Published in final edited form as:

Ultrasound Med Biol. 2018 January ; 44(1): 199–213. doi:10.1016/j.ultrasmedbio.2017.09.007.

Real-time spatiotemporal control of high-intensity focused ultrasound thermal ablation using echo decorrelation imaging in *ex vivo* bovine liver

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Abstract

The ability to control high-intensity focused ultrasound (HIFU) thermal ablation using echo decorrelation imaging feedback was evaluated in *ex vivo* bovine liver. Sonications were automatically ceased when the minimum cumulative echo decorrelation within the region of interest exceeded an ablation control threshold, determined from preliminary experiments as -2.7 ($\log_{10}$ -scaled decorrelation per millisecond), corresponding to 90% specificity for local ablation prediction. Controlled HIFU thermal ablation experiments were compared with uncontrolled experiments employing 2, 5, or 9 sonication cycles. Means and standard errors of the lesion width, area and depth, as well as receiver operating characteristic curves testing ablation prediction performance, were computed for each group. Controlled trials showed significantly smaller average lesion area, width, and treatment time than 5-cycle or 9-cycle uncontrolled trials, and also showed significantly greater prediction capability than 2-cycle uncontrolled trials. These results suggest echo decorrelation imaging is an effective approach to real-time HIFU ablation control.

Keywords

Echo decorrelation imaging; high-intensity focused ultrasound; thermal ablation; real-time control

Introduction

Local thermal ablative techniques have been shown an effective treatment for nonresectable hepatocellular carcinoma and colorectal metastases in liver (van Sonnenberg et al., 2005), offering potential tumor control and in some cases long-term, disease-free survival. However, it remains difficult to use these techniques to treat tumors located near major

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blood vessels while avoiding blood vessel injury (Ng et al., 2004). High-intensity focused ultrasound (HIFU) can cause coagulative necrosis at a precise focal point within the tissue without harming overlying and adjacent structures, even for those within the beam path (Zhang et al., 2009), providing sufficient residual liver tissue for patient survival (Aubry et al., 2013). HIFU has been employed to treat unresectable liver cancer in thousands of cases in China (Luo et al., 2015).

Imaging modalities are needed for real-time monitoring of tissue changes during HIFU thermal ablation. Magnetic resonance-guided HIFU (MRg-HIFU) and ultrasound-guided HIFU (USg-HIFU) are the most commonly used platforms for HIFU treatment guidance and control. MRg-HIFU has been used for monitoring and control of thermal ablation using real-time MR thermometry (Hynynen et al., 1993; Darkazanli et al., 1993; Bohris et al., 1999; Hynynen and McDannold, 2004; Bour et al., 2017). However, MR imaging increases treatment complexity and cost (Zhou, 2011; Napoli et al., 2013). USg-HIFU is more portable and inexpensive, while its high frame rate and real-time capability potentially make it more capable of ablating moving organs (e.g. liver (Wu et al., 2004; Kennedy et al., 2004) and kidney (Illing et al., 2005; Klingler et al., 2008)).

Various monitoring algorithms have been developed to detect HIFU-induced thermal lesioning based on ultrasound pulse-echo imaging, including B-mode (Vaezy et al., 2001; Gudur et al., 2012), M-mode (Kumon et al., 2012), backscatter (Seip et al., 2002; Chen et al., 2011; Shishitani et al., 2013), echo strain imaging (Souchon et al., 2005), harmonic motion imaging (HMI) (Han et al., 2015; Grondin et al., 2015; Yang et al., 2017), elastography (Iwasaki et al., 2016), and acoustic radiation force imaging (Lizzi et al., 2003). All these methods aim to provide feedback on the progress of thermal treatment and to confirm the completeness of ablation. However, these methods are sensitive to errors caused by the formation of microbubble clouds at the ablation site (Varghese et al., 2004), the complex nonlinear dependence of tissue acoustic and viscoelastic properties on temperature (Suomi et al., 2016) and decorrelation of the backscattered signal caused by tissue motion (Curiel et al., 2009), bubble activity (Kumon et al., 2012; Casper et al., 2013), and heat-induced changes in tissue structure (Majaron et al., 1999; Kruskal et al., 2001; Hooi et al., 2015).

Echo decorrelation imaging (Mast et al., 2008; Matsuzawa et al., 2012; Subramanian et al., 2014; Sasaki et al., 2014; Fosnight et al., 2017) potentially overcomes these errors by utilizing heat-induced, millisecond-scale echo variations to spatially map thermal lesioning during HIFU thermal ablation. Echo decorrelation imaging has been successfully validated for prediction of *in vivo* HIFU thermal lesioning in normal and VX2 rabbit tumor liver tissue (Fosnight et al., 2017, 2014).

Real-time feedback control algorithms for HIFU therapy based on pulse-echo ultrasound imaging have been tested in previous research (Seip et al., 2002; Curiel et al., 2009; Sugiyama et al., 2015; Takagi et al., 2016), to reduce treatment time and to enhance ablation accuracy (Al-Bataineh et al., 2012). Curiel et al. (2009) evaluated the feasibility of using localized HMI to monitor and control HIFU ablation in VX2 rabbit tumors *in vivo*. Treatments were automatically stopped if the localized harmonic motion amplitude at the

focus dropped below a specified amplitude threshold in two consecutive measurements. This algorithm was able to successfully control ablation in 69% of the trials; echo decorrelation associated with tissue state changes and motion was considered a source of error. Similarly, Sugiyama et al. (2015) used real-time localized motion imaging (LMI) as a feedback method to control HIFU thermal lesion length in porcine liver. Their feedback control system successfully provided lesion lengths close to their target (8–12 mm) with root-mean square error (RMSE) of 2.51 mm. However, errors were found in the estimated lesions' size and overtreatment occurred due to system delay after ceasing the HIFU sonication. Qu et al. (2016) improved the LMI method by employing a dynamic cross-correlation window, which decreased the RMSE of the estimated coagulation length to 1.69 mm during porcine liver ablation ($N=49$), but increased processing time for displacement estimation from 2.05 ± 0.05 s to 2.64 ± 0.36 s for 28 A-mode scan lines.

Recently, Takagi et al. (2016) developed an approach for real-time monitoring and control of HIFU treatment based on cross-correlation of ultrasound signals, similar to echo decorrelation imaging. The cross-correlation coefficient between echo signals, calculated between the target frame and the first (reference) frame was used to detect thermal coagulation during HIFU treatments in chicken breast. HIFU exposures were automatically terminated when the average cross-correlation coefficient within a focal region of interest (ROI) fell below a predetermined threshold. Feedback-controlled, HIFU treated samples were found to be more homogeneously coagulated compared to uncontrolled cases. However, they noted that further investigation is needed to determine the optimum coagulation threshold and to increase the monitoring frame rate for better detection of tissue changes in the presence of motion, boiling and cavitation. In addition, their approach of cross-correlating with a fixed, pre-treatment reference frame may not be amenable to control of *in vivo* HIFU ablation, where motion-induced decorrelation can be large (Subramanian et al., 2014; Fosnight et al., 2017).

In the research reported here, the feasibility of controlling HIFU thermal ablation using 2D echo decorrelation imaging was assessed with a view toward *in vivo* implementation. The echo decorrelation imaging approach employed here tracks changes in echo signals over millisecond-scale intervals, potentially providing more robust control than cross-correlation with a fixed reference frame. The main goals of this research were to (i) implement an automated control algorithm based on real-time echo decorrelation imaging, (ii) determine the optimum echo decorrelation threshold for HIFU control, (iii) test the algorithm in *ex vivo* bovine liver experiments, and (iv) statistically assess the effect of control on thermal lesioning results.

Materials and Methods

In this section, materials and methods for a series of experiments on controlled ablation of *ex vivo* liver tissue are discussed. In these experiments, both HIFU ablation and echo decorrelation imaging were performed using a linear, 5 MHz image-treat array, with treatments ceased after echo decorrelation exceeded a predetermined threshold throughout the focal region of interest. An overview of the experimental setup is shown in Fig. 1 (a)–(b).

Echo decorrelation feedback control algorithm

In order to control HIFU ablation using echo decorrelation imaging, a real-time, closed-loop control algorithm was developed. This algorithm was integrated with a real-time imaging application used to compute B-mode and echo decorrelation images from beamformed radiofrequency (RF) echo signals acquired immediately after each sonication pulse, during the rest period of the HIFU transducer, as shown in Fig. 1(d).

