Diffusion and extracellular space volume in the cerebral cortex in a mouse model of Alzheimer's disease

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Introduction

Alzheimer's disease is accompanied by β-amyloid deposition, neuronal loss and changes in synaptic transmission. Symptoms such as forgetfulness, sleeplessness, anxiety and depression, may be related to impaired extrasynaptic transmission [1], which is based on the diffusion of substances in the extracellular space (ECS). We therefore studied diffusion in the cerebral cortex of transgenic APP23 mice, which develop a pathology similar to Alzheimer's disease. We employed the real-time iontophoretic tetramethylammonium (TMA) method to measure the ECS volume fraction (α = ECS volume/total tissue volume) and the apparent diffusion coefficient of TMA in the extracellular space (ADC_{TMA}). Although this method provides detailed information about diffusion in the brain, it cannot be used in humans. Diffusion-weighted (DW) MRI, which is widely available along with MRI scanners for clinical use, was used to determine the apparent diffusion coefficient of water (ADC_W) in the tissue, and the results were compared to TMA measurements.

Subjects and methods

Experiments were performed *in vivo* on 7- and 23-month-old hemizygous APP23 mice (both males and females) and age-matched controls (B6D2 mice). APP23 mice over-express the mutated human amyloid beta precursor protein (APP751, Swedish double mutation) and develop extracellular β-amyloid deposits [3]. The TMA method monitors the diffusion of the small TMA cation after its iontophoretic application. The local concentration of TMA^+ was measured in the primary somatosensory cortex using ion-selective microelectrodes [2]. TMA^+ diffuses predominantly in the ECS and therefore the TMA method measures the ECS diffusion parameters: α and ADC_{TMA}. The DW-MRI method measures the apparent diffusion coefficient of water in both the extracellular and intracellular compartments. Using a stimulated echo sequence, we acquired 4 axial slices with the following parameters: FOV 2 cm, TR 1.2 s, TE 45.7 ms, 0.8 mm slice thickness, 0.4 mm gap, 256 x 128 image matrix, slice diffusion gradient direction and 6 b-values (136 - 1826 s/mm^2). ADC_W maps were evaluated in the somatosensory cortical region (Fig. 1). Additionally, fixed brain sections were stained using the polyclonal antibody NT12, directed against β-amyloid, to assess the plaque load.

Results

In 7-month-old APP23 mice, the mean ECS volume fraction, ADC_{TMA} and ADC_W were not significantly different from age-matched controls (α = 0.201 ± 0.004, ADC_{TMA} = 594 ± 10 μm^{2}s^{-1}, ADC_W = 600 ± 8 μm^{2}s^{-1}). No differences between males and females were seen. Aging in 23-month-old controls was accompanied by a decrease in the ECS volume fraction, significantly more in females (α = 0.132 ± 0.006) than in males (α = 0.161 ± 0.008). In contrast, the deposition of β-amyloid in aged, 23-month-old APP23 mice was associated with an increase in ECS volume fraction (0.224 ± 0.008) and with a decrease in ADC_{TMA} (565 ± 5 μm^{2}s^{-1}), compared to age-matched controls. The increase of ECS volume fraction was proportional to the amyloid plaque load. Significant changes in ADC_W during aging in control and APP23 mice were observed only in females, who generally showed a greater change in ECS volume fraction than males. The values of ECS volume fraction, ADC_{TMA} and ADC_W in females are summarized in Table 1.

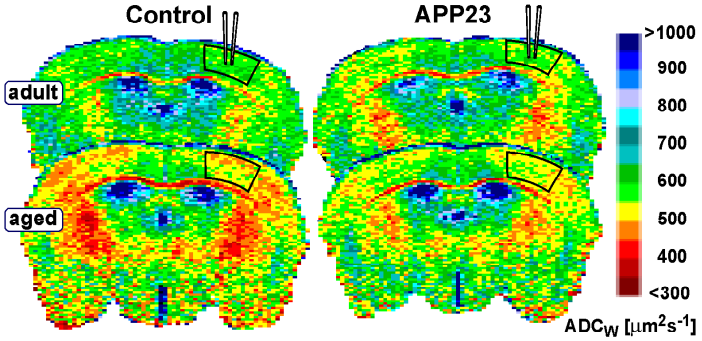

Fig. 1: ADC_W maps acquired in the brain of control and APP23 females. The mean values of ADC_W were calculated in the delineated areas. Thereafter, microelectrodes were inserted in the same region to perform TMA measurements.

	α	ADC_{TMA} (x 10^{-6}mm^{2}s^{-1})	ADC_W (x 10^{-6}mm^{2}s^{-1})
7-month-old control mice	0.199 ± 0.006 n = 6	602 ± 16 n = 6	602 ± 10 n = 6
23-month-old control mice	0.132 ± 0.006* n = 11	606 ± 9 n = 11	547 ± 8* n = 7
7-month-old APP23 mice	0.194 ± 0.009 n = 7	599 ± 5 n = 7	597 ± 15 n = 8
23-month-old APP23 mice	0.226 ± 0.011† n = 13	553 ± 9*† n = 13	574 ± 9† n = 8

Table 1: Table shows the diffusion parameters in aged control and APP23 females. Data are expressed as mean ± S.E.M. Significant differences (two-tailed Student's t-test, p<0.05) compared to 7-month-old (23-month-old) control mice are marked with asterisks (crosses).

Discussion and conclusion

Our data show that aging leads to a decrease in the ECS size (α) in the cerebral cortex of mice. Similarly low values of α were previously observed in aged rats [4]. However, the diffusion of small molecules in the ESC (represented by TMA^+) remained unchanged during normal aging. In transgenic APP23 mice, in which aging is associated with the development of β-amyloid plaques, the ECS volume fraction increased. These changes were more pronounced in females than in males, which may correspond to sex differences in disease progression. In addition, aged APP23 females showed an increase in ECS diffusion barriers (manifested by an ADC_{TMA} decrease). ADC_W changes predominantly corresponded to the variations in ECS volume. Altered ECS size and diffusion may influence synaptic as well as extrasynaptic transmission and thus lead to impaired brain function.

References and acknowledgement

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Supported by grants AVOZ5039906, LN00A65 and J13/9811130004.