Discoid Shaped Multimodal Nanoprobes to Target Cerebrovascular Amyloid

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Introduction

Cerebral amyloid angiopathy (CAA) results from the accumulation of Aβ proteins primarily within the media and adventitia of small arteries and capillaries of the cortex and leptomeninges [1]. CAA affects a majority of Alzheimer's disease (AD) patients and is associated with a rapid decline in cognitive reserve. Unfortunately, there is no pre-mortem diagnosis available for CAA. However, MRI has emerged as a tool to image and identify AD in animal models. Furthermore, treatment options are few and relatively ineffective. This work builds upon previously reported and published work aimed to design nanovehicles (nanoparticles-IgG4.1) capable of targeting cerebrovascular amyloid (CVA) and serving as early diagnostic and therapeutic agents [2,3]. Here we have modified the nanovehicle for increased MRI detection and labeling efficiency.

Experimental

Magnevist (Dy-DTPA), a MRI contrast agent was conjugated to chitosan using carbodiimide reaction which was then dialyzed against water to remove free Magnevist. Chitsoan-polycarbophil nanoparticles were then prepared by slowly adding anionic charged polycarbophil to Magnevist-chitosan at pH 3.5 to form the polyelectrolyte complex. An anti-amyloid antibody (IgG4.1) was then conjugated to the surface using carbodiimide reaction enable it to target CVA deposits. The nanovehicles were also loaded with anti-inflammatory agent (cyclophosphamide) which were encapsulated with a cyclophosphamide/chitosan solution and added to the polycarbophil solution and allowed to equilibrate for 30 min prior to the formation of nanovehicles. This modified nanovehicle was shown to have a flat discoid shape, as seen using atomic force imaging. To maintain this shape and to prevent the nanovehicle from falling apart when the pH is raised, the chitosan was cross-linked to the polycarbophil during the carbodiimide reaction. The nanovehicles were then characterized with animal models of (AD). Using the 21.1-T (900 MHz) magnet at the NHMFL, and the 11.75 T magnet at the FSU-FAMU college of engineering the nanovehicle was evaluated using high resolution T_2* weighted MRI sequences on *ex vivo* mouse brains to assess the nanovehicle potential to target CVA.

Results and Discussion

Previous work with a similar particles show successful passing from the blood flow to the vascular wall and targeting of the CVA (Figure 1) [2,3]. The modified particle described in this work show increased labeling efficiency at 21.1 T as seen by the dark voids in the cortex and hippocampus in Figure 2. In addition similar contrast is also seen at the lower 11.75 T field strength showing its potential for translational clinical research.

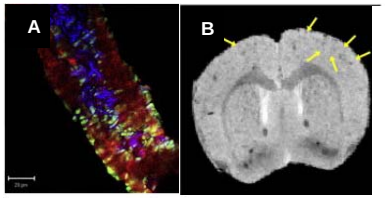

Figure 1 a: Uptake of nanovehicles (red) of a Alzheimer's animal model. **b:** Ex vivo MRI of an Alzheimer's mouse injected with the Gd-conjugated nanocvehicles and showing the subsequent MRI

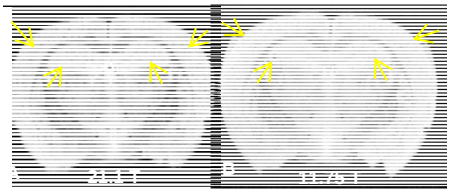

Figure 2: Ex vivo MRI at 21.1 (a) and 11.75 T (b) showing increased labeling efficiency of the modified nanovehicle as presented in this paper using a Alzhimer's mouse model.

Conclusions

This paper demonstrates our successful modified design of a multi-modal nanovehicle and demonstrates its ability to target CVA deposits and increased MRI contrast. This nanovehicle demonstrates the potential for translational research to lower clinical field strengths and for detection of CVA in humans. Histological investigation is underway to confirm the MRI findings and the effectiveness of the immunosuppressant.

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References [1] Calhoun, M.E., *et al*., Proc Natl Acad Sci USA, **96**, 14088-93 (1999). [2] Jaruszewski, K.M., *et al.,* Biomaterial, **35(6),**1967-76 (2014). [3] Agyare, E.K., *et al.,* Journal of Controlled Released,**185**, 121-129 (2014).