A flow chart of the control algorithm is shown in Fig. 1(c). Echo decorrelation images, comprising maps of the local echo decorrelation per millisecond, were computed as (Subramanian et al., 2014)

$$\Delta(y, z, t) = 2 \left[\frac{\beta^2(y, z, t) - |R_{01}(y, z, t)|^2}{\tau [\beta^2(y, z, t) + \overline{\beta^2}(t)]} \right], \quad (1)$$

where y and z are azimuthal (array) and range (depth) image coordinates and τ is the inter-frame time in milliseconds. R_{01} is the position-dependent, zero-lag, windowed spatial cross-correlation between two consecutive complex image frames, while $\beta^2(y, z, t)$ is an integrated backscatter term determined from the corresponding autocorrelations of the two frames:

$$\begin{aligned} R_{01}(y, z, t) &= \langle I(y, z, t) * I(y, z, t + \tau) \rangle, \\ \beta^2(y, z, t) &= \langle |I(y, z, t)|^2 \rangle \langle |I(y, z, t + \tau)|^2 \rangle, \end{aligned} \quad (2)$$

where $\langle \cdot \rangle$ represents two-dimensional convolution with a Gaussian window (Subramanian et al., 2014) and $\overline{\beta^2}$ denotes the spatial mean average of $\beta^2(y, z, t)$. The echo decorrelation term in Eq. (1) is normalized by the inter-frame time τ to define the echo decorrelation map per millisecond (Fosnight et al., 2017) and by the average of $\beta^2(y, z, t)$ and $\overline{\beta^2}$ to reduce decorrelation artifacts in hypoechoic and hyperechoic regions respectively (Mast et al., 2008). More detailed derivation and analysis of echo decorrelation imaging is given elsewhere (Mast et al., 2008; Subramanian et al., 2014; Fosnight et al., 2014, 2017; Hooi et al., 2015).

Echo decorrelation maps were computed from image frames recorded immediately after each HIFU sonication cycle. For each cycle, an ensemble-averaged echo decorrelation map $\overline{\Delta}(y, z, m)$ was computed as

$$\overline{\Delta}(y, z, m) = \frac{1}{K-1} \sum_{k=1}^{K-1} \Delta(y, z, k\tau), \quad (3)$$

where m is the therapy cycle index, K is the number of image frames recorded per cycle, and k is the frame index.

Cumulative echo decorrelation maps $(y, z, m)_{cum}$ were defined as the accumulated temporal maximum of the ensemble-averaged echo decorrelation maps for each pixel (y, z) :

$$\Delta(y, z, m)_{cum} = \max(\bar{\Delta}(y, z, m-1), \bar{\Delta}(y, z, m)). \quad (4)$$

The cumulative echo decorrelation $(y, z, m)_{cum}$ was used as a feedback predictor for closed-loop ablation control, qualitatively representing the time history of thermal energy deposition in the tissue. The imaging application overlaid the cumulative echo decorrelation map over the B-mode image at the end of each sonication cycle, resulting in a frame rate of 0.344 Hz (inverse of the 2.9 s cycle length). Treatments were automatically terminated when either the minimum cumulative echo decorrelation within a focal ROI exceeded the predetermined ablation control threshold or when the current sonication cycle index m exceeded the maximum number of sonication cycles M .

HIFU ablation experiments

Tissue handling followed procedures employed in previous studies of echo decorrelation imaging during *ex vivo* thermal ablation (Mast et al., 2008; Subramanian et al., 2016). A freshly excised bovine liver was obtained from a local slaughterhouse and placed in ice-cold, deionized, degassed phosphate-buffered saline (PBS). Liver tissue specimens were cut to fit a square cuboid acrylic cuvette with dimensions $85 \times 85 \times 60 \text{ mm}^3$ with an open top for ultrasound imaging and treatment, as shown in Fig. 1a. The tissue surface was covered with a layer of aqueous gel to couple the specimen and transducer while avoiding tissue dehydration. Experiments were performed at room temperature, with an average liver temperature of 21°C measured before each trial.

HIFU treatments and imaging were controlled by the Iris 2 ultrasound imaging and therapy system (Ardent Sound Mesa, AZ, USA) (Barthe et al., 2004). A custom image-treat linear array transducer (64 elements, $4.8 \times 24.4 \text{ mm}^2$ aperture, frequency 5–5.4 MHz) performed both pulse-echo imaging ($> 40\%$ fractional bandwidth, transmit focal depth 3.5 cm, F -number 4) and HIFU therapy (maximum acoustic power 35 W). This array was integrated with a 23 mm water-filled standoff, filled with deionized, degassed water. The Iris 2 system was interfaced with a secondary computer that acquired beamformed RF echo signals using a 14-bit A/D converter (Compuscope 14200, Gage Applied) and initiated HIFU sonications via a custom programmable serial interface.

A graphical user interface (GUI), shown in Fig. 2, facilitated treatment planning, monitoring, and control for HIFU therapy. Before each ablation experiment, the user defined the maximum number of sonication cycles M , the ROI size/location, and the ablation control threshold thresh by parameters input to this GUI.

For HIFU treatments, a series of electronically focused exposures were fired in cycles with 0.7 s sonication followed by pulse-echo imaging and RF data acquisition during a 2.2–2.8 s rest period. The timing sequence of the HIFU exposures and echo data acquisition is shown in Fig. 1(d). Immediately after each HIFU exposure, twenty beamformed echo frames were

acquired at a frame rate of 116 Hz. Echo data were sampled at 33.3 MHz and stored to a solid-state flash drive.

Recorded echo signals were filtered by a Hilbert-transform filter providing in-phase and quadrature (IQ) complex signal components, then demodulated using a 5 MHz carrier frequency and decimated by a factor of 6. Demodulated IQ frames were used to compute B-mode images, maps of the local echo decorrelation per millisecond, and corresponding ensemble-averaged and cumulative echo decorrelation maps for each cycle (Fosnight et al., 2017) in real time. In the GUI, instantaneous and cumulative echo decorrelation maps were superimposed on B-mode images for each frame for real-time feedback, as shown in Fig. 2.

To directly relate echo decorrelation images to the desired outcome of tissue ablation, images were compared with tissue histology, as done in many previous studies of ultrasound monitoring approaches (e.g., Zhou et al. (2010); Wijlemans et al. (2015); Bour et al. (2017); Yang et al. (2017)). After treatments, ablated liver tissue was frozen overnight at -80°C in the same cuvette, maintaining tissue shape and orientation for accurate registration with ultrasound images. The tissue was then sectioned along the image plane and stained with 2% triphenyl tetrazolium chloride (TTC) vital stain. Ablated tissue histology was assessed based on TTC uptake, with regions of full TTC uptake (stained red) interpreted as non-ablated liver tissue and regions of partial or no TTC uptake interpreted as ablated tissue (Scheffer et al., 2014). TTC-stained sections were optically scanned at 1200 dpi (CanoScan 8800F, Canon, Japan). Scanned histologic images were then manually segmented into ablated and non-ablated regions (Fosnight et al., 2017).

Image registration of segmented tissue sections with corresponding B-mode images was performed using 2D rigid registration with two translational and one rotational degree of freedom (Fosnight et al., 2017). Landmark points placed at the left, right and top edges of the HIFU lesion were used to center the histologic section at zero azimuth of the B-mode image. At the same time, the histologic section was rotated and translated to align with the segmented tissue surface on the B-mode image. Registration errors were estimated based on the vertical (range/depth direction) distance between the proximal tissue surfaces segmented on the B-mode and histologic images. Mean and root-mean-square (RMS) values of this distance, as well as the 95% confidence interval of the mean, were determined over all proximal boundary points from all controlled and uncontrolled trials.

The capability of echo decorrelation imaging to predict local tissue ablation was assessed using receiver operating characteristic (ROC) curve analysis, as performed previously (Subramanian et al., 2014; Fosnight et al., 2017). For ROC curve analysis, cumulative echo decorrelation maps were computed using MATLAB (The MathWorks, Natick, MA, USA), including correction for decorrelation associated with electronic noise and tissue motion (Hooi et al., 2015; Fosnight et al., 2017). ROC curves were computed by comparing thresholded cumulative echo decorrelation maps to histologic binary masks for each trial (Subramanian et al., 2014; Fosnight et al., 2017), plotting the true-positive ratio [Sensitivity=TP/(TP+FN)] versus the false-positive ratio [1-Specificity=FP/(TN+FP)] over all image pixels and the entire range of echo decorrelation cutoffs (Hanley and McNeil, 1982). In these expressions, TP (true positives) is the number of correctly predicted ablated

points, FP (false positives) is the number of incorrectly predicted unablated points, TN (true negatives) is the number of correctly predicted unablated points, and FN (false negatives) is the number of incorrectly predicted ablated points. Area under the ROC curve (AUC) was computed using the trapezoidal rule; an AUC value of 1 represents the best possible prediction performance while a value of 0.5 indicates no predictive capability (Hanley and McNeil, 1982).

