Acceleration of ultrasound thermal therapy by patterned acoustic droplet vaporization

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One application of acoustic droplet vaporization (ADV), a method of converting biocompatible microdroplets into microbubbles, is to enhance locally high intensity focused ultrasound (HIFU) therapy. Two objectives are pursued here: (1) the controlled creation of a bubble trench prior to HIFU using ADV and (2) use of the trench for increasing ablation volumes, lowering acoustic powers, and decreasing therapy duration. Thermally responsive phantoms were made with perfluorocarbon emulsion. Compound lesions were formed in a laboratory setting and a clinical magnetic resonance imaging (MRI)-guided HIFU system. Linear and spiral patterned compound lesions were generated in trenches. A larger fraction of the HIFU beam is contained to increase the generation of heat. Using the laboratory system, a 90 mm linear length spiral trench was formed in 30 s with mechanical beam steering. Comparatively, the clinical HIFU system formed a 19.9 mm linear length spiral trench in approximately 1 s with electronic beam steering. Lesions were imaged optically and with MRI. A uniform thermal ablation volume of 3.25 mL was achieved in 55.4 s (4-times faster than standard clinical HIFU and 14-times larger volume versus sum of individual lesions). Single lesions showed a 400% volume increase.

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I. INTRODUCTION

High intensity focused ultrasound (HIFU) therapy is a non-invasive and non-ionizing technique wherein focused ultrasound beams are emitted from a high-powered transducer to thermally ablate the tissue volume of interest (Crouzet *et al.*, 2010). HIFU systems commonly operate in a frequency range of 0.5–5 MHz, generating high focal intensities up to 1–10 kW/cm² (Shaw and Hodnett, 2008). Such intensities can increase tissue temperature beyond 60 °C within the focal volume in seconds, resulting in irreversible protein denaturation, cell destruction, and associated coagulative necrosis (Hill and ter Haar, 1995). Two principal mechanisms, direct absorption of the transmitted pressure wave and acoustic cavitation, can cause tissue heating synergistically (Coussios and Roy, 2008). The bioeffects of focused ultrasound on tissues have been investigated for decades (Bamber and Hill, 1979; Parker, 1983; Frizzell, 1988) and the physical principles associated with HIFU are becoming better understood as HIFU becomes an accepted modality for treatment (ter Haar and Coussios, 2007).

Focused HIFU energy is sufficient to induce thermal coagulation in a small volume within seconds, and therefore blood perfusion effects acting as thermal sinks are minimal (Billard *et al.*, 1990). Although promising and effective for small tumors, attempts to expand the application of HIFU thermal ablation to larger masses have had limited success (Yamakado *et al.*, 2006; Gervais *et al.*, 2003). A major difficulty has been creating homogenous, reproducible, and uniformly shaped lesions in a time period that is clinically manageable for treating large volumes. In addition, some patients complain about local pain after HIFU therapy, which indicates normal tissue overheating, especially at sites of large acoustic impedance mismatch (Li *et al.*, 2009; Zhang *et al.*, 2010b). Therefore, methods localizing treatment and thus protecting sensitive tissues would greatly benefit HIFU and potentially yield new treatment locations.

It is well known that the thermal contribution of microbubbles can be significant. Ultrasound contrast agents (UCAs) have been investigated for the enhancement of thermal effects at the HIFU focus (e.g., Tung *et al.*, 2006). However, UCAs also move the greatest heating position from the focus by up to 2 cm for a given transducer geometry and aberrating tissues intervening the acoustic path as well as create surface lesions for concentrations greater than 0.1% (v/v), with some damage well outside the lesion (Tung *et al.*, 2006). Alternatively, acoustic droplet vaporization (ADV) is an ultrasound-based method of converting biocompatible microdroplets (Fabiilli *et al.*, 2013) into microbubbles. When exposed to acoustic pressures above the

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vaporization threshold, injected perfluorocarbon microdroplets—which are similar in size to UCAs—produce microbubbles that are more than 100-fold larger in volume than the original microdroplets, resulting in a local increase in the acoustic absorption. Hence, the increased heating with ADV can be limited to the acoustic focus where pressures exceed the ADV threshold, allowing for lower source intensities and/or shorter exposure times for given therapeutic effects at the focus. Increased absorption by bubbles at the focus also reduces risk to distal tissues by decreasing the transmitted acoustic energy (Lo *et al.*, 2006).

