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ENHANCEMENT OF GAS-FILLED MICROBUBBLE MAGNETIC SUSCEPTIBILITY BY IRON OXIDE NANOPARTICLES

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INTRODUCTION
Gas-filled microbubbles were originally developed as an intravascular contrast agent to enhance backscattering in ultrasound imaging. Microbubbles possess the ability to be an MR susceptibility contrast agent due to the induction of large local magnetic susceptibility differences by the gas-liquid interface. Feasibility of microbubbles as an MR pressure sensor, based on the susceptibility change caused by pressure-induced microbubble size change, has been explored through theoretical1,2 and phantom studies. Gas-filled microbubbles have also been shown as an MR susceptibility contrast agent in vivo.3 However, microbubble susceptibility effect is relatively weak when compared with other intravascular MR susceptibility contrast agents. By optimizing the microbubble size distribution and choice of shell coating material and core gas, it is possible to substantially enhance the microbubble susceptibility effects1 and reduce the dosage requirement for MR applications. In this study, we aim to demonstrate that microbubble susceptibility effects can be improved by embedding and entrapping iron oxide nanoparticles.

METHODS
Synthesis of iron oxide nanoparticles embedded albumin-coated microbubbles: Iron oxide nanoparticles embedded albumin-coated microbubbles (AMB) were produced by an adapted sonication method4. Briefly, 18 mg of monocrystalline iron oxide nanoparticles (MION; MGH) was added into a 5% solution of bovine serum albumin (10857, USB Corporation). The mixture was preheated to about 70°C and sonicated under aspetic conditions using an ultrasound frequency of 20 kHz.

Synthesis of iron oxide nanoparticles entrapped polymeric microbubbles: Iron oxide nanoparticles entrapped polymeric microbubbles (polymeric MB) were produced by an adapted double emulsion method. Briefly, 0.5 g poly(D,L-lactide-co-glycolic acid 50:50, PLGA; Sigma) was dissolved in 10 mL of ethyl acetate (Sigma). 1 mL of MION solution (1.164mg/mL) was added to the polymer solution and sonicated for 30 s. The W/O emulsion was then poured into a 5% poly(vinyl alcohol) (PVA; Sigma) solution and homogenized for 5 min. The double (W/O)W emulsion was then poured into a 2% isopropyl alcohol (Sigma) and stirred at room temperature for 1 hour. The capsules were collected by centrifugation, washed once with distilled water, centrifuged at 15°C for 5 min, at 3000g and the supernatant discarded. The capsules were then washed three times with hexane (Sigma). The capsules were frozen in a -80°C freezer and lyophilized using a freeze dryer to fully dry the capsules and sublimate the encapsulated water. MRI and Data Analysis: All MRI experiments were performed on a 7 T Bruker MRI scanner. Microbubble phantom study was performed with 38-mm quadrature resonator for RF transmission and receiving. AMB were diluted from a well-mixed microbubble suspension to 4% volume fraction. Gas-filled microbubbles were prepared by adding saline of 2 mL to 50 mg of the lyophilized powder. The microbubbles were then placed in separate 2-mL cylindrical phantom tubes. Each phantom tube was slowly warmed to room temperature and gently mixed for 2 min outside the magnet prior to MR measurements. To ensure uniform suspension of microbubbles, the phantom was then continuously stirred by rotation inside the magnet. It was then arrested in horizontal position immediately before the start of MR acquisition sequence. Apparent transverse relaxation rate enhancement (ΔR2) was measured by acquiring multi-echo gradient echo (GE) images in GE-EPI sequence. The GE-EPI sequence was acquired with the following parameters: TE = 10 ms, TR = 3.5, 7, 10.5, 14, 17, 21, 24.5, 28 ms, flip angle = 30° and NEX = 1. Phantom R2* values were computed by monoexponential fitting of the peak magnitude of the multi-echo GE signals using a software toolkit developed in MATLAB (MathWorks). Initially, there was a uniform suspension of microbubbles. As GE signals were acquired, microbubbles started to migrate upward; therefore, in the final state the microbubbles aggregated in the upper part of the tube. Microbubble induced ΔR2 was then calculated as the difference between R2* in the initial state and that in the final state. To demonstrate that MION were embedded and entrapped, R2* was measured before and after cavitation, which was performed by applying ultrasound of frequency 40 kHz. R2* maps of the suspending solutions were acquired before and after cavitation with multiple gradient echo sequences.

RESULTS AND DISCUSSIONS
Values of R2* were plotted against time in Figure 1 for different microbubbles. As GE signals were acquired, microbubbles started to migrate upward; therefore, in the final state values of R2* were due to the suspending solution. The different amounts of free MION in the suspended solution accounted for the difference in the R2* of the suspending solution. Microbubble induced ΔR2 of different microbubbles were depicted in Figure 2. The MION embedded and entrapped would enhance the susceptibility effect and increased the values of ΔR2 as MION would float along with the microbubbles, resulting in a larger ΔR2 values. Similarly, microbubble induced ΔR2* for AMB without and with MION after cavitation were found to be increased by 1.41 s^-1 and 23.91 s^-1 respectively. Figure 4(a) shows one of the preinjection GE-EPI T2*-weighted images, while (b) shows the postinjection GE-EPI T2*-weighted image with the maximum susceptibility contrast for polymeric MB with MION injection. Figure 5 depicts the T2*-weighted signal time course during polymer MB with MION injection with ROI selected from homogeneous liver region (indicated in Figure 4(a)). Similar signal time course was also observed during AMB with MION injection. The T2*-weighted signals after microbubble injection did not return to the preinjection baseline, which may be caused by accumulation of released MION in Kupffer cells after in vivo microbubble clearance.

CONCLUSIONS
In this study, we demonstrated, for the first time, that embedding iron oxide nanoparticles onto shells of albumin-coated microbubbles could greatly enhance iron oxide microbubbles’ susceptibility. This work is currently underway to characterize the in vivo susceptibility effects of these microbubbles. With such approach, microbubble susceptibility can be significantly enhanced, so that microbubbles can be monitored with high sensitivity and low concentrations under MRI.

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REFERENCES

Figure 1 Values of R2* were plotted against time for different microbubbles.

Figure 2 Microbubble induced ΔR2* of different microbubbles. The error bars represent one standard deviation.

Figure 3 Transmission electron micrograph shows MION was embedded onto shells of AMB.

Figure 4 GE-EPI T2*-weighted image for polymeric MB w/ MION (a) preinjection image and (b) postinjection with the maximum susceptibility contrast.

Figure 5 T2*-weighted signal time course in a homogeneous liver region during polymeric MB w/ MION injection.