AUC values were tested for statistical significance against the null hypothesis ($AUC = 0.5$) using a general model for the AUC standard error (Hanley and McNeil, 1982) (one-tailed, significance criterion $p < 0.05$). The statistical significance of differences between AUC values was tested using DeLong's method (DeLong et al., 1988; Krzanowski and Hand, 2009) (significance criterion $p < 0.05$, two-tailed). To account for non-independence of predictions for closely located pixels, statistical tests of AUC were adjusted using effective sample sizes determined from the maximum packing density (hexagonal packing) of circular windows with diameter matching the spatial resolution of echo decorrelation images ($d = 2.35$ mm for a Gaussian correlation window width $\sigma = 1.0$ mm) (Subramanian et al., 2014; Fosnight et al., 2017).

Determination of ablation control threshold

The echo decorrelation threshold employed for controlled HIFU exposures was determined using a process outlined by Fosnight et al. (2017) in an *in vivo* study of echo decorrelation imaging during ultrasound ablation. For this purpose, a set of preliminary HIFU ablation experiments was performed without use of the control algorithm. HIFU thermal ablation trials ($N = 13$) were carried out on fresh *ex vivo* bovine liver using the experimental setup and methods described above.

In each of the ablation trials, nine HIFU sonication cycles (20% duty cycle, 5.4 MHz frequency, as in the *in vivo* study reported by Fosnight et al. (2017)) were performed using continuous-wave, high-intensity pulses, electronically focused along the array axis 2 mm below the tissue surface with 803–1125 W/cm² *in situ* spatial-peak, temporal-peak intensity (I_{SPTP}). I_{SPTP} was calculated based on measured acoustic power and simulated acoustic fields (Mast et al., 2005) as described previously (Fosnight et al., 2017). For each cycle, the array was fired for 0.7 s (HIFU on) followed by a 2.8 s rest period during which pulse-echo images were acquired. Before the therapy cycles of each trial, nine sham sonications were employed to estimate decorrelation caused by electronic noise and tissue motion; this estimated artifactual decorrelation was then used to compensate decorrelation maps during the HIFU treatment (Fosnight et al., 2017; Hooi et al., 2015).

Using simulations, temperatures in the focal plane were predicted for *ex vivo* liver tissue using previously reported methods and parameters (Mast et al., 2005), assuming 0.5 dB/cm/MHz absorption, 21 °C initial temperature, and no perfusion. According to these simulations, one sonication cycle was sufficient to achieve ablative temperatures in all cases. For the lowest HIFU exposure intensity employed (5.4 MHz, 20% duty cycle, 803 W/cm² I_{SPTP}), the maximum estimated tissue temperature after one sonication cycle was 74 °C, while maximum tissue temperature reached the boiling point (100 °C) after two sonication cycles.

Treated tissue was sectioned, stained, and segmented and ROC curves were computed as described above. The sensitivity and specificity of pixel-by-pixel ablation prediction were determined as a function of the echo decorrelation threshold, and the control threshold was chosen as that producing 90% prediction specificity.

Controlled HIFU experiments

To test the performance of echo decorrelation imaging feedback for controlling HIFU thermal ablation, a series of experiments was carried out over a range of sonication intensities and exposure durations. The real-time closed loop algorithm described above was implemented to control the array therapy module (ATM) of the Iris 2 system using the Qt-Serial library. Code specifying each HIFU exposure, including amplitude, phasing, and timing, was sent to a custom serial interface module by a standard serial communication protocol (RS232, 115200 bps, 8N1) and stored in the buffer memory of the serial interface module before being sent to the ATM for execution. Sonications were repeated by sending a serial command to the serial interface module to execute currently stored therapy code.

A small ROI of $1 \times 1 \text{ mm}^2$ was selected and located 2 mm below the tissue surface within the focal zone, as shown in Fig. 2. The control ROI was chosen to be smaller than the spatial resolution of the echo decorrelation map, estimated as 2.35 mm for the Gaussian window employed here (Subramanian et al., 2014). This ensured that decorrelation values computed within this ROI characterized echo changes at the focal position. A larger ROI would increase the likelihood of false-positive ablation predictions due to any artifacts. To examine the effect of ROI size on the control algorithm, a *post hoc* analysis was performed on the cumulative echo decorrelation maps from all controlled trials. Means and standard errors of the minimum echo decorrelation inside square ROIs of different diameters (1–10 mm) were computed. To compare with approaches employing average decorrelation within an ROI (Takagi et al., 2016), means and standard errors of the average decorrelation within the same ROIs were also computed.

HIFU exposures (5.0 MHz, 24% duty cycle) were performed with I_{SPTP} *in situ* ranging from 1064–1385 W/cm², with each therapy cycle comprising 0.7 s HIFU sonication followed by a 2.2 s rest period including pulse-echo imaging. The rest period corresponded to the time elapsed during acquisition of RF echo signals (0.5 s), transfer of echo data (0.4 s), computation of echo decorrelation and B-mode images (1.0 s), and firing of the next therapy cycle (0.3 s). Simulations showed a small increase in overall heating rate, compared to the 2.8 s rest period used in the preliminary experiments. After each cycle, echo decorrelation was computed; treatment was then continued if decorrelation failed to exceed its predetermined control threshold throughout the ROI, up to a maximum of $M = 18$ cycles. After each cycle, echo decorrelation was computed; treatment was then continued if decorrelation failed to exceed its predetermined control threshold throughout the ROI, up to a maximum of $M = 18$ cycles (twice the maximum number of cycles from a previous *in vivo* study of echo decorrelation imaging (Fosnight et al., 2017)). This procedure was followed for a total of 10 controlled ablation trials.

For comparison, series of uncontrolled HIFU ablation trials (5.0 MHz, 24% duty cycle) were also performed. The maximum number of uncontrolled therapy cycles was selected to be 9,

corresponding to the number previously found sufficient to ensure *in vivo* ablation of both rabbit liver and VX2 tumor (Fosnight et al., 2017, 2014). A total of 10 uncontrolled trials were performed using this maximum duration (total time 26.1 s for 9 cycles). An additional 10 uncontrolled trials were performed employing 5 therapy cycles (14.5 s), corresponding to the average sonication time required in the preliminary HIFU trials ($N=13$) for the cumulative echo decorrelation to exceed the selected ablation control threshold throughout the ROI. Based on results of initial controlled trials, the sonication time required to reach the predetermined decorrelation threshold was found to vary from 1–3 cycles (2.9–8.7 s), with an average of 2 cycles (5.8 s). Hence, a third uncontrolled group ($N=10$) was added to represent the average sonication time of the controlled trials, equal to 5.8 s (2 cycles).

After tissue sectioning, staining, registration, and segmentation as described above, thermal lesions for each trial were characterized by their width, area, and depth, computed in MATLAB based on binary maps of segmented tissue ablation. The lesion depth was measured as the vertical distance from the tissue surface to the deepest ablated point along the array axis (zero azimuth on the ultrasound images). The lesion width was measured as the azimuthal extent of ablated tissue along the horizontal (azimuthal) line taken at half the measured lesion depth. The lesion area was computed as the total area occupied by pixels mapped as ablated tissue.

Statistical analysis was done using R software (R Foundation, Vienna, Austria). Means and standard errors of the lesion width, depth, and area were computed for all groups. The normality of the width, depth and area for all groups was tested using the Shapiro-Wilk test with the significance criterion $p < 0.05$. For normally distributed groups, analysis of variance (ANOVA) was applied to test the hypothesis of no difference in means (significance criterion $p < 0.05$). If significant, a *post hoc* Tukey's honest significance difference (HSD) test was carried out for multiple comparison of means. For non-normally distributed groups, a Kruskal-Wallis rank sum test (Kruskal and Wallis, 1952) (significance criterion $p < 0.05$) was conducted to test the hypothesis of no median difference, followed by Dunn's test (Dunn, 1964) for multiple pairwise comparisons with Bonferroni adjustment (Holland and Copenhagen, 1988). Equality of variances was tested statistically using the Bartlett test for the normally distributed groups and the Levene test for the non-normally distributed groups. The average treatment time of the controlled group was tested against the fixed treatment times of 5-cycle and 9-cycle uncontrolled groups using one-sample t tests (two-tailed, significance criterion $p < 0.025$ including Bonferroni correction).

