the lateral ventricle, as well as in the third ventricle and fourth ventricle. As a secretory source of vitamins, peptides and hormones for neurons, the choroid plexus provides substances for brain homeostasis [2].

The auto-antibodies against GD1a gangliosides are associated with acute motor axonal neuropathy and acute motor-sensory axonal neuropathy [8]. Anti-gangliosides complex antibodies may be useful diagnostic and prognostic markers of the demyelination, neurodegeneration and correlated with the loss of integrity of the blood brain barrier (BBB) [3, 4, 9]. Patients with a positive titer of anti-ganglioside antibodies have significant alterations of motor conduction parameters and contribute to the neuropathy [10]. GM1 are the main myelin gangliosides and the high titer of IgG anti-GM1 antibodies is correlated with the demyelination [11]. Serum gangliosides GM3 could be used as biomarkers of BBB destruction [12].

The aim of the present investigation is to establish the changes of the microvasculature and to value the titers of the IgG anti-GD1a, anti-GM1 and anti-GM3 antibodies in aging rats.

Materials and Methods

Wistar rats aged 1, 13, 18, 22 and 32 months (five animals per group) were used in the present study. The animals were fixed by intracardial perfusion, embedded in Durcupan and examined with JEOL JEM 1200EX transmission electron microscope and Light microscope Carl Zeiss Jena (morphometric analysis) and Leica DM5000B. Results are reported as mean values \pm SEM and as relative part in %, and statistically analyzed by Student's t-test using statistical package. Differences were regarded as significant at $p < 0.05$.

We made slight modifications of the method of Mizutamari et al. [5]. The serum titers of IgG anti-GD1a, anti-GM1 and anti-GM3 were estimated by the enzyme-linked immunosorbent assay (ELISA) [8]. The results were accounted on the ELISA reader TECAN TM, Sunrise, Austria.

Results

Changes in relative part of the blood vessels divided in four subgroups in young (1 month) and adult rats (13 and 22 months) were analyzed and comprised (**Fig. 1**). Blood vessels with luminal diameter $>30 \mu\text{m}$ have not been found in young rats. This may be related to a large number of the blood vessels with small luminal diameter, i.e. capillaries (85.80%) and blood vessels of $15.5\text{--}30.0 \mu\text{m}$ in diameter (14.20%). Relative part of the capillaries in adult rats (13 and 22 months) was 71.75% and 65.70% respectively.

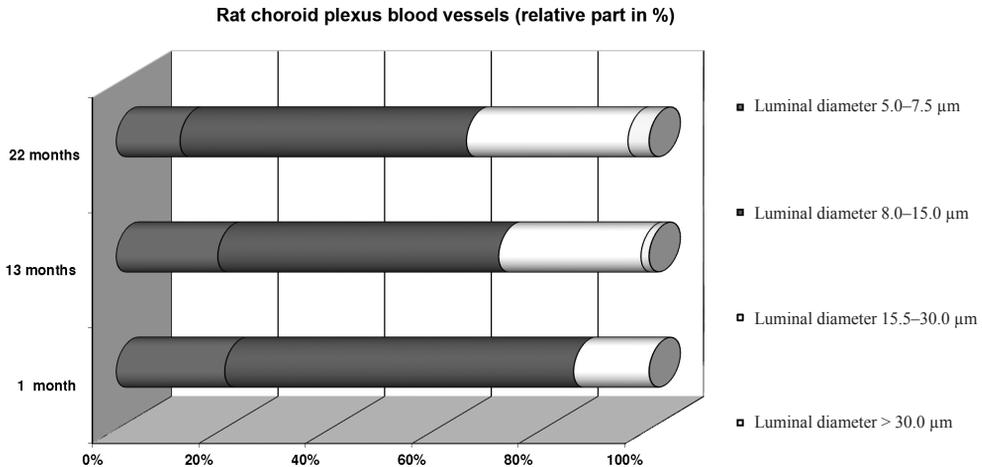


Fig. 1. Comparative analyses of the number (%) of choroid plexus blood vessels in young and adult rats

The surface of the young rat choroid plexus consists of small villi each covered with a single layer of large cuboidal epithelial cells, electron-dense epithelial cytoplasm, well differentiated connective tissue elements and capillary with many fenestrations (**Fig. 2**). In the epithelial cytoplasm of the aging rat choroid plexus were observed many lipid droplets, imbibing mitochondria and dense bodies (13, 22 months).

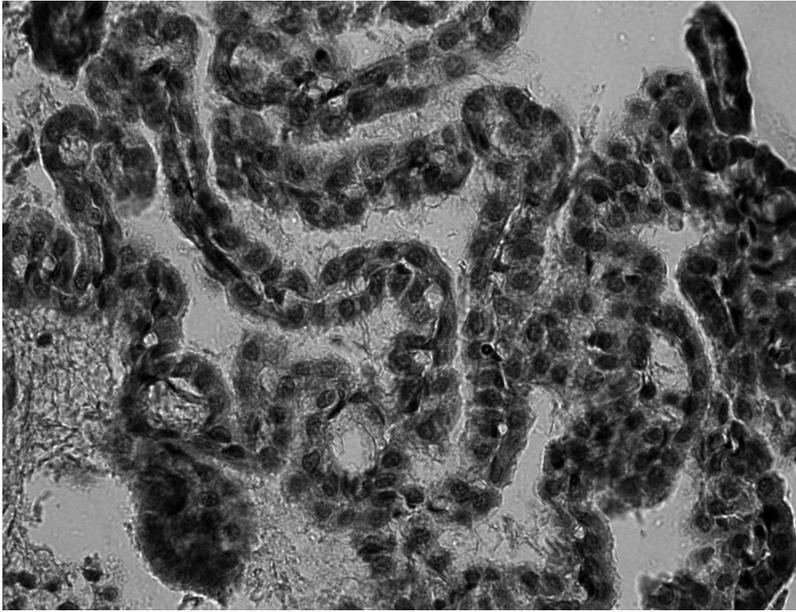


Fig. 2. Choroid plexus in the IV ventricle of the young rat brain. H&E stain, $\times 40$

There was a significant increase in the titers value of the IgG anti-GD1a, anti-GM1 and anti-GM3 only in adults – 32 month rats. These changes began in rats after the 22nd month and are probably related to the beginning of neurodegenerative changes in brain at this age.

Discussion

Normal aging is associated with various metabolic and vascular changes, which are systemic and organ-specific [1]. In the brain, both structure and function are altered with age. In the present study quantitative information has been available regarding the vessels of the brain, in particular, regarding the vessels of the rat choroid plexus. Blood vessels with luminal diameter $>30 \mu\text{m}$ have not been found in young rats. In our previous study the blood vessels with luminal diameter $>30 \mu\text{m}$ were found for the first time in rats aged 2 months [7]. This may be related 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 capillaries number was reduced by 20% in old rats and this compensatory reaction may be related with structural changes with age. 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-old 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 [6,13]. The value of IgG anti-GD1a, anti-GM1 and anti-GM3 titers determined by ELISA technique revealed the presence of the immune-mediated neurodegeneration, concomitant demyelination, growing with age, and impaired integrity of the BBB.

Conclusion

Our morphological and immunological studies demonstrated that in adult rats changes in the structure of the plexus choroideus are present also demyelination, neurodegeneration and damage of the integrity of the BBB are present. The observed changes are evidence of age-related neurodegenerative changes, which are important for the homeostatic regulation of CNS.

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