Trans-Blood-Brain Barrier Drug Delivery via Ultrasound and Microbubbles for Neurodegenerative Diseaseses

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Introduction

Treatment of neurological disorders is often hampered by the inability of drugs to cross the blood-brain barrier (BBB). Over the last several years, novel techniques have been developed to temporarily open the BBB, allowing therapeutic agents to enter the brain. A novel technique uses ultrasound (US) energy in combination with microbubble (μB) contrast agents to reversibly open up the BBB [1-3]. Foundational studies have been carried out in several animal models, including mice [4-8]. BBB opening is readily verified with MRI using gadolinium contrast agents. To comprehensively evaluate these methods for drug delivery, it will be extremely useful to directly image the distribution of drugs in the brain in vivo. With this goal in mind we have initiated studies that combine US-mediated BBB opening with high-resolution γ-ray imaging of radiolabeled molecules. The technique is demonstrated in a mouse model of Niemann-Pick type C (NPC) disease, a childhood disease that involves errors in cholesterol trafficking which results in neurodegeneration.

Methods

Mice were imaged prior to BBB opening in a 7T Bruker Biospec MRI system. A 72 mm ID birdcage coil was used for excitation and a 4-channel phased array coil was used for reception. The mice were secured in an MRI cradle with ear bars and a bite bar. Rapid whole-brain 3D T1-weighted GRE images (5 minute acquisition) were obtained prior to and after IP injection of Gd-DTPA. BBB opening utilized a 40 μL bolus of custom gas filled μBs, similar to Definity® (Lantheus Medical Imaging, Inc.), that were injected into the tail vein, followed by a 120 μL saline flush. Immediately after the injection, 3.3 MHz US was applied to the brain for 3 minutes (37% duty cycle, 6 ms pulse width, 0.52 MPa peak negative pressure) with a 3 second pause every 30 seconds. A custom built positioning apparatus (Fig. 1) was used to position and hold the transducer while the mice remained secured in the cradle. After US, the mice were returned to their original position in the MRI magnet. The same T1-weighted imaging was initiated within 5 minutes after US and repeated 6 times. After this, a higher resolution image was obtained using a 3D-FSE sequence. Other mice underwent the same procedure, except that no US was applied. Mice were allowed to recover and showed no obvious deficits in neurologic function. Within 3 hours of the MRI procedure, two mice were injected with ^{123}I-tyr-beta-cyclodextrin (^{123}I-BCD) and imaged simultaneously using a custom-built γ-ray imaging system. BCD is a promising treatment for NPC disease when it can be delivered directly to brain tissue.

Results and Discussion

MRI images before and after the administration of Gd-DTPA, μBs and US are shown in Fig. 2. There is visible increase in signal in the brain parenchyma in the experimental mouse that received US compared to the control mouse that just received Gd-DTPA and μBs. Percent signal enhancement maps from the two mice are shown in Fig. 2c and indicate opening of the BBB to Gd-DTPA in the exprimental mouse. γ-ray scintigraphy images of the two mice are shown in Fig. 3. An increase in signal is seen in the brain region of the experimental mouse compared to the control. This increase was validated by a greater than twofold higher residual radioactivity in the extracted brain of the experimental mouse (242.0 kcounts/min/g) compared to the control mouse (95.1 kcounts/min/g). This shows that the BBB opening procedure allows passage of ^{123}I-BCD into the brains of mice. While these experiments are directed towards NPC disease, they could have a significant impact on other common neurological disorders (e.g. Alzheimer's and Parkinson's).

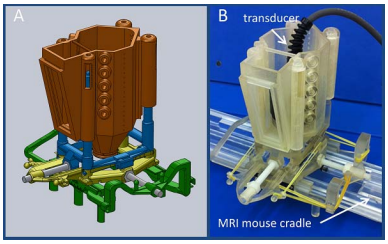

Fig 1. *An apparatus that couples US transducer beam to mice brains and allows for 3D translation. (A) SolidWorks rendering. (B) Photograph of printed apparatus (generated by a 3D polymer printer) attached to a MRI mouse cradle.*

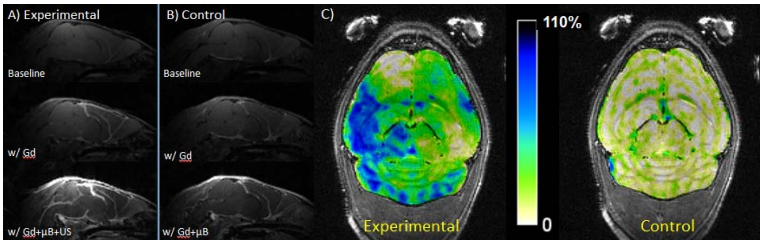

Fig 2. *(A) MRI images of a mouse brain obtained at baseline, 10 minutes after the IP injection of Gd-DTPA (w/Gd) and 20 minutes after the injection of 40 μL solution of μB followed by the application of 3 minutes of US. (B) MRI images of control mouse that did not receive US. (C) Percent signal enhancement maps from Gd-DTPA of mouse that underwent the ultrasound opening procedure and a control mouse that did not receive US.*

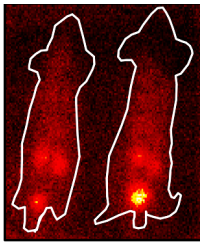

Fig 3. *γ–ray image 30 minutes post ^{123}I-BCD injection. Increased intensity is seen in the brain region of the experimental mouse (left) compared to the control mouse (right).*

References

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