To test the performance of echo decorrelation imaging as an ablation predictor in the controlled and uncontrolled trials, ROC curves and AUC values were computed for each group by the same method described above. The significance difference of AUC values against the null hypothesis (AUC=0.5) and against each other (controlled vs uncontrolled groups) were tested as described in the previous subsection.

Results

Based on results of the preliminary experiments designed to determine the ablation control threshold, an ROC curve was computed to test the performance of echo decorrelation

imaging as a predictor of local tissue ablation. Echo decorrelation imaging predicted local ablation significantly better than chance ($AUC = 0.941$, $p < 10^{-16}$). The sensitivity and specificity of local ablation prediction using echo decorrelation imaging are plotted as a function of the echo decorrelation threshold in Fig. 3. The echo decorrelation threshold for the controlled ablation experiments was selected as $\text{thresh} = -2.7$ ($\log_{10}$ -scaled decorrelation per ms) to achieve the prescribed specificity of 90% in the preliminary experiments. The corresponding sensitivity of ablation prediction at this threshold was 83%.

In the 10 controlled HIFU trials, successful control was achieved in all 10 cases. That is, in all 10 cases, the echo decorrelation threshold of -2.7 ($\log_{10}$ -scaled decorrelation per ms) was reached throughout the focal ROI. The number of cycles required to reach this threshold ranged from 1 cycle (0.7 s sonication time or 2.9 s total therapy time) to 3 cycles (2.1 s sonication time or 8.7 s total therapy time). The mean number of cycles required for successful control was 2.0 (1.4 s sonication time or 5.8 s total therapy time), equal to the duration for the 2-cycle group (5.8 s therapy time) but significantly smaller than the 5-cycle group (14.5 s, $p = 1.8 \times 10^{-7}$) or the 9-cycle group (26.1 s, $p = 1.0 \times 10^{-10}$).

Means and standard errors of minimum (min) and average (avg) cumulative echo decorrelation values within ROIs of diameter 1–10 mm from all controlled trials are shown in Fig. 4. The mean of min exceeded the selected ablation threshold thresh in the $1 \times 1 \text{ mm}^2$ ROI (mean $\pm$ standard error -2.23 ± 0.13) only. However, the mean of avg exceeded thresh in ROIs of size $1 \times 1 \text{ mm}^2$ (-2.02 ± 0.098) to $5 \times 5 \text{ mm}^2$ (-2.6 ± 0.116).

Representative ablation results from the controlled HIFU ablation trials, as well as from uncontrolled comparison trials, are shown in Fig. 5. For each of eight trials, the cumulative corrected echo decorrelation map is shown superimposed on a B-mode image from the end of the trial, together with a co-registered map of the vitally stained, segmented tissue histology. Column (a) and (b) show results from trials ablated with average and maximum acoustic powers 26.6 W (*in situ* intensity $I_{\text{SPTP}} = 1192 \text{ W/cm}^2$) and 31 W ($I_{\text{SPTP}} = 1385 \text{ W/cm}^2$) respectively. Row (I) shows two representative controlled trials, while rows (II)–(IV) show representative results from uncontrolled trials with 2, 5 and 9 sonication cycles respectively. In each echo decorrelation image, the $\log_{10}$ -scaled ablation control threshold is indicated by a yellow line, while white lines indicate tissue boundaries segmented from the B-mode images. In the TTC-stained tissue sections, red and green lines indicate segmented tissue boundaries and ablated regions respectively. The RMS image registration error measured over all controlled and uncontrolled trials was 0.67 mm, while the mean registration error was 0.52 mm with a 95% confidence interval of 0.47–0.57 mm. Lesion dimensions (width, depth, and area) of the representative trials are close to the reported means for each group, as shown in Fig. 6(b)–(d).

ROC curves computed for the four experimental groups are shown in Fig. 6a. All AUC values were significantly greater than chance, including the controlled group ($AUC = 0.956$, $p < 10^{-16}$) as well as the 5-cycle ($AUC = 0.977$, $p < 10^{-16}$), 9-cycle ($AUC = 0.981$, $p < 10^{-16}$), and 2-cycle ($AUC = 0.722$, $p = 0.0243$) uncontrolled groups. No significant differences occurred between AUC values for the controlled, 9-cycle, and 5-cycle groups ($p > 0.3$). However, the 2-cycle uncontrolled group had a significantly smaller AUC value,

corresponding to poorer predictive capability, compared to the controlled ($p = 0.0179$), 5-cycle ($p = 6.8 \times 10^{-3}$), and 9-cycle ($p = 5.3 \times 10^{-3}$) groups.

Statistics of segmented lesion dimensions for controlled and uncontrolled trials are shown in Fig. 6(b)–(d). For lesion width [Fig. 6(b)], means $\pm$ standard errors were 1.29 ± 0.10 mm for the controlled group, 1.89 ± 0.09 mm for the 5-cycle group, 2.40 ± 0.11 mm for the 9-cycle group, and 1.01 ± 0.08 mm for the 2-cycle group. The lesion width passed the Shapiro-Wilk normality test ($p > 0.05$). The null hypothesis of the ANOVA test was rejected ($p = 1.3 \times 10^{-11}$), indicating a significant difference among lesion widths for the four groups. Pairwise comparisons of mean lesion widths from the Tukey HSD test are shown in Table 1. The controlled group had significantly smaller mean lesion width than the 5-cycle or 9-cycle uncontrolled groups, but statistically equivalent lesion width compared to the 2-cycle uncontrolled group. The mean width of the 9-cycle group was significantly larger than the 2-cycle and 5-cycle groups, while the mean width of the 5-cycle group was also significantly larger than the 2-cycle group. Results of the Bartlett test showed no significant difference between variances of the lesion width among all groups ($p = 0.892$).

For the segmented lesion area [Fig. 6(c)], mean $\pm$ standard error values were 4.78 ± 0.47 mm² for the controlled group, 7.02 ± 0.56 mm² for the 5-cycle group, 9.31 ± 0.58 mm² for the 9-cycle group, and 5.63 ± 0.61 mm² for the 2-cycle group. Lesion area passed the normality test ($p > 0.05$). The ANOVA test indicated a significant difference among the lesion areas for all groups ($p = 9.31 \times 10^{-6}$). Pairwise comparisons of mean lesion areas from the Tukey HSD test are shown in Table 2. As for the lesion width, controlled trials had mean lesion area significantly smaller than 5-cycle or 9-cycle uncontrolled trials but statistically equivalent to 2-cycle uncontrolled trials. Mean lesion area for the 9-cycle uncontrolled group was also significantly larger than the 2-cycle and 5-cycle uncontrolled groups. Variances of lesion area were not significantly different among the four groups ($p = 0.807$).

For the lesion depth [Fig. 6(d)], mean $\pm$ standard error values were 4.39 ± 0.31 mm for the controlled group, 4.28 ± 0.30 mm for the 5-cycle group, 4.66 ± 0.24 mm for the 9-cycle group, and 6.28 ± 0.49 mm for the 2-cycle group. The null hypothesis of the Shapiro-Wilk test was rejected ($p = 4.5 \times 10^{-3}$), indicating a non-normal data distribution. Median ablation depths for the controlled, 5-cycle, and 9-cycle groups were not significantly different based on the Kruskal-Wallis rank-sum test ($p > 0.05$). Median depth for the 2-cycle group was not significantly larger than the 9-cycle group ($p = 5.9 \times 10^{-2}$), but was significantly larger than the controlled group ($p = 7.1 \times 10^{-3}$) and the 5-cycle group ($p = 4.2 \times 10^{-3}$), respectively). Results of the Levene test indicated no significant difference in the lesion depth variance among all four groups ($p = 0.218$).