ADV bubbles are stable for several minutes or longer and can cause vascular occlusion when lodged in a capillary or other small vessels (Kripfgans *et al.*, 2000), which is favorable for HIFU therapy. These microbubbles are highly echogenic (Zhang *et al.*, 2010a, Kripfgans, 2002), thus facilitating real-time image-guided therapy. Therefore, the ultrasonic monitoring of microbubbles created during the ADV process may offer opportunities to visualize the location and spatial extend of the HIFU process. Additionally, a dense bubble plane forms a barrier for acoustic waves and impedes wave propagation to distal tissues and can double pressure amplitudes proximal to the bubble wall (Lo *et al.*, 2006). Similar studies by Zhang *et al.* (2013) demonstrated *in vitro* that the inclusion of 0.008% (v/v) phase-shift nanoemulsion (PSNE) in phantoms significantly reduced the power required to generate 9 mm³ lesions compared to HIFU without PSNE.

This study is focused on substantially enhancing HIFU by increasing thermal lesion volume (or equivalently shortening treatment time) through precisely controlled production of ADV microbubbles from micrometer-sized droplets. This technology is expected to be applicable across a broad range of current and future uses of HIFU ablative therapy.

II. MATERIALS AND METHODS

A. Thermal phantom preparation

Thermally responsive phantoms were fabricated based on previously described methods (Takegami *et al.*, 2004). The phantoms consisted of 33% (v/v) of an aqueous acrylamide solution (30% wt/vol, Sigma-Aldrich, St. Louis, MO), 31.4% (v/v) of degassed deionized water, 35% (v/v) of egg white, 0.5% (v/v) of 10% ammonium persulfate (Sigma-Aldrich), and 0.1% (v/v) tetramethylethylenediamine (Sigma-Aldrich). Lipid droplets were made according to a previously described protocol (Fabiilli *et al.*, 2009) and stored in the refrigerator (5 °C) for up to 4 weeks. Prior to use, the droplet size distribution and number density were measured with a Coulter counter (Multisizer 3, Beckman Coulter Inc., Fullerton, CA). Approximately 99% of droplets in the emulsion were smaller than 8 μm in diameter. The mean diameter of the droplets was 2.0 ± 0.1 μm.

During phantom preparation, the gel solution was degassed before the addition of droplets to minimize spurious gas inclusions that could act as cavitation nuclei. The concentration of droplets within the phantom was 10⁵ droplets/mL, which corresponds to 0.00004% (v/v). These egg white-based phantoms were optically transparent. If HIFU increased the

temperature in a portion of the phantom to >60 °C for >5 s, the egg white protein would denature and coagulate, resulting in a permanently opaque, optically observable lesion. The acoustic attenuation of these phantoms was approximately 0.3 dB/cm at the resonance frequency of our HIFU transducer (1.44 MHz), which is also very close to the frequency of the clinical HIFU system of 1.45 MHz.

