T2* quantification can differentiate Alzheimer's plaques burden following anti-amyloid therapy

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Introduction.

Amyloid β plaques (Aβ) are one of the main hallmarks of Alzheimer's disease (AD). A non invasive method that would accurately estimate Aβ plaques longitudinally would be very valuable for testing and monitoring ongoing therapeutical strategies aimed at reducing amyloid burden in transgenic mouse models of AD. *In Vivo* mouse MRI approaches have enabled Aβ plaque visualization using both endogenous contrast when enough iron accumulates in the plaques (1), and exogenous contrast by magnetically labeling the Aβ plaques using Aβ based peptides as targeted tagging contrast agent (2). Despite this important progress to specifically delineate individual plaques, a significant portion of the plaque population remains unaccounted for when assessing the amyloid burden (1) and thus preclude its current practical use for precise measurement of Aβ brain loads. In this study, we tested whether T2* quantification could be sensitive enough to distinguish relatively small differences in amyloid plaque burden. Two age-matched transgenic mouse groups with histologically confirmed differences in Aβ plaques burden were used. The first group was treated with memantine, a neuroprotective agent previously shown to decrease Aβ proteins levels (3) and compared to a vehicle-treated AD mouse control group. Both groups were injected with the gadolinium labeled tagging ligand 4-h prior to euthanasia and perfusion for subsequent imaging and histology.

Methods.

Animals, Treatment and Histology We used two groups (7 mice/group) of 2-3 month old transgenic APP/PS1 mice. Progressive cerebral Aβ deposits and associated pathology in these animals begins at approximately 6-8 weeks of age. Each group was treated with either memantine (10mg/kg/day; ip) or vehicle (water) for a period of 4 months. At the end of the treatment, Gd-Aβ1-40 peptide was subsequently administered in conjunction with mannitol, via intracarotid injection, to label the plaques i*n vivo* in the brain of both mouse groups. After a 4-hour post-injection delay all mice were anesthetized and perfused. Brain were extracted and subjected to ex vivo μMRI. Aβ burden (% area occupied by Aβ) was quantified from histological brain sections with Aβ immunostaining.

MRI: All experiments were assessed with a 7T SMIS/Magnex system (gradient 250mT/m, 200μs rise time). A litz coil (Doty Scientific: ID=25mm, length=22 mm) was used to incorporate a 30cc syringe (OD=24mm, ID=20.5mm) in order to increase the throughput up to four extracted brains, glued into place in each quadrant of the syringe plunger and immersed in fomblin. A T2*-weighted 3DGE sequence was used to provide image data sets with 50μm isotropic resolution optimized for plaque visualization. A 2D-T2* maps was acquired as well (4 echoes, TR=1.5s, TE=7.24, Echo spacing ES=7.5ms, FA=55°, 100μmx100μmx250μm, 1-h).

Results and Discussion: 50μ resolution scans of both mouse groups at 6-7 months of age revealed few visually evident Aβ plaques. More dark spots could be seen in water-treated mice (Fig.2A, red arrows) compared to memantine-treated mice (Fig.2B). Immunostaining confirmed the visual findings in the MRI with stereological documentation of a significant difference in Aβ burden comparing vehicle-treated mice to memantine-treated mice in both brain regions (fig 3 A&B, hippocampus: *p=.0219; cortex: *p=.0469). When T2* quantification was used, a significant difference was obtained in the cortex (*p=.04). In the hippocampus, difficult in delineating the anatomical boundaries by MRI limited the comparison. In conclusion : T2* mapping in combination with targeted tagging contrast agents can be an excellent global approach to longitudinally evaluate the Aβ burden in mice. Ongoing *in vivo* experiment are testing this approach.

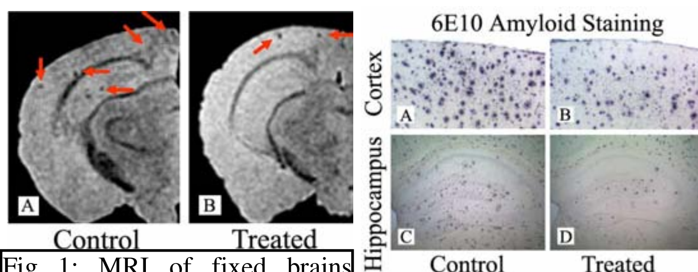

Fig 1: MRI of fixed brains (50μm isotropic resolution TR=50ms, TE=15ms, FA=18°, 14h35min). Both (A) water- and (B) memantine-treated mice reveal few plaque enhancement seen as dark spots (read arrow).

Fig 2: Immunostaining reveals abundant Aβ plaques (dark spots) both in the cortex (A&B) and hippocampus (C&D). Note the difference in plaque density between structures and brains.

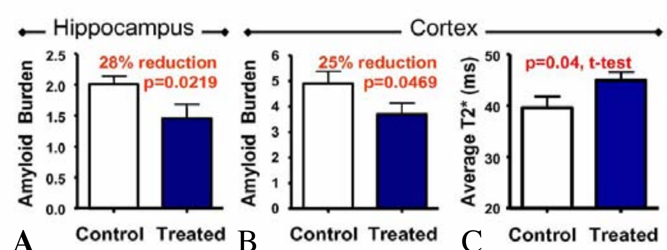

Fig 3: Aβ plaque burden quantification from brain histology show a significant statistical difference between control (water-treated) and memantine-treated mice. T2* quantification demonstrated a significant difference in the cortex but not in the the hippocampal region that may be explained due to partial volume effect of this compartmental brain structure.

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References:1. Jack CR *et al.*, MRM 2004,52(6):1263; Vanhoutte G *et al.*, MRM 2005,53(3):607. **2.** Wadghiri YZ *et al.*, MRM 2003,50(2):293-302; Wadghiri YZ *et al.*, Meth Mol Biol 2005;299:365 **3.** Lahiri DB *et al.,* 34th Meet. Soc for Neurosci, 2004; San Diego