Discussion

This study evaluated the potential of echo decorrelation imaging as a feedback approach for real-time control of HIFU thermal ablation in *ex vivo* bovine liver. Our methods and results can be compared with a similar recent study, in which Takagi et al. (2016) controlled HIFU exposures in *ex vivo* chicken breast. In that study, tissue coagulation was detected in real time during HIFU treatment by computing the cross-correlation of echo signals during HIFU

treatment with reference signals recorded before treatment. In contrast to the study reported here, Takagi et al. used separate transducers for therapy (1.25 MHz, 128 element, geometrically focused HIFU array) and for imaging (3 MHz sector-format array placed in the center of the HIFU array). They computed cross-correlation maps over longer time scales (minimum 0.3 s, maximum > 5 s inter-frame time between cross-correlated frames), compared to the short time scales (8.6 ms inter-frame time) employed for decorrelation computations here. They also employed larger dimensions for the control ROI ($10 \times 10 \text{ mm}^2$ vs $1 \times 1 \text{ mm}^2$). They observed more homogeneous multi-focal lesions when using treatment feedback control with correlation-coefficient thresholds 0.7 or 0.8, compared to the threshold of 0.9. Average exposure times for each focal lesion were found to range from 4.5–7.8 s for controlled exposures using correlation-coefficient thresholds 0.7, 0.8, and 0.9, comparable to the average exposure time for the controlled group here (5.8 ± 1.9 s). They noted the need for more systematic investigation of optimal control thresholds to judge complete tissue coagulation. Notably, the large inter-frame time used for cross-correlation calculations used in their method can be expected to lessen the specificity of ablation detection in the presence of tissue motion *in vivo*.

Based on the preliminary *ex vivo* experiments reported here ($N = 13$), the echo decorrelation threshold to achieve 90% specificity in predicting local liver ablation was found to be -2.7 ($\log_{10}$ -scaled decorrelation per ms). Analogous thresholds for 90%-sensitivity prediction of *in vivo* ultrasound ablation were found by Fosnight et al. (2017) to be -2.9 for rabbit liver parenchyma and -2.3 for VX2 tumor within rabbit liver. Sensitivity of local ablation prediction in the preliminary *ex vivo* experiments reported here was 83% at this control threshold, higher than the corresponding sensitivities found previously for both ablated liver (65%) and VX2 tumor (43%) *in vivo* (Fosnight et al., 2017). These discrepancies may be associated with differences between *ex vivo* and *in vivo* ablation, including changes in thermal energy deposition due to blood perfusion and tissue motion. Consequently, longer sonication times (Chen et al., 1993) may be required for feedback-controlled HIFU ablation *in vivo* employing the decorrelation thresholds determined here. Other considerations for *in vivo* and clinical controlled ablation are described later in this section.

ROC curves for ablation prediction by echo decorrelation imaging, shown in Fig. 5a, are consistent with previous studies on echo decorrelation monitoring of HIFU thermal ablation (Kumon et al., 2012; Fosnight et al., 2017), with statistically significant prediction of local tissue ablation in every case. Notably, the overall AUC value for controlled HIFU ablation trials (0.956) was significantly higher than for the uncontrolled 2-cycle group (0.722), even though the average sonication time was the same for both groups (5.8 s). The likely reason is that for controlled trials, the minimum cumulative echo decorrelation $(y, z, m)_{cum}$ within the ROI exceeded the control threshold $_{thresh}$, chosen to predict local ablation with 90% specificity, in every case. However, in the 2-cycle group, six of the ten trials failed to meet this threshold within the same ROI. A representative ablation result from a 2-cycle trial failing to meet this threshold is shown in Fig. 5(a)-II. In contrast, the AUC value for the controlled group was slightly lower but statistically equivalent to that for the 5-cycle and 9-cycle groups (AUC = 0.977 and 0.981 respectively). For the 5-cycle and 9-cycle groups, the minimum $(y, z, m)_{cum}$ within the ROI exceeded $_{thresh}$ in all cases, resulting in effective ablation prediction. In addition, the larger lesion areas obtained from longer HIFU exposures

resulted in fewer false-positive predictions of local ablation near the focal zone, thus slightly increasing AUC for these groups.

Substantial false-positive predictions were found outside the focal zone in 30–50% of controlled and uncontrolled trials due to decorrelation artifacts. One likely source of these artifacts is the presence of air-filled cavities in the *ex vivo* tissue, which can move due to US radiation force during sonication. These artifacts did not affect the present control algorithm because of the small dimensions of the control ROI ($1 \times 1 \text{ mm}^2$), which was placed at the focal position. These artifacts are likely not relevant to *in vivo* echo decorrelation imaging, because air cavities do not normally exist within the liver *in vivo*. ROC values for all groups remained high despite these artifacts because high fractions of the ablated and unablated points were still predicted correctly, corresponding to high prediction sensitivity and specificity respectively.

Sensitivity of the control algorithm to ROI size was assessed for the controlled trials, as shown in Fig. 4. Smaller ROI sizes ($\leq 2 \text{ mm}$) showed small differences between the minimum decorrelation $\min$ and the average decorrelation avg within the ROI. However, larger ROIs resulted in much smaller $\min$ than avg values. Thus, when $\min$ is employed as a control end point as in the present study, a small ROI size is appropriate. When avg is used as a control end point, as done by others (Takagi et al., 2016), a larger ROI may also be usable; however, large ROIs may be more susceptible to echo decorrelation artifacts outside the focal region.

Smaller and more consistent thermal lesion areas were obtained here in the controlled ablation group (lesion area mean $\pm$ standard error $4.78 \pm 0.47 \text{ mm}^2$) trials than in the 5-cycle ($7.02 \pm 0.56 \text{ mm}^2$), 9-cycle ($9.31 \pm 0.58 \text{ mm}^2$), or 2-cycle ($5.63 \pm 0.61 \text{ mm}^2$) uncontrolled trials. A reason for the apparently more consistent thermal lesioning in controlled trials is that echo decorrelation control may automatically compensate for tradeoffs between *in situ* acoustic intensities and sonication times required for thermal ablation (Dasgupta et al., 2010). Another possible reason is that the control algorithm may have ceased HIFU treatments either before significant heat-induced tissue vaporization (boiling) occurred, or soon after the onset of any vaporization. Consistent with this interpretation, the control threshold employed here ($-2.7 \log_{10}$ -scaled echo decorrelation per ms) corresponds approximately to the optimal threshold previously found for prediction of temperatures $> 80^\circ \text{C}$ in *ex vivo* radiofrequency ablation (Subramanian et al., 2016). These results suggest that HIFU treatments controlled by echo decorrelation imaging can provide consistent, reproducible, completely ablated thermal lesions regardless of changes in the tissue heating rate, which may be caused by differences in the delivered acoustic power, distortion of the HIFU beam, or variations in tissue properties.

The trials reported here included only individual HIFU exposures, with no mechanical or electronic scanning to create larger lesions. However, the results obtained here have implications for scanned HIFU ablation to create larger thermal lesions. The controlled group showed thermal lesions with significantly smaller average area (4.78 mm^2), width (1.29 mm), and treatment time (5.8 s) than the 5-cycle (7.02 mm^2 , 1.89 mm, 14.5 s) or 9-cycle (9.31 mm^2 , 2.4 mm, 26.1 s) uncontrolled trials. These results suggest that HIFU

ablation controlled by echo decorrelation imaging may be capable of ablating target volumes in shorter-duration procedures compared to uncontrolled ablation. For instance, based on average lesion widths from the present study, a target region of 10 mm width within the image plane could be ablated using an estimated 8 lesions performed by controlled ablation (46.4 s), compared to 6 lesions using 5-cycle uncontrolled ablation (87 s) or 5 lesions using 9-cycle uncontrolled ablation (130.5 s).

Scanning of HIFU exposures using control by echo decorrelation imaging is anticipated to provide uniform thermally coagulated volumes (Takagi et al., 2016) by automatically varying HIFU exposure time as needed. With HIFU exposure times controlled to achieve consistent echo decorrelation, and thus consistent ablation of each focal lesion, heat accumulation induced by successive HIFU exposures will also be more consistent (Takagi et al., 2016), avoiding undesired over- or under-treatment (Ebbini and ter Haar, 2015). The present control algorithm can be straightforwardly extended to monitor and control multiple HIFU lesions (Seip et al., 2002) using echo decorrelation imaging by consecutively placing control ROIs for individual HIFU exposures filling the targeted region.

For future *in vivo* experiments testing HIFU ablation control by echo decorrelation imaging, real-time correction for artifactual decorrelation caused by tissue motion and electronic noise will be needed. A motion and noise compensation method already established for echo decorrelation imaging (Hooi et al., 2015; Fosnight et al., 2017) has been integrated with the present control algorithm by utilizing the computed cumulative echo decorrelation from sham treatments preceding HIFU sonication. In the case of extracorporeal HIFU treatments, respiratory gating may also be useful to reduce motion effects, both for accurate placement of HIFU foci and for accurate monitoring (Okada et al., 2006; van Breugel et al., 2016).

As implemented here, the real-time control algorithm using echo decorrelation imaging provided feedback at a frame rate of 0.34 Hz, the inverse of the 2.9 s sonication cycle length. Temporal resolution of this method could be improved by computing decorrelation using fewer frames of echo data, which would reduce the time consumed in data acquisition, transfer and computation. For example, on the system employed here, acquiring five RF frames instead of 20 frames per cycle would allow echo data acquisition, processing, and transfer to occur in 0.33 s (17% of the currently required time), thus reducing the rest period of each sonication cycle to 0.63 s (29% of its current length) and increasing the feedback frame rate to 0.76 Hz. Temporal resolution could also potentially be improved by shortening the duration of each sonication pulse, with the tradeoff of a smaller HIFU duty cycle.

Other considerations important for *in vivo* use of this control algorithm include the accuracy of HIFU ablation near critical tissue structures such as the hepatic vein and bile ducts. In addition to use of the present control algorithm to ensure ablation within targeted ROIs, distinct ROIs could be defined to include critical structures meant to be spared from thermal ablation. In this context, the control algorithm would cease HIFU exposures if the maximum cumulative echo decorrelation within the safety ROI exceeded the control threshold.

Clinical translation of the proposed real-time feedback control algorithm is feasible, given necessary additional investigations. Optimal echo decorrelation thresholds for control should

be determined for different tumor types (e.g., hepatocellular carcinoma and colorectal metastases) and liver parenchyma states (e.g., cirrhotic or fatty liver), each of which has different acoustic and thermal properties. Uncontrolled preliminary clinical experiments are needed to determine these optimal thresholds before testing of echo decorrelation-controlled ablation in patients. These experiments could determine control thresholds similarly to the procedure employed here, with ROC curves computed by point-by-point comparison of cumulative echo decorrelation maps with ablation results from radiologic imaging or histology. Echo decorrelation thresholds could then be chosen to achieve the required specificity and sensitivity of ablation prediction in each tissue type.

Ablation control using echo decorrelation imaging could be incorporated into commercial extracorporeal USg-HIFU systems used for treating unresectable liver tumors (e.g., Model JC, Chongqing HAIFU Tech Ltd, China). In the Model-JC HAIFU system (Wu et al., 2001), sections of the targeted treatment volume are ablated sequentially with monitoring by 2D B-mode imaging. Using a single-exposure mode (Wu, 2006), repeated HIFU sonications > 1 s duration at each position are employed. After ablation, treatment end points are assessed visually by comparing pretreatment and post-treatment B-mode US images of the treated area. Using the same workflow, 2D echo decorrelation maps could be computed using the RF echo signals acquired after HIFU sonication in each slice. To implement ablation control, treatments would stop once the echo decorrelation exceeded an ablation threshold within an ROI for the current focal position. The system would then proceed to the next focal position and repeat this procedure to ablate the entire targeted volume. This control approach could improve ablation accuracy while reducing treatment times, since only the necessary number of HIFU sonication cycles would be fired at each position.

3D echo decorrelation imaging has also been implemented, based on analysis of volumetric ultrasound echo data (Hooi et al., 2015). Real-time, 3D echo decorrelation imaging has been proposed for monitoring of thermal ablation (Fosnight et al., 2017), and could also be applied to real-time ablation control using methods similar to those reported here. For best performance, a 3D real-time implementation of echo decorrelation imaging would employ 2D matrix array probes (De Luca et al., 2015). True 3D echo decorrelation imaging is necessary when a targeted tissue volume is ablated at once, as in radiofrequency and microwave ablation of liver tumors (Wells et al., 2015; Hansen et al., 2015).

Conclusions

HIFU ablation of *ex vivo* bovine liver tissue was successfully controlled using real-time echo decorrelation imaging feedback. Controlled treatments ablated liver tissue more precisely and in less treatment time than uncontrolled 5-cycle or 9-cycle HIFU treatments. Compared to 2-cycle uncontrolled treatments, controlled treatments provided equivalent ablation time but significantly better capability to confirm local ablation. The precise lesions produced by controlled HIFU exposures could be used to ablate larger volumes by electrically or mechanically scanning, while sparing critical tissue structures. This automatic, ultrasound-based control approach may have advantages over current ultrasound-guided HIFU ablation methods employing manual control with visual feedback.

Acknowledgments

This work was supported by National Institutes of Health (NIH) grant R01 CA158439.

References

- Al-Bataineh O, Jenne J, Huber P. Clinical and future applications of high intensity focused ultrasound in cancer. *Cancer Treat Rev.* 2012; 38:346–353. [PubMed: 21924838]
- Aubry JF, Pauly KB, Moonen C, Haar G, Ries M, Salomir R, Sokka S, Sekins KM, Shapira Y, Ye F, Huff-Simonin H, Eames M, Hananel A, Kassell N, Napoli A, Hwang JH, Wu F, Zhang L, Melzer A, Kim YS, Gedroyc WM. The road to clinical use of high-intensity focused ultrasound for liver cancer: Technical and clinical consensus. *J Ther Ultrasound.* 2013; 1:13. [PubMed: 25512859]
- Barthe PG, Slayton MH, Jaeger PM, Makin IRS, Gallagher LA, Mast TD, Runk MM, Faidi W. Ultrasound therapy system and ablation results utilizing miniature imaging/therapy arrays. *IEEE Int Ultrason Symp.* 2004; 3:1792–1795.
- Bohris C, Schreiber WG, Jenne J, Simiantonakis I, Rastert R, Zabel H, Huber P, Bader R, Brix G. Quantitative MR temperature monitoring of high-intensity focused ultrasound therapy. *Magn Reson Imaging.* 1999; 17:603–610. [PubMed: 10231187]
- Bour P, Marquet F, Ozenne V, Toupin S, Dumont E, Aubry JF, Lepetit-Coiffe M, Quesson B. Real-time monitoring of tissue displacement and temperature changes during MR-guided high intensity focused ultrasound. *Magn Reson Med.* 2017
- Casper AJ, Liu D, Ballard JR, Ebbini ES. Real-time implementation of a dual-mode ultrasound array system: *In vivo* results. *IEEE Trans Biomed Eng.* 2013; 60:2751–2759. [PubMed: 23708766]
- Chen L, ter Haar G, Hill CR, Dworkin M, Carnochan P, Young H, Bensted JPM. Effect of blood perfusion on the ablation of liver parenchyma with high-intensity focused ultrasound. *Phys Med Biol.* 1993; 38:1661. [PubMed: 8272440]
- Chen W, Sanghvi NT, Carlson R, Uchida T. Real-time tissue change monitoring on the Sonablate® 500 during high intensity focused ultrasound (HIFU) treatment of prostate cancer. *AIP Conf Proc.* 2011; 1359:391–396.
- Curiel L, Huang Y, Vykhodtseva N, Hynynen K. Focused ultrasound treatment of VX2 tumors controlled by local harmonic motion. *Phys Med Biol.* 2009; 54:3405. [PubMed: 19436103]
- Darkazanli A, Hynynen K, Unger EC, Schenck JF. On-line monitoring of ultrasonic surgery with MR imaging. *J Magn Reson Imaging.* 1993; 3:509–514. [PubMed: 8324310]
- Dasgupta S, Wansapura J, Hariharan P, Pratt R, Witte D, Myers MR, Banerjee RK. HIFU lesion volume as a function of sonication time, as determined by MRI, histology, and computations. *J Biomech Eng.* 2010; 132:081005. [PubMed: 20670054]
- De Luca V, Benz T, Kondo S, Knig L, Lbke D, Rothlbbers S, Somphone O, Allaire S, Bell MAL, Chung DYF, Cifor A, Grozea C, Gnther M, Jenne J, Kipshagen T, Kowarschik M, Navab N, Rhaak J, Schwaab J, Tanner C. The 2014 liver ultrasound tracking benchmark. *Phys Med Biol.* 2015; 60:5571. [PubMed: 26134417]
- DeLong ER, DeLong DM, Clarke-Pearson DL. Comparing the areas under two or more correlated receiver operating characteristic curves: A nonparametric approach. *Biometrics.* 1988; 44:837–845. [PubMed: 3203132]
- Dunn OJ. Multiple comparisons using rank sums. *Technometrics.* 1964; 6:241–252.
- Ebbini ES, ter Haar G. Ultrasound-guided therapeutic focused ultrasound: Current status and future directions. *Int J Hyperthermia.* 2015; 31:77–89. [PubMed: 25614047]
- Fosnight TR, Hooi FM, Keil RD, Ross AP, Subramanian S, Akinyi TG, Killin JK, Barthe PG, Rudich SM, Ahmad SA, Rao MB, Mast TD. Echo decorrelation imaging of rabbit liver and VX2 tumor during *in vivo* ultrasound ablation. *Ultrasound Med Biol.* 2017; 43:176–186. [PubMed: 27712923]
- Fosnight TR, Hooi FM, Keil RD, Subramanian S, Barthe PG, Yang W, Xiaoping R, Ahmad S, Rao MB, Mast TD. Motion-corrected echo decorrelation imaging of *in vivo* focused and bulk ultrasound ablation in a rabbit liver cancer model. *IEEE Int Ultrason Symp.* 2014:2161–2164.