B. Laboratory HIFU lesion generation

The HIFU therapy system consisted of a high power spherical section transducer (63.5 mm diameter, f-number of 1, resonance frequency 1.44 MHz, Etalon 940501, Lebanon, IN) driven with a power amplifier (A-300, ENI Inc., Rochester, NY). Low power pulse-echo operation of the HIFU transducer allowed for focal localization in the phantom by detection of phantom boundaries by use of a micro positioning system. Also under pulse-echo guidance, a needle thermocouple (300 μm diameter, Omega, Stamford, CT) was inserted through a guide into the side of the phantom 2 mm away from the transducer focus, and a data logger (TC-08, Pico Technology, Cambridge, UK) was used to record the temperature. A function generator (3314A, Agilent, Palo Alto, CA) was gated with a secondary function generator (33120A, Agilent) to produce continuous wave (CW) signals to the amplifier for ultrasound exposures with durations of 5 s. The lateral beam width and axial depth of field at the focus [full-width at half maximum (FWHM) pressure] were 1.6 mm and 8.2 mm, respectively. The therapy transducer was calibrated using a membrane hydrophone (ST223, Sonic Industries, Hatboro, PA) in degassed water at room temperature, and the output waveforms from the hydrophone were digitized (9314L, LeCroy Corp., Chestnut Ridge, NY). A peak negative pressure of 7.4 MPa and a peak positive pressure of 17.7 MPa were measured at the focus. The maximum focal intensity was then estimated from the measured peak positive pressure (Shaw and Hodnett, 2008) by approximating the 2D acoustic beam profile with a Gaussian function and assuming linear acoustic (first order assumptions)

$$p(x, y) = p_+ e^{-(x^2/2\sigma_x^2 + y^2/2\sigma_y^2)},$$

$$P_{\text{acoustic}} = \iint_{-\infty}^{\infty} \frac{p(x, y)^2 dx dy}{Z},$$

$$I_{\text{SPTP}} = \frac{P_{\text{acoustic}}}{\pi(FWHM/2)^2},$$

where $p(x, y)$ is the focal plane pressure distribution, p_+ is the peak positive pressure amplitude of 17.7 MPa, σ_x and σ_y are related to the FWHM beam width by $\sigma_{x/y} = FWHM/2.35$, P_{acoustic} is the acoustic power to be determined, Z is the wave impedance of 1.5 MRayl, dx times dy compose an area element dA in the focal plane and I_{SPTP} is the spatial peak, temporal peak, acoustic intensity derived from the total acoustic power over the FWHM focal plane area. For the laboratory HIFU system, P_{acoustic} is computed to 304 W and I_{SPTP} to 15.1 kW/cm².

FIG. 1. (Color online) Three-dimensional schematic depiction of the acoustic trench created for HIFU. (A) First a back wall bubble layer is created by means of Acoustic Droplet Vaporization. (B) Then a bubble spiral wall is created on top of it to form a trench. (C) Inside the bubble trench, the acoustic field of a HIFU beam is used to create thermal lesions. Discrete lesions are placed along the spiral track of the trench and result in a solid lesion of the shape of a thick disk of 15 mm height (D). Trench geometries depend on the HIFU point spread function and steering capabilities of the HIFU beam. Note: Approximate aspect ratios are shown. The beam widths of the clinical and laboratory HIFU systems are 1.0 and 1.6 mm, respectively, and the trench walls are spaced approximately 5 mm. A 3.25 mL lesion can be created in 55.4 s. Actual heating required 40 s while the creation of the acoustic trench required 15.4 s.

A programmable 3-axis positioning system was used to translate the focused transducer in a spiral pattern as illustrated in Fig. 1. A short tone burst of 10 cycles at 1.44 MHz with a 0.25% duty cycle was applied to the therapy transducer to vaporize the droplets in thermal phantom blocks (5.5 cm $\times$ 5.5 cm $\times$ 6.5 cm), forming a bubble back wall [10 mm in thickness, Fig. 1(A)] and then a spiral sidewall [3 mm in thickness, Fig. 1(B)]. The purpose of the back and sidewalls is to reflect and largely contain the fraction of the incoming and bubble-scattered HIFU beam, confining a greatly increased fraction of the heat generation within the trench.

