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Reduction of microvascular length in dentate gyrus of the hippocampus of diseased Alzheimer's rats

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Abstract

Alzheimer's disease (AD) is accompanied by changes in vascularization and exhibits many emotional abnormalities. In response to the growing interest in the use of a novel transgenic AD model, we aimed of this study to descript features of microcirculation using modern approaches of unbiased stereology to quantify various microcirculation parameters in the pyramidal cell layers of different hippocampal zones of TgF344-AD rats.

Motivation

Neurodegeneration during the Alzheimer disease (AD) are accompanied by changes of vascularization and exhibit many emotional abnormalities. Mice models of AD are commonly used in investigations. Nevertheless, rats are closer to human evolutionally. Therefore, the aim of this study was to descript a features of microcirculation of the layers of pyramidal cells in CA1, CA2/3 and DG region of hippocampus of TgF344-AD rats.

Materials and Methods

A year old wailed type (n=5) and TgF344-AD transgenic rats (n=6) were sacrificed and brains were collected. After standards histological process serials of 18-µm thick paraffin embed sections with horizontal orientation were prepared. Every thirtieth slice was stained by polyclonal rabbit anti-laminin antibodies to detect cerebellar microvessels. The stereology approaches [1] were used to collect the data. The total regional volumes of cell layers were estimated using the Cavalieri principle [2], (see Fig. 1A). The vessel numbers were founded using optical dissector [3, 4], (see Fig. 1C). The total length of microvessels was found using the space ball probe [5] (see Fig. 1D). All the quantitative estimations were done using the Stereologer 11 software (SRC Biosciences, Tampa, Florida, USA) including the running on a PC equipped with with a Nikon Eclipse Ti-U microscope which equipped with a standard line of lenses, camera (Promicra 3-3cc) and XYZ motorized stage.

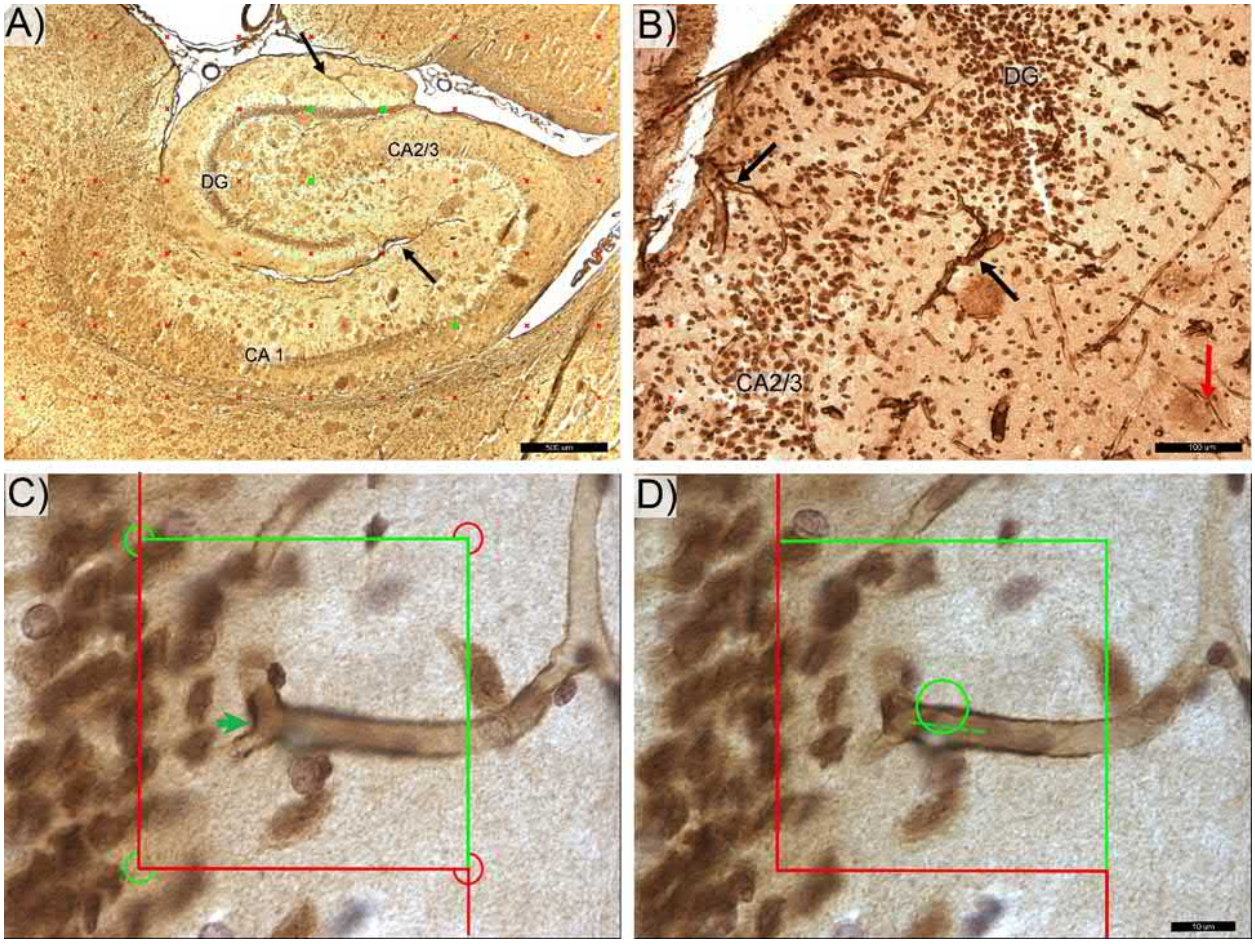

Fig. 1.: Photomicrograph of the anti-laminin antibody labeled hippocampal microvessels in control and transgenic rats. DG - stratum pyramidale of dentate gyrus; CA 2/3 - was combined CA3 and CA2 zones; CA1 - labeled respective zone. Space bars: A – 500 μm; B – 100 μm; C,D – 20 μm.

- A – A randomly positioned systematic-uniform grid of equidistant points (red) was generated by the software over each outlined region of interest. Green points captured the interesting object.
- B – The vessels serve as the main source of hippocampal blood supply shown by black arrows. Thinner hair-like capillaries (red arrows) are common in transgenic rats.
- C – The optical disector frame was used for quantifications of "capillary saddle points" (green arrow) at branch-points or nodes. The corners of the frame beyond the region of interest were excluded (red circles) from the analysis.
- The capillary length was found using the Space Balls method (D) by counting the number of intersections between surface of the sphere probe and a spline through the center of each capillary (dotted line).

Results and Discussion

The results (see Tab 42.1) revealed significant reductions in the total length of the capillary in DG of transgenic rats compared to non-transgenic littermates, with a pronounced trend towards a decrease in the total number of capillary segments. Mean capillary length is longer in transgenic rats (p=0.036) compared to non-transgenic rats. This trend is most pronounced in the CA1 area of the hippocampus. An increased

Table 42.1.: Complete quantitative parameters of the microvascular bed of hippocampus of non-Tg control and TgF344-AD rats.

	Total		CA 2/3		CA 1		DG	
	non-Tg	TgF344-AD	non-Tg	TgF344-AD	non-Tg	TgF344-AD	non-Tg	TgF344-AD
Volume (mm^3)	46.4 ±4.11	49.12 ±6.39	2 ±0.56	1.4 ±0.45	1.56 ±0.23	1.29 ±0.44	2.05 ±0.30	1.98 ±1.09
Nsegs	1228668 ±358030	932315 ±262556	77976 ±33907	90070 ±69728	253061 ±299613	28355 ±8444	69840 ±37760	45521 ±34532
Length (mm)	75992527 ±22812530	82800257 ±19085224	3545777 ±462590	4492204 ±3043746	2936692 ±529583	3083259 ±765560	3401354 ±1337359	1720442* ±290845
Diffusion distance (mm)	7.19 ±1.36	7.01 ±1.16	6.65 ±0.86	5.62 ±1.49	6.54 ±0.66	5.74 ±0.54	7.28 ±1.71	9.31 ±2.67

frequency of kinked, twisted, and string-like vessels has been found as well as in transgenic mouse AD models [6]. Nevertheless, the total number of capillaries in white matter (corpus callosum of the mice) is significantly reduced [7] in contrast with our results.

Conclusions

This study demonstrate the reduction of total capillary length on 49% in the DG region. DG seems to be the area contained the most expressed changes of microcirculation in TgF rats. Unlike the mice AD model we were unable to show a significant change in basal quantitative parameters of the microcirculation of the hippocampus of a year old TgF344-AD rats. These findings support the view that the expression of mutant human genes for beta-amyloid peptides alters the normal architecture of cerebral capillary vessels in the hippocampus of rat brain, which may model microvasculature changes reported in AD.

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