- Grondin J, Payen T, Wang S, Konofagou EE. Real-time monitoring of high intensity focused ultrasound (HIFU) ablation of *in vitro* canine livers using harmonic motion imaging for focused ultrasound (HMIFU). *JOVE J Vis Exp*. 2015; 105:53050.
- Gudur M, Kumon R, Zhou Y, Deng C. High-frequency rapid B-mode ultrasound imaging for real-time monitoring of lesion. *IEEE Trans Ultrason Ferroelectr Freq Control*. 2012; 59:1687–1699. [PubMed: 22899116]
- Han Y, Hou GY, Wang S, Konofagou E. High intensity focused ultrasound (HIFU) focal spot localization using harmonic motion imaging (HMI). *Phys Med Biol*. 2015; 60:5911. [PubMed: 26184846]
- Hanley JA, McNeil BJ. The meaning and use of the area under a receiver operating characteristic (ROC) curve. *Radiology*. 1982; 143:64–71.
- Hansen PD, Cassera MA, Wolf RF. Ablative technologies for hepatocellular, cholangiocarcinoma, and metastatic colorectal cancer of the liver. *Surg Oncol Clin N Am*. 2015; 24:97–119. [PubMed: 25444471]
- Holland BS, Copenhaver MD. Improved Bonferroni-type multiple testing procedures: Correction to Holland and Copenhaver. *Psychol Bull*. 1988; 104:299.
- Hooi FM, Nagle A, Subramanian S, Mast TD. Analysis of tissue changes, measurement system effects, and motion artifacts in echo decorrelation imaging. *J Acoust Soc Am*. 2015; 137:585–597. [PubMed: 25697993]
- Hynynen K, Darkazanli A, Unger E, Schenck JF. MRI-guided noninvasive ultrasound surgery. *Med Phys*. 1993; 20:107–115. [PubMed: 8455489]
- Hynynen K, McDannold N. MRI guided and monitored focused ultrasound thermal ablation methods: A review of progress. *Int J Hyperthermia*. 2004; 20:725–737. [PubMed: 15675668]
- Illing RO, Kennedy JE, Wu F, ter Haar GR, Protheroe AS, Friend PJ, Gleeson FV, Cranston DW, Phillips RR, Middleton MR. The safety and feasibility of extracorporeal high-intensity focused ultrasound (HIFU) for the treatment of liver and kidney tumours in a Western population. *Br J Cancer*. 2005; 93:890–895. [PubMed: 16189519]
- Iwasaki R, Takagi R, Nagaoka R, Jimbo H, Yoshizawa S, Saijo Y, Umemura SI. Monitoring of high-intensity focused ultrasound treatment by shear wave elastography induced by two-dimensional-array therapeutic transducer. *Jpn J Appl Phys*. 2016; 55:07KF05.
- Kennedy J, Wu F, ter Haar G, Gleeson F, Phillips R, Middleton M, Cranston D. High-intensity focused ultrasound for the treatment of liver tumours. *Ultrasonics*. 2004; 42:931–935. [PubMed: 15047409]
- Klingler HC, Susani M, Seip R, Mauermann J, Sanghvi N, Marberger MJ. A novel approach to energy ablative therapy of small renal tumours: Laparoscopic high-intensity focused ultrasound. *Eur Urol*. 2008; 53:810–818. [PubMed: 18069120]
- Kruskal JB, Oliver B, Huertas JC, Goldberg SN. Dynamic intrahepatic flow and cellular alterations during radiofrequency ablation of liver tissue in mice. *J Vasc Interv Radiol*. 2001; 12:1193–1201. [PubMed: 11585886]
- Kruskal WH, Wallis WA. Use of ranks in one-criterion variance analysis. *J Am Stat Assoc*. 1952; 47:583–621.
- Krzanowski, WJ., Hand, DJ. ROC curves for continuous data. 1. Chapman and Hall/CRC; 2009.
- Kumon RE, Gudur MS, Zhou Y, Deng CX. High-frequency ultrasound M-mode imaging for identifying lesion and bubble activity during high-intensity focused ultrasound ablation. *Ultrasound Med Biol*. 2012; 38:626–641. [PubMed: 22341055]
- Lizzi FL, Muratore R, Deng CX, Ketterling JA, Alam S, Mikaelian S, Kalisz A. Radiation-force technique to monitor lesions during ultrasonic therapy. *Ultrasound Med Biol*. 2003; 29:1593–1605. [PubMed: 14654155]
- Luo J, Ren X, Yu T. Efficacy of extracorporeal ultrasound-guided high intensity focused ultrasound: An evaluation based on controlled trials in China. *Int J Radiat Biol*. 2015; 91:480–485. [PubMed: 25758333]
- Majaron B, Plestenjak P, Luka M. Thermo-mechanical laser ablation of soft biological tissue: Modeling the micro-explosion. *Appl Phys B*. 1999; 69:71–80.

- Mast TD, Makin IRS, Faidei W, Runk MM, Barthe PG, Slayton MH. Bulk ablation of soft tissue with intense ultrasound: Modeling and experiments. *J Acoust Soc Am*. 2005; 118:2715–2724. [PubMed: 16266191]
- Mast TD, Pucke D, Subramanian S, Bowlus W, Rudich S, Buell J. Ultrasound monitoring of *in vitro* radio frequency ablation by echo decorrelation. *J Ultrasound Med*. 2008; 27:1685–1697. [PubMed: 19022994]
- Matsuzawa R, Shishitani T, Yoshizawa S, Umemura SI. Monitoring of lesion induced by high-intensity focused ultrasound using correlation method based on block matching. *Jpn J Appl Phys*. 2012; 51:07GF26.
- Napoli A, Anzidei M, Ciolina F, Marotta E, Cavallo Marincola B, Brachetti G, Di Mare L, Cartocci G, Boni F, Noce V, Bertaccini L, Catalano C. MR-guided high-intensity focused ultrasound: Current status of an emerging technology. *Cardiovasc Intervent Radiol*. 2013; 36:1190–1203. [PubMed: 23474917]
- Ng KKC, Lam CM, Poon RTP, Shek TWH, Fan ST, Wong J. Delayed portal vein thrombosis after experimental radiofrequency ablation near the main portal vein. *Br J Surg*. 2004; 91:632–639. [PubMed: 15122617]
- Okada A, Murakami T, Mikami K, Onishi H, Tanigawa N, Marukawa T, Nakamura H. A case of hepatocellular carcinoma treated by MR-guided focused ultrasound ablation with respiratory gating. *Magn Reson Med Sci*. 2006; 5:167–171. [PubMed: 17139143]
- Qu X, Azuma T, Sugiyama R, Kanazawa K, Seki M, Sasaki A, Takeuchi H, Fujiwara K, Itani K, Tamano S, Takagi S, Sakuma I, Matsumoto Y. Improved highly accurate localized motion imaging for monitoring high-intensity focused ultrasound therapy. *Jpn J Appl Phys*. 2016; 55:07KF04.
- Sasaki S, Takagi R, Matsuura K, Yoshizawa S, Umemura SI. Monitoring of high-intensity focused ultrasound lesion formation using decorrelation between high-speed ultrasonic images by parallel beamforming. *Jpn J Appl Phys*. 2014; 53:07KF10.
- Scheffer HJ, Nielsen K, van Tilborg AAJM, Vieveen JM, Bouwman RA, Kazemier G, Niessen HWM, Meijer S, van Kuijk C, van den Tol MP, Meijerink MR. Ablation of colorectal liver metastases by irreversible electroporation: Results of the COLDFIRE-I ablate-and-resect study. *Eur Radiol*. 2014; 24:2467–2475. [PubMed: 24939670]
- Seip R, Tavakkoli J, Carlson R, Wunderlich A, Sanghvi N, Dines K, Gardner T. High-intensity focused ultrasound (HIFU) multiple lesion imaging: Comparison of detection algorithms for real-time treatment control. *IEEE Int Ultrason Symp*. 2002; 2:1427–1430.
- Shishitani T, Matsuzawa R, Yoshizawa S, Umemura SI. Changes in backscatter of liver tissue due to thermal coagulation induced by focused ultrasound. *J Acoust Soc Am*. 2013; 134:1724–1730. [PubMed: 23927213]
- Souchon R, Bouchoux G, Maciejko E, Lafon C, Cathignol D, Bertrand M, Chapelon JY. Monitoring the formation of thermal lesions with heat-induced echo-strain imaging: A feasibility study. *Ultrasound Med Biol*. 2005; 31:251–259. [PubMed: 15708465]
- Subramanian S, Rudich SM, Alqadah A, Karunakaran CP, Rao MB, Mast TD. *In vivo* thermal ablation monitoring using ultrasound echo decorrelation imaging. *Ultrasound Med Biol*. 2014; 40:102–114. [PubMed: 24239361]
- Subramanian S, Schmidt DT, Rao MB, Mast TD. Dependence of ultrasound echo decorrelation on local tissue temperature during *ex vivo* radiofrequency ablation. *Phys Med Biol*. 2016; 61:2356. [PubMed: 26943026]
- Sugiyama R, Kanazawa K, Seki M, Azuma T, Sasaki A, Takeuchi H, Fujiwara K, Itani K, Tamano S, Yoshinaka K, Takagi S, Matsumoto Y. Real-time feedback control for high-intensity focused ultrasound system using localized motion imaging. *Jpn J Appl Phys*. 2015; 54:07HD15.
- Suomi V, Han Y, Konofagou E, Cleveland RO. The effect of temperature dependent tissue parameters on acoustic radiation force induced displacements. *Phys Med Biol*. 2016; 61:7427. [PubMed: 27694703]
- Takagi R, Jimbo H, Iwasaki R, Tomiyasu K, Yoshizawa S, Umemura SI. Feasibility of real-time treatment feedback using novel filter for eliminating therapeutic ultrasound noise with high-speed ultrasonic imaging in ultrasound-guided high-intensity focused ultrasound treatment. *Jpn J Appl Phys*. 2016; 55:07KC10.

- Vaezy S, Shi X, Martin RW, Chi E, Nelson PI, Bailey MR, Crum LA. Real-time visualization of high-intensity focused ultrasound treatment using ultrasound imaging. *Ultrasound Med Biol.* 2001; 27:33–42. [PubMed: 11295268]
- van Breugel JMM, Wijlemans JW, Vaessen HHB, de Greef M, Moonen CTW, van den Bosch MAAJ, Ries MG. Procedural sedation and analgesia for respiratory-gated MR-HIFU in the liver: A feasibility study. *J Ther Ultrasound.* 2016; 4:19. [PubMed: 27478615]
- van Sonnenberg, E., McMullen, W., Solbiati, L. Tumor ablation. 1. Springer-Verlag; New York, New York: 2005.
- Varghese T, Techavipoo U, Zagzebski JA, Lee FTJ. Impact of gas bubbles generated during interstitial ablation on elastographic depiction of *in vitro* thermal lesions. *J Ultrasound Med.* 2004; 23:535–544. [PubMed: 15098873]
- Wells SA, Hinshaw JL, Lubner MG, Ziemlewicz TJ, Brace CL, Lee FT. Liver ablation. *Radiol Clin North Am.* 2015; 53:933–971. [PubMed: 26321447]
- Wijlemans JW, de Greef M, Schubert G, Bartels LW, Moonen CT, van den Bosch MA, Ries M. A clinically feasible treatment protocol for magnetic resonance-guided high-intensity focused ultrasound ablation in the liver. *Invest Radiol.* 2015; 50:24–31. [PubMed: 25198833]
- Wu, F. Clinical applications of high intensity focused ultrasound in the treatment of patients with solid malignancy. In: Wu, J., Nyborg, W., editors. *Emerging Therapeutic Ultrasound*. Vol. Ch. 8. World Scientific; 2006. p. 279-339.
- Wu F, Chen WZ, Bai J, Zou JZ, Wang ZL, Zhu H, Wang ZB. Pathological changes in human malignant carcinoma treated with high-intensity focused ultrasound. *Ultrasound Med Biol.* 2001; 27:1099–1106. [PubMed: 11527596]
- Wu F, Wang ZB, Chen WZ, Zhu H, Bai J, Zou JZ, Li KQ, Jin CB, Xie FL, Su HB. Extracorporeal high intensity focused ultrasound ablation in the treatment of patients with large hepatocellular carcinoma. *Ann Surg Oncol.* 2004; 11:1061. [PubMed: 15545506]
- Yang H, Shutao W, Thomas P, Elisa K. Fast lesion mapping during HIFU treatment using harmonic motion imaging guided focused ultrasound (HMIgFUS) *in vitro* and *in vivo*. *Phys Med Biol.* 2017; 62:3111. [PubMed: 28323638]
- Zhang L, Zhu H, Jin C, Zhou K, Li K, Su H, Chen W, Bai J, Wang Z. High-intensity focused ultrasound (HIFU): Effective and safe therapy for hepatocellular carcinoma adjacent to major hepatic veins. *Eur Radiol.* 2009; 19:437–445. [PubMed: 18795303]
- Zhou S, Petruzzello J, Anand A, Sethuraman S, Azevedo J. Validating ultrasound-based HIFU lesion-size monitoring technique with MR thermometry and histology. *AIP Conf Proc.* 2010; 1215:53–56.
- Zhou YF. High intensity focused ultrasound in clinical tumor ablation. *World J Clin Oncol.* 2011; 2:8–27. [PubMed: 21603311]

Figure 1.

Experimental setup. (a) 64 element image-treat transducer aligned with *ex vivo* bovine liver during the *in vitro* experiment, (b) Schematic of the experimental setup, (c) The developed real-time feedback control algorithm flow chart, (d) Timing diagram of the treatment scenario.

Figure 2.
 GUI of the implemented Qt-based C++ application used for the HIFU thermal ablation control and imaging; the controlling ROI is bounded by a yellow line.

Figure 3.

Ablation prediction sensitivities and specificities for preliminary uncontrolled HIFU exposures in *ex vivo* liver ($N=13$) plotted vs. echo decorrelation threshold. The blue line represents the chosen ablation control threshold of -2.7 ($\log_{10}$ -scaled echo decorrelation per ms).

Figure 4. Means and standard errors of minimum ($\Delta_{\min}$) and average (Δ_{avg}) cumulative echo decorrelation values vs. ROI size for the controlled group; the blue line represents the ablation control threshold.

Figure 5.

Representative vital-stained histologic sections and hybrid echo decorrelation/B-mode images for controlled and uncontrolled HIFU trials. (I) Controlled group. (II) 2-cycle group. (III) 5-cycle group. (IV) 9-cycle group. Column (a) shows representative treatments at the average acoustic intensity, while column (b) shows representative treatments at the maximum acoustic intensity employed. White lines indicate tissue boundaries segmented in the B-mode images, while yellow dashed lines show contours of the control threshold ($-2.7 \log_{10}$ -scaled echo decorrelation per millisecond) in the echo decorrelation maps. In the tissue sections, red and green boundaries indicate the segmented tissue and ablated regions. Lesion dimensions (width/depth/area) are close to reported means for each group.

Figure 6. Statistical results. (a) ROC curves showing the performance of the echo decorrelation imaging as an ablation predictor for each group. (b) Means and standard errors of ablated lesion width; (c) lesion area; (d) lesion depth. (* $p < 0.05$; ** $p < 10^{-2}$; *** $p < 10^{-3}$)

Table 1

Results of unpaired, two-tailed *t* test comparing thermal lesion widths [*t* statistic (*p* value)].

	Controlled	5-cycle	9-cycle
5-cycle	4.33 (6.3×10^{-4})		
9-cycle	8.01 ($<10^{-16}$)	3.67 (0.004)	
2-cycle	-2.05 (0.188)	6.38 (1.3×10^{-6})	10.06 ($<10^{-16}$)

Table 2

Results of unpaired, two-tailed t test comparing measured thermal lesion areas [t statistic (p value)].

	Controlled	5-cycle	9-cycle
5-cycle	2.84 (0.036)		
9-cycle	5.72 (9.5×10^{-6})	2.88 (0.032)	
2-cycle	1.06 (0.714)	1.77 (0.302)	4.66 (2.4×10^{-4})