INTRODUCTION

Gas-filled microbubbles possess the potential to become a unique MR contrast agent because of their magnetic susceptibility effect, high biocompatibility and unique cavitation and sonoporation properties. However, in vivo demonstration of microbubble susceptibility effect is limited so far. In this study, we aim to further demonstrate and characterize the in vivo MR susceptibility effect induced by using both custom-made albumin-coated microbubbles (AMB) and commercially available lipid-based clinical ultrasound microbubble contrast agent (SonoVue®) in rat livers at 7 Tesla using dynamic susceptibility weighted MRI.

METHODS

Microbubbles Preparation: Air-filled AMBs were produced by sonication as previously described. SonoVue® microbubbles (Bracco) are commercially available, which consist of sulphur hexafluoride gas stabilized in aqueous dispersion by phospholipid monolayer. Animal Procedures: Normal SD rats (~250-350 g) (n = 5) were used. Femoral vein catheterization was performed with a 1-n long tube connected to a 27-gauge needle under anesthesia using IP injection of ketamine (100 mg/kg) and xylazine (10 mg/kg). The dead space in the catheter was about 0.2 mL. During MRI, animals were anesthetized with isoflurane/air using 1.0-1.5% via a nose cone. Microbubbles Administration: Microbubbles were first warmed slowly to room temperature and mixed to achieve uniform suspension. For each imaging session, 0.2 mL of microbubble suspension (~4% volume fraction for AMB and ~3.5% volume fraction for SonoVue®) was slowly injected at a rate of 1.2 mL/min to avoid possible microbubble destruction due to high pressure and shear stress. MRI: All MRI experiments were performed on a 7 T Bruker MRI scanner using a 60-mm quadrature RF coil. Anatomical images were acquired with 2D FLASH sequence with resolution = 0.20 × 0.20 × 2.0 mm³. Dynamic susceptibility weighted liver MRI was performed with respiratory-gated single-shot GE-EPI sequence using TR = 1000 ms, TE = 10 ms, FA = 90º, FOV = 50 mm × 50 mm, slice thickness = 2 mm, acquisition matrix = 64 × 64, BW = 221 kHz and NEX = 1. To allow sufficient clearance of the microbubbles before the next injection, a minimum lapse of 10 min was used. The susceptibility effect of microbubbles was compared with that of a well-established intravascular contrast agent, monocrystalline iron oxide nanoparticles (MION; MicroThermics), by single dose of 0.6 mg Fe/kg injection using identical injection protocol and imaging sequence. Image Analysis: GE-EPI images were first co-registered using AIR5.2.5. AR₀ maps were computed on a pixel-by-pixel basis as AR₀ = ln (S₀/S₀(pre))/TE, where S₀ is the average intensity in 100 preinjection images and S₀(pre) is the average intensity in 40 postinjection images with maximum susceptibility contrast for microbubbles (or 100 postinjection images at steady-state contrast for MION). To quantify the AR₀ values, ROIs were manually drawn in homogeneous liver region (LV) and the region covering inferior vena cava (IVC) based on the high resolution FLASH images. Assuming that AR₀ is proportional to microbubble concentration C(t) at time t, C(t) can be estimated as C(t) = k ln (S₀/S₀(i))/TE + C₀, where S₀(i) is the intensity at time t, k a proportionality constant, and C₀ a constant residue to account for any postinjection baseline. Given the relatively low injection time and the limited lifetime of microbubbles in vivo, C(t) were approximately modeled with a gamma-variate function by curve fitting. Full width at half maximum (FWHM) and time-to-peak were then measured from the fitted C(t) time courses.

RESULTS AND DISCUSSIONS

Figure 1 shows the in vivo measurements of AR₀, FWHM and time-to-peak of AMB and SonoVue® as well as the AR₀ and time-to-peak of MION in liver among all rats studied. The similar AR₀ susceptibility effects of AMB and SonoVue® at the dosage used are comparable to that of 0.6 mg Fe/kg MION in liver tissue at 7 T. However, time-to-peak was found to be shorter for MION. This is largely expected as microbubbles, with size comparable to that of red blood cells, flow slower than blood plasma while nanosized MIONs flow together with plasma. In few of the rats studied (2 out of 5), the T₂*-weighted signals after microbubble injection did not return to the preinjection baseline, this may be caused by microbubble trapping in local tissue vasculature. Possible uptake of intact microbubbles by Kupffer cells in liver may also contribute to such observation.

CONCLUSIONS

Substantial susceptibility induced changes were observed and characterized using gas-filled microbubbles for MRI at 7 T in rat livers, using custom-made albumin-coated microbubbles and a commercially available clinical ultrasound microbubble contrast agent. With the increasing availability of high-field MRI systems in both clinical and research setting, gas-filled microbubbles offer the promise as a viable in vivo MRI contrast agent. With the potential for MRI guidance of microbubble-based drug delivery and therapies, Microbubble fabrication technology is also advancing. For example, substantially increased in vivo lifetime has been demonstrated recently using surfactant molecules with multiphase mixing technique. Lastly, molecular targeting capability can also be achieved by microbubble surface modification.

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REFERENCES


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