ONLINE SUPPLEMENT

SUPPLEMENTARY MATERIALS

Supplemental Table S1. Summary of ultrasound parameters for *in vitro* and *in vivo* experiments performed with an in-house built high frequency ultrasound imaging system and FUS sonication platform.

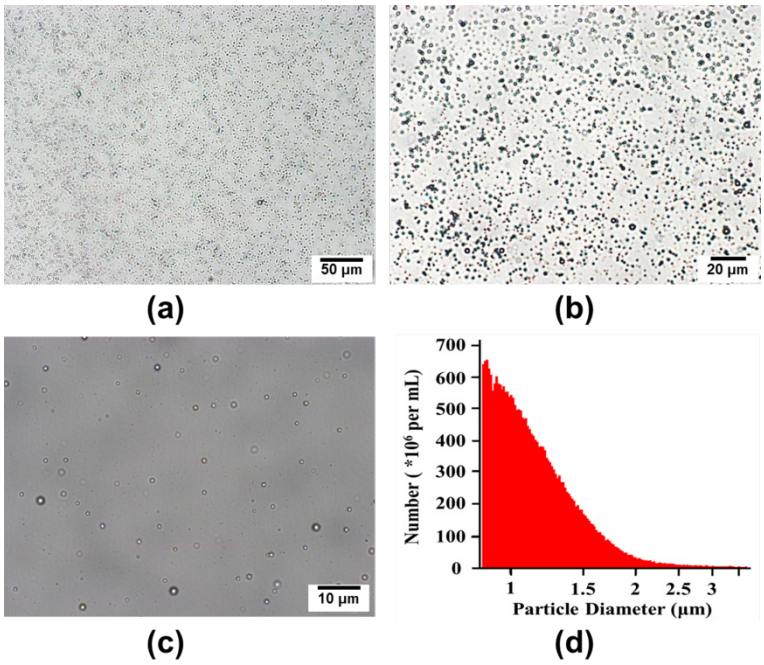
	Central frequency (MHz)	Acoustic pressure (MPa)	Number of cycles	PRF* (kHz)	Exposure Time (s)
In vitro experiments					
Imaging	40	2.4	3	N/A	0.125/frame
MB destruction	2	0.6	10	4	0.5
In vivo experiments					
Imaging	40	2.4	3	N/A	0.125/frame
MB destruction	2	0.6	10	4	0.5
BBB opening	2	0.5/0.7**	2000	0.001	60

*PRF: Pulse repetition frequency. **_In vivo_ animals were mostly exposed to a pressure level of either 0.5 or 0.7 MPa.

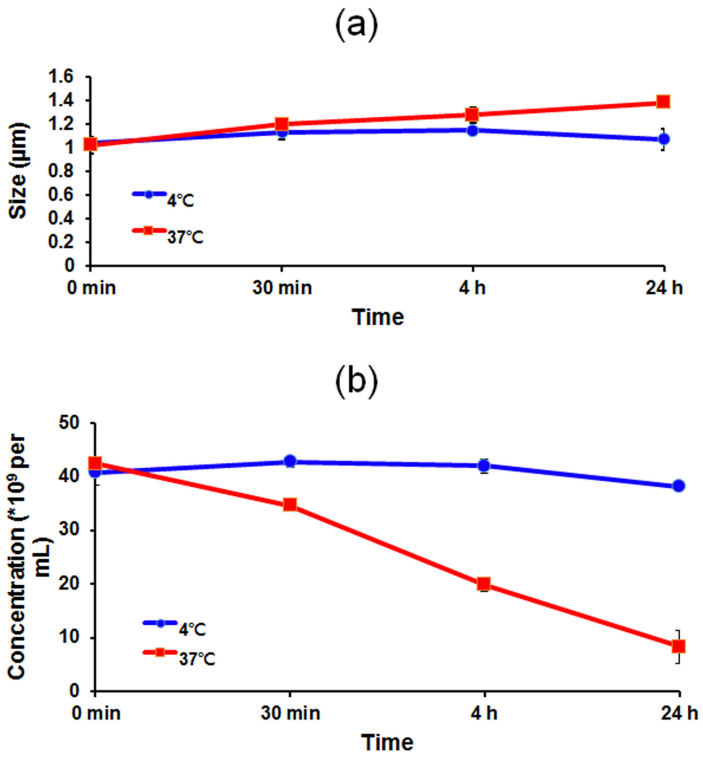
Supplemental Table S2. Summary of ultrasound parameters for *in vivo* experiments performed with a commercial ultrasound imaging system and FUS sonication setup.

	Central frequency (MHz)	Power (%)	Number of cycles	PRF* (Hz)	Exposure Time (s)
Imaging	21	5	N/A	N/A	0.03/frame
MB destruction	21	100	10	N/A	2
BBB opening	2	0.5/0.7 MPa	2000	1	60

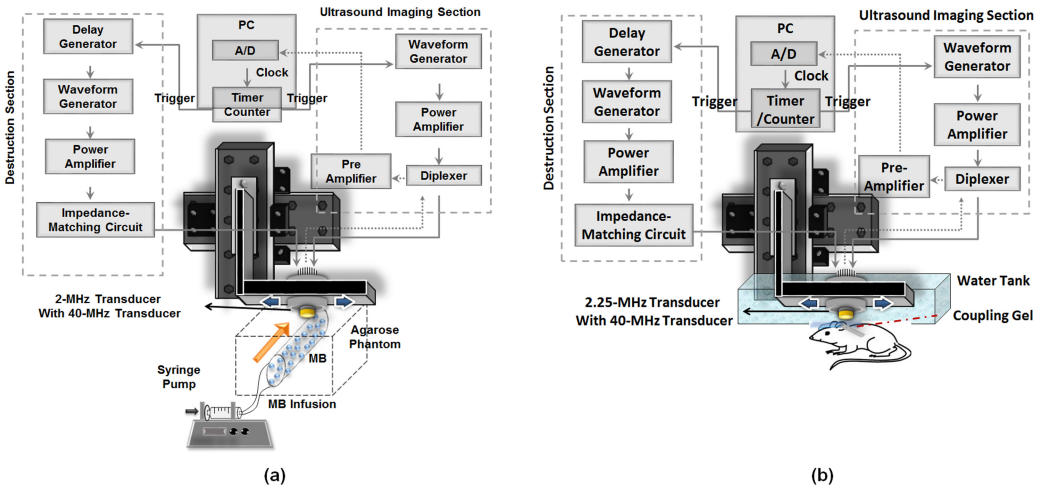
*PRF: Pulse repetition frequency.



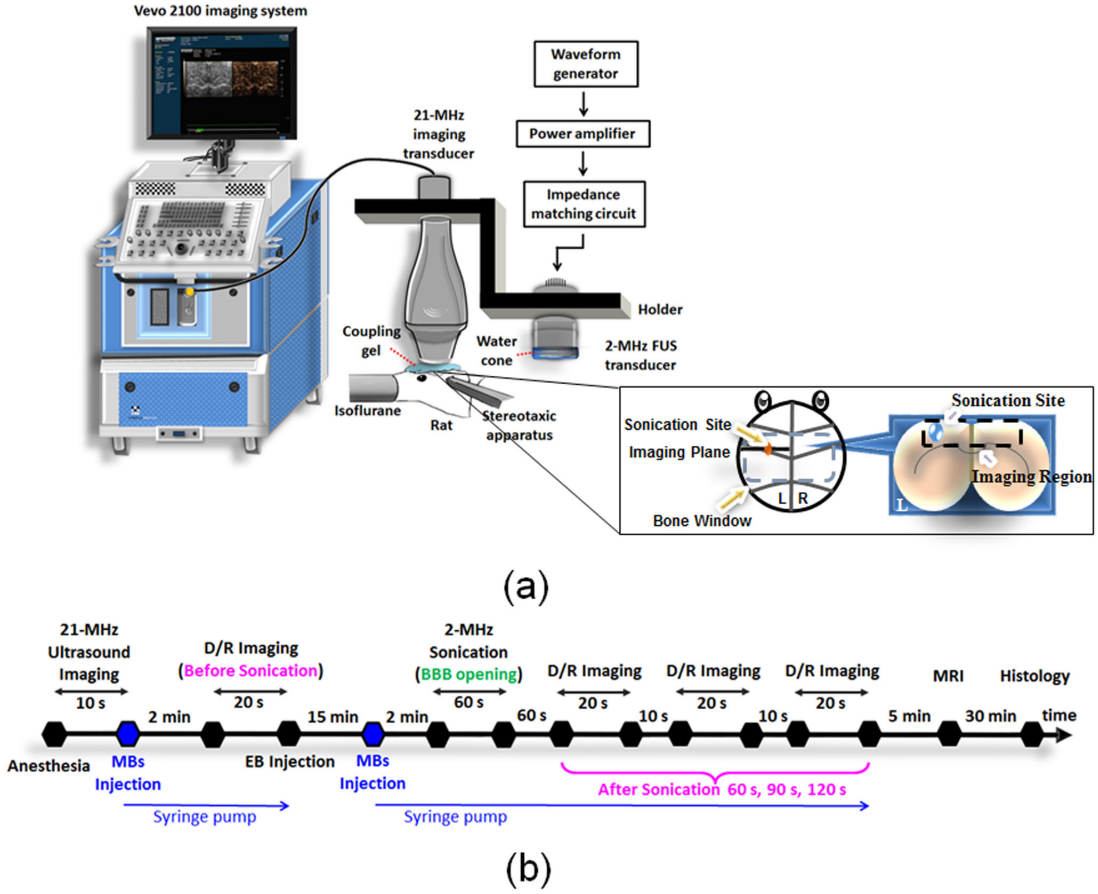
Supplemental Figure S1. (a-c) Light microscopic images of in-house manufactured MBs at 10× (a), 40× (b), and 200× (c) magnification. (d) Measured MB size distribution.



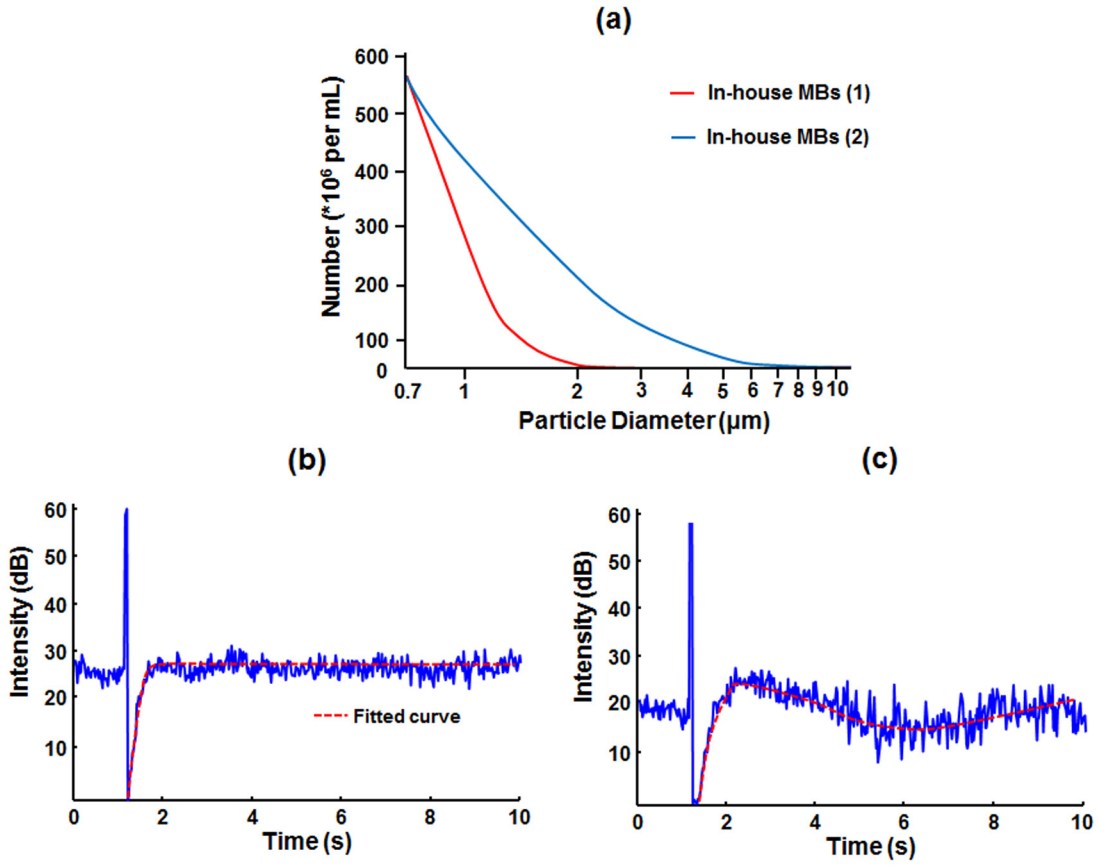
Supplemental Figure S2. The (a) size distribution and (b) concentration of in-house manufactured MBs at 4 °C or 37 °C at different time point (post bubble preparation 0 min, 30 min, 2 h, 4 h and 24 h).



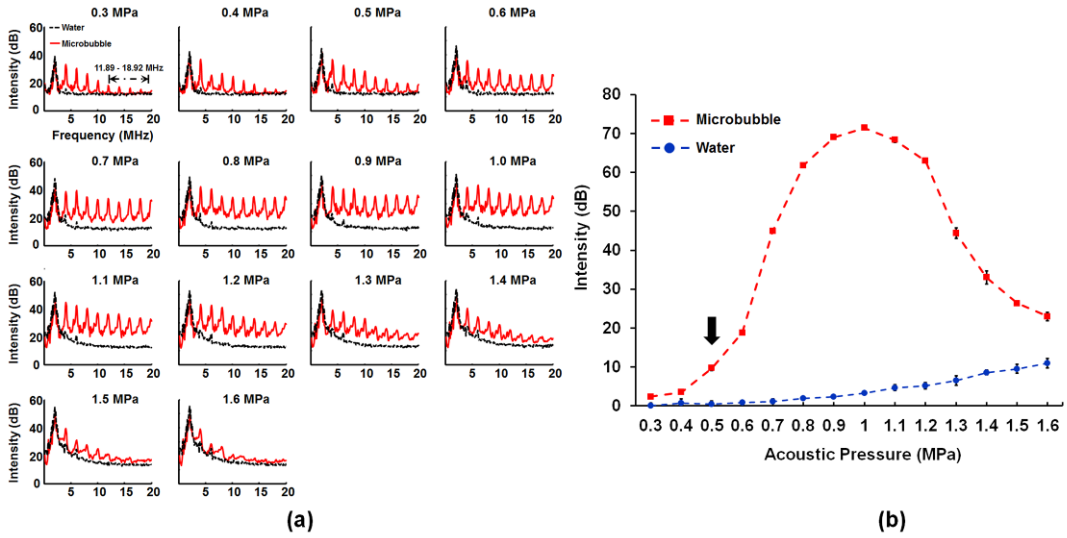
Supplemental Figure S3. (a) *In vitro* experimental setup. (b) *In vivo* experimental setup.



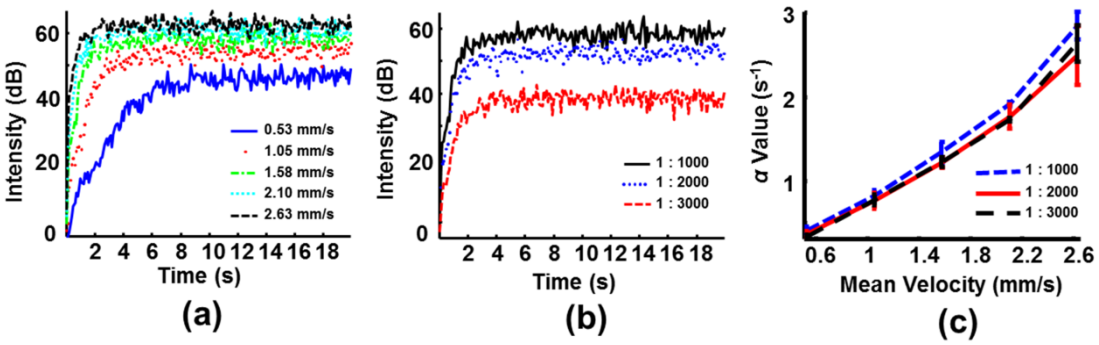
Supplemental Figure S4. (a) *In vivo* experimental setup. (b) *In vivo* experimental protocol.



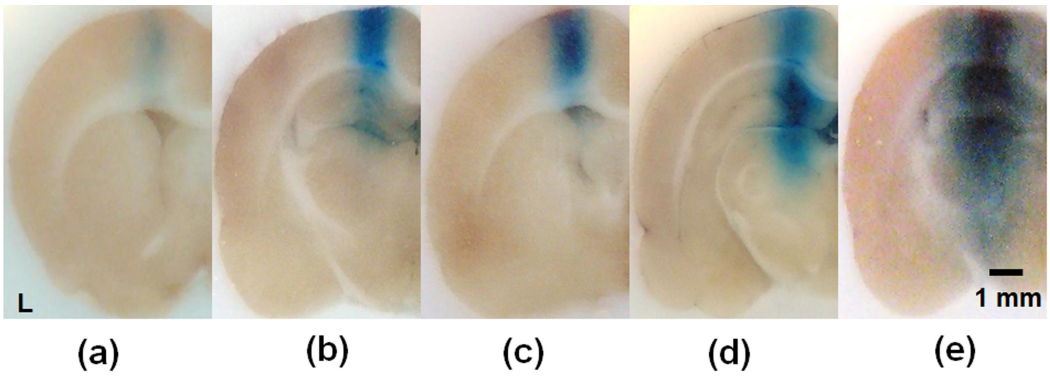
Supplemental Figure S5. (a) Size distribution measurement of two in-house made MBs. Size distribution of the in-house made MBs (2) (0.7 – 9 μm) was wider than (1) (0.7 – 3 μm). (b) TIC response of in-house made MBs (1). (c) TIC response of in-house made MBs (2).



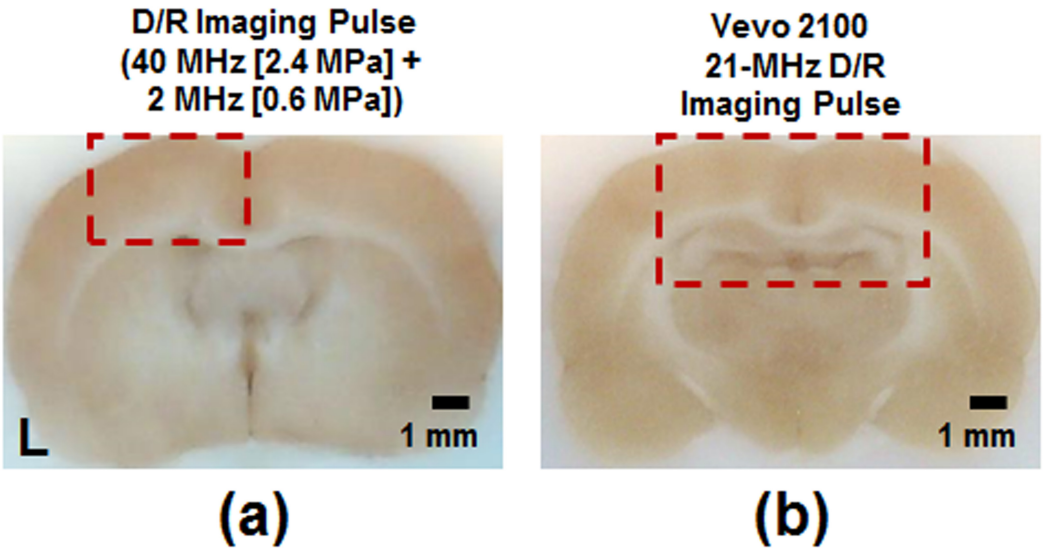
Supplemental Figure S6. (a) Frequency response measured in an *in vitro* vessel phantom under various excitation pressures (ranging from 0.3 to 1.6 MPa; black dashed line = water; red solid line = MB solution); (b) The relationship between inertial cavitation intensity and applied acoustic pressure (arrow: the destruction threshold of MBs).