The formation of the spiral shaped trench was achieved within 30 s. Subsequently, compound lesions were formed by 5 s exposures of the HIFU beam inside the trench [Fig. 1(C)] separated by 5 s delays. As shown here, ultrasound was focused at discrete locations along the trench thereby generating a solid thermal lesion disk [Fig. 1(D)]. Center-to-center spacing between neighboring lesions within the trench was 5.5 mm (right column in Fig. 2). The geometry of the acoustic trenches was verified prior to HIFU exposure, using a GE Logiq 9 ultrasound scanner (GE Ultrasound, Milwaukee, WI) with a 7 L array operating at 7 MHz.

C. Lesion size estimation using visible macroscopic imaging and MRI

All lesions were imaged at 2 T with T_2 -weighted magnetic resonance imaging (MRI) with 1 mm $\times$ 0.5 mm $\times$ 0.25 mm

resolution, and optical macroscopic observations in sliced phantoms. In MRI, the transverse magnetization relaxation time constant, T_2 , is sensitive to slow molecular motions of water. The denatured and cross-linked proteins caused a significant decrease in the water proton mobility and thereby a decrease in the water proton T_2 (Lambelet *et al.*, 1991), which was observed as a darker region in Fig. 2. Therefore, MRI provided an alternative, independent, and direct method of measuring protein coagulation, and it was utilized to assess the uniformity of compound thermal lesions. The spin-echo MRI imaging sequence with T_2 weighting was performed within 1 h after thermal lesion production in the phantoms.

The repetition time TR, the amount of time between successive pulse sequences applied to the same slice, was 4.5 s. The echo time TE, the time between the 90° pulse and the peak of the spin echo signal and inversion recovery pulse, was 120 ms. After MRI, the lesions were sliced and dissected based on the visible color change. Lesion volumes of the laboratory HIFU system were also estimated by a fluid displacement method.

D. Lesion formation using an MRI guided clinical HIFU system

A Philips Sonalleve MRI-HIFU system utilizing a 256-element phased array was used to generate HIFU lesions. For the generation of spiral shaped trenches a first generation Sonalleve V1 system with a 120 mm HIFU transducer focal length, implemented on a 1.5 T MR scanner, was employed. A newer system, a Sonalleve V2 (3 T MR scanner, 140 mm HIFU transducer focal length), was employed for the generation of single lesions. Both Sonalleve systems, while not identical, are capable of generating either lesion shape. At the employed transmit center frequency of 1.45 MHz, a maximum electronic steering of ± 10 mm was achieved at a depth of 10 cm for spiral pattern generation analogous to that of the single element transducer. Emulsions were vaporized by use of continuous wave excitation. For the creation of the bubble back wall and bubble spiral trench using the Sonalleve V1 system, an exposure of 50 W was employed for 100 ms. For the creation and characterization of single thermal lesions, the Sonalleve V2 system was employed using exposures ranging from 30 to 200 W for 20 s at 1.45 MHz center frequency. Calibrations under free field conditions and 1 W excitation in a water tank yielded a pressure amplitude of 1.12 MPa and a point spread function FWHM intensity of 1.00 mm lateral and 7.99 mm axial. Under the assumption of linear propagation, 200 W excitation in water would result in a pressure amplitude of 15.9 MPa. Assuming that 66% of the beam intersects the FWHM of the focus as a planar wave traveling in water with 1.5 MRayl impedance, this pressure corresponds to 12.7 kW/cm² (16.2 MPa).

Lesion volumes were measured by MRI and computed as all volumetric pixels with T_2 values deviating from background as described by Zhang *et al.* (2011). Lesion volume increases due to the presence of perfluorocarbon emulsions in the phantoms during HIFU were quantified as the ratios of

FIG. 2. (Color online) Optical (top row) and T_2 -weighted MR images (bottom two rows) of lesions (i.e., denatured protein) generated within thermal phantoms using the laboratory HIFU system. The left and right columns depict lesions generated with and without ADV-enhancement (see Fig. 1), respectively. The scale bar is 1 cm. The solid, double and dashed lines superimposed to the middle and bottom row MRI scans indicate where the lateral-elevational and the lateral-axial images intersect.

lesion volume with and without emulsions present. Lesion sizes were quantified as a function of applied acoustic power ranging from 30 to 200 Watts and *in situ* temperature monitoring with a therapeutic endpoint of 75°C (see Fig. 3). Statistically significant differences between lesions sizes (i.e., with and without ADV) were determined using a Student's *t*-test. A significance level of 0.05 was used for all comparisons.

In Fig. 3(A), real-time heating is assessed and then displayed via a pseudo color map whose scale is shown on the right colorbar. The temperature map is overlaid on the continuously updated magnitude image resulting from the gradient echo sequence that is used to measure temperature. Since

the thermal map scan sequence is not optimized for good contrast, but rather for fast acquisition and good signal to noise at the center, the image appears gray and is subject to some zebra artifacts (Stadler *et al.*, 2007) at the edge of image. In this sequence, ADV was performed at an output of 100 W. A temperature of 83°C was reached in part of the lesion. Conversely, Fig. 3(B) displays the real-time temperature mapping employing a static T_2 weighted spin echo image sequence acquired before the heating sequence. The T_2 weighted spin sequence is slow but provides better contrast without artifact or image distortion, which allows discrimination of the various layers in the phantom and its surroundings more clearly.

FIG. 3. (Color online) MRI-guided temperature readout during HIFU exposure. (A) A temperature map, scale bar on the right side, is overlaid on the magnitude image of the MRI gradient echo sequence. The gradient echo sequence is optimized for fast acquisition and signal-to-noise at the center. (B) Conversely, using a T_2 weighted spin sequence once, a static background image is generated and placed behind the continuously updated heating color map, therefore providing better contrast for distinguishing phantom details and other important structures. Note: The HIFU beam enters from the left into the phantom and the spatial extent of the beam is illustrated by the two shaded opposing triangles.

FIG. 4. (Color online) Optical images (side view) of linear compound lesions (white) in doped phantoms, generated with the laboratory system, with (left) and without (right) a bubble wall. The bubble back wall is visible below the lesion on the left. Note that the tadpole shape characteristic of bubble-enhanced HIFU lesions is not evident with the bubble wall. A thermal ablation volume of 15 mL was achieved in 4 min with ADV and the acoustic trench. The scale bar is 1 cm. The dashed arrowhead line shows propagation direction of the HIFU.

III. RESULTS AND DISCUSSION

A. Laboratory HIFU lesion generation

Compound lesions in a spiral pattern were formed in phantoms with and without droplets, in the following referred to as “doped phantom” and “control phantom,” respectively. Initial results, shown in Fig. 2, were similar for both optical macroscopic and T_2 -weighted MR images. In MR images, a high contrast between the mean gray levels inside the lesions and the surrounding background was observed, which showed a difference of at least nine standard deviations. The coefficients of variance were found to be similar in the lesion and in the background medium (i.e., the standard deviations did not exceed 5% of the mean value). Therefore, a simple amplitude thresholding was applied in each section to discriminate pixels included in the lesions from pixels of the background, with a threshold set to the mid-value between the mean gray level values in the lesion and in the background.

For lesions generated in a spiral pattern (Fig. 2), a 2.5-fold increase in the axial dimension of the lesion was obtained with the acoustic trench. Figure 4 shows optical images of linear compound lesions. Given the homogeneously complete filling of the treatment space, there is a considerably larger volume that is treated when using the ADV enhancement and acoustic trench approach. For instance, here a uniform thermal ablation volume of 15 mL was

achieved in 4 min with ADV and the acoustic trench. A common term in the HIFU literature is the time spent on a lesion of a given volume. Here 4 min for a 15 mL lesion yields 16 s/mL.

In both optical and MR images of the HIFU lesion in the presence of ADV bubbles, the back bubble wall was visible below the lesion. The tadpole shape characteristic of bubble-enhanced HIFU lesions was not evident with the bubble wall in place (Fig. 4). As seen in Fig. 4, the thermal lesion, indicated by the opaque denatured egg-white, ended abruptly and uniformly across the back wall. The layer of this exposure plane (i.e., proximal to the transducer) can then be the back wall for the next layer. It is expected that ADV occurs during lesion formation as both ADV and lesion formation were executed with the same pressure amplitudes though longer exposure times were required for lesion formation. The function of the acoustic trench sidewalls is to reflect the lateral and elevational beam components hereby resulting in a homogeneous lesion versus the tadpole shape.

B. Lesion formation results using a clinical HIFU system

Rapid repetition of electronically steered therapy pulses (40 pulses per second, 50 W) in the clinical HIFU system allowed for the generation of nearly homogeneous composite lesions at 17.0 s/mL or better (Fig. 5). The volume of compound lesion (3.24 mL) is 14-times larger than the sum total single volumes ($8 \times 28.4 \mu\text{L} = 227 \mu\text{L}$). The time to create a spiral compound lesion is 55 s (i.e., 17.0 s/mL), versus 40 s (i.e., 176 s/mL) to create eight individual lesions with no back wall and no spiral trench. Thus, a 1400% increase in lesion volume was achieved with only a 37% increase in treatment time.

Consistent with earlier results on the laboratory system (see also Zhang *et al.*, 2011), for acoustic power levels ranging from 30 to 200 W, lesion volumes of doped phantoms increased compared to lesion volumes in control phantoms (see Fig. 6). Lesion temperature was recorded in real-time and used as the therapy end-point while insonifying at the employed acoustic power. Lesion size was measured over a range of power levels and axial as well as lateral lesion dimensions. The minimum power level at which lesions

FIG. 5. B-mode images of single and compound lesions from the clinical HIFU system are shown in panels (A), (B), and (C), respectively. (A) Eight individual bubble clouds and HIFU from ADV produced with a 5 s per cloud sweep speed and illustrate the extent of single lesions. (B) A spiral shaped trench wall is created within 4 s by rapid succession of individual acoustic electronically steered pulses for ADV. Forty-four individual beam positions were targeted with 100 ms each to form the shown spiral. (C) In a second step, as schematically illustrated in Fig. 1, the formed acoustic trench was filled-in by eight additional individually positioned HIFU beams (50 W, 5 s each), which then created the solid lesion that is shown. A backside wall (not shown) was created before the spiral with 110 beams in 11 s. The total lesion (3.25 mL) creation time including bubble clouds was 55 s (see Fig. 1) or 17.0 s/mL.

FIG. 6. (Color online) (A) Lesion sizes were estimated using T_2 -weighted MR imaging. Doped phantoms (open circles) showed larger lesion volumes than control phantoms (stars). To reach an average lesion volume of 0.105 mL in the control phantom, the acoustic power must be more than doubled (213%). At 150 W acoustic power the lesion volume ratio of doped versus control exceeds 8-times. Vertical lines indicate the thresholds, below which lesions are not observed. (B) and (C) Axial and lateral lesion diameters were measured. Lesion volumes in doped phantoms showed a greater functional gradient with applied acoustic power compared to control phantoms. Data are shown as the mean $\pm$ standard deviation for $n=4$. “#” indicate $p < 0.05$ versus control phantoms.

were observed was 40 W (doped phantom), which corresponded to a lesion volume of $17 \pm 11 \mu\text{L}$. Figure 6 shows that at 150 W acoustic power, doped phantoms yielded eight-times larger lesions for the same acoustic power than control phantoms. To reach an average lesion volume of 0.105 mL in the control phantom, the acoustic power must be more than doubled (213%) compared to the power used in the doped phantom. Doped lesions with volumes statistically different from controls are marked with a “#” symbol in Fig. 6(A). Regression analyses [Figs. 6(B) and 6(C)] show that both the lateral and axial lesion lengths increase with $\sim 56.0\%$ greater slope when comparing doped to control phantoms. A constant offset of the fitting function over the tested acoustic power possibly indicates the larger depth of field compared to the lateral beam diameter.

As shown in Fig. 7, the lesion shapes within control and doped phantoms differed, with lesions within control phantoms showing higher symmetry along the axial direction. Doped phantoms exhibited lesions that progressed toward the sound source and were cone shaped, except for a tail distal to the triangle. At each axial position of a lesion the

lateral-elevational cross-sectional area A was measured. Then an effective radius was calculated as $r_{\text{eff}} = \sqrt{A/\pi}$. Line plots of r_{eff} are shown as a function of axial position to illustrate the three-dimensional character of the lesions.

The acceleration of HIFU procedures by use of administered or induced gas bodies has been investigated by numerous research groups. Peng *et al.* (2012) compared ultrasound-guided HIFU treatment for uterine fibroids to treatment enhanced by SonoVue, administered 10 min before HIFU as a bolus of 2.0 mL normal saline with 10.0 mg SonoVue via the hand vein. For fibroids smaller than 4 cm in diameter, average sonication times decreased from 69 s/mL (HIFU only) to 34 s/mL (HIFU + SonoVue), meanwhile the range decreased as well 2–619 s/mL versus 15–334 s/mL, but is not significantly different at $p=0.354$. For ADV accelerated HIFU, in comparison, single lesion formation was achieved in the clinical HIFU system in less than 20 s, resulting in a 0.67 mL volume, hence allowing for approximately 30 s/mL lesioning rates. For lesions based on spiral trenches, the rate improved to 17.0 s/mL.

During image-guided ultrasound therapy, the rate of massive gray scale changes during lesion formation was 50.0% (HIFU only) versus 77.8% (HIFU + SonoVue) (Peng *et al.*, 2012). Chung *et al.* (2012) reported that the volume of coagulative necrosis was significantly larger for contrast-assisted HIFU in comparison to HIFU alone. Here the volume increased from 0.83 mL to 1.75 mL ($p < 0.05$). Simultaneously, tissue ablation times significantly decreased from 12.1 s to 8.6 s ($p < 0.05$). Illing *et al.* (2005) stated that observed adverse events were localized to the area of treatment. Following HIFU treatment, skin toxicity occurred in 27% of the cases (8/30). These were not clinically relevant (except for one case) and resolved spontaneously. Ablation at large depth (e.g., kidney tumors) may be more difficult and less reliable than at more superficial depths (e.g., liver tumors). Illing *et al.* speculated that this decreased reliability “may be due to the greater depth of renal tumor[s] and the presence of a perinephric fat layer, both of which attenuate the ultrasonic beam.”

To treat rapidly enough for efficient use of expensive facilities and staff as well as reasonable duration of anesthesia, it is essential to enhance heating without damaging the skin and other overlying tissues. For example, a single uterine fibroid with a volume of 17 mL requires approximately 20 min of treatment time using current standard HIFU. Using ADV enhanced trenches, this time would decrease to 5 min. The treatment time for a 6 cm diameter hepatocellular adenoma, which currently requires 2 h, would be reduced to 30 min.

ADV shows the potential to provide local control of heat deposition while minimizing tissue damage caused by microbubbles in the path of therapeutic ultrasound beams, as is the case in use of UCAs. In this study, an appropriate bubble density was achieved consistent with therapeutic ultrasound while providing uniform ablation. The required droplet concentration was comparable to diagnostic UCA use. While the emulsion core material, dodecafluoropentane (C_5F_{12}), is an inert fluid with high biocompatibility, *in vivo* administration of large quantities may cause bioeffects (Zhang *et al.*, 2011). In this paper, 10^5 droplets/mL were used to enhance HIFU

FIG. 7. (Color online) (Left) MRI images showing coronal cross-sections of generated HIFU lesions at 200 W acoustic power and 20 s exposure for doped and control phantoms. The propagation direction of the HIFU was from left to right. Achieved lesion volumes and volumetric creation speeds were 0.654 mL and 30.6 s/mL as well as 0.185 mL and 108 s/mL for doped (top) and control (bottom) phantoms, respectively. (Right) For each lesion the lateral-elevational cross-section was fit to a circle. The radius of the fitted circle is plotted as a function of axial position through the lesion. Lesion geometry asymmetries between doped and control phantoms can be seen.

lesions. In a highly perfused organ such as liver, with an average fractional blood volume of 31% (Schwickert *et al.*, 2005), 3.23×10^5 droplets/mL of blood are required for HIFU lesions in a 70 kg human with 5 L blood volume. Systemically, this yields 2.3×10^7 droplets/kg, which is 282-times lower in concentration than a recent biodistribution study where a similar emulsion was safely tolerated at 6.5×10^9 droplets/kg (Fabiilli *et al.*, 2013).

Our phantom experiments demonstrated no heating effects such as protein denaturation in the pre-focal or post-focal ultrasound propagation pathway (see Fig. 3). In addition, use of an ADV-bubble back wall limits distal ultrasound propagation, thus limiting damage beyond the wall. It is possible to create these distal barriers prior to treatment as an additional layer of protection for sensitive tissues such as nerves in clinical implementations. It is therefore hypothesized that, building upon previously produced lesions with ADV bubbles should allow for the reduction of the transmitted acoustic energy, which can be advantageous in sensitive tissue regions. These bubble walls can be created using low duty cycle, microsecond pulses that will not generate thermal lesions. Further investigations of the requisite bubble wall characteristics and the level of protection afforded by the wall are warranted as are studies of the optimal location of the ultrasound beam focus for heating in front of the barrier wall. Our results also indicated that for the treatment of a large volume, the ADV-enhanced HIFU would require fewer targeting sites to create a compound lesion throughout a predetermined volume, because the size of the individual lesions was increased. Lesion size diminishes with decreasing acoustic power, as seen when treating tissues at large depth. Compensatory increase in acoustic power is limited due to its consequential damage of overlying tissues. However, an increase in the maximum depth of treatment is achievable by means of ADV enhanced HIFU as the required acoustic power is on average lowered by 47% for the same depth lesion of 0.1 mL volume, when compared to non-ADV HIFU (Fig. 6). Therefore, currently untreatable regions due to depth or restricted aperture should be realizable by the use of ADV.

Another parameter warranting further study is the optimum size for the emulsion. In this paper, a micron-sized emulsion, similar in size to UCA, was used. A previous *in vitro* study (Zhang *et al.*, 2013) utilized a 50-fold higher concentration of submicron-sized emulsion, which could ultimately accumulate within a tumor via extravasation through leaky vasculature. This type of passive targeting could further enhance localized heating effects, though the relationship between emulsion size and the concentration required for enhanced heating is not fully elucidated.

IV. SUMMARY AND CONCLUSION

ADV greatly enhanced HIFU therapy by increasing the efficiency of focal energy deposition. Large, compound lesions can be generated at a rate of 17 s/mL of lesion volume. Uniform stacking of spiral lesions occurred without the distal gaps characteristic of lesions using bubble enhancement, which allows for efficient stacking of compound lesions. The thermal lesion had a sharp back wall margin, allowing precise protection of tissue structures distal to the targeted region. Perfluorocarbon emulsions demonstrate great potential for enhancing clinically relevant HIFU performance, with four-times faster treatment times as well as improved lesion homogeneity. This initial phantom study serves as an appropriate test of our methods and also provides preliminary indications for *in vivo* animal study and clinical trials. Efficient lesion formation with real-time imaging feedback at droplet concentrations less than 1% of tested safety limits supports feasibility for high biocompatibility of this mechanism to accelerate thermal therapy. This technology may be applicable across a broad range of HIFU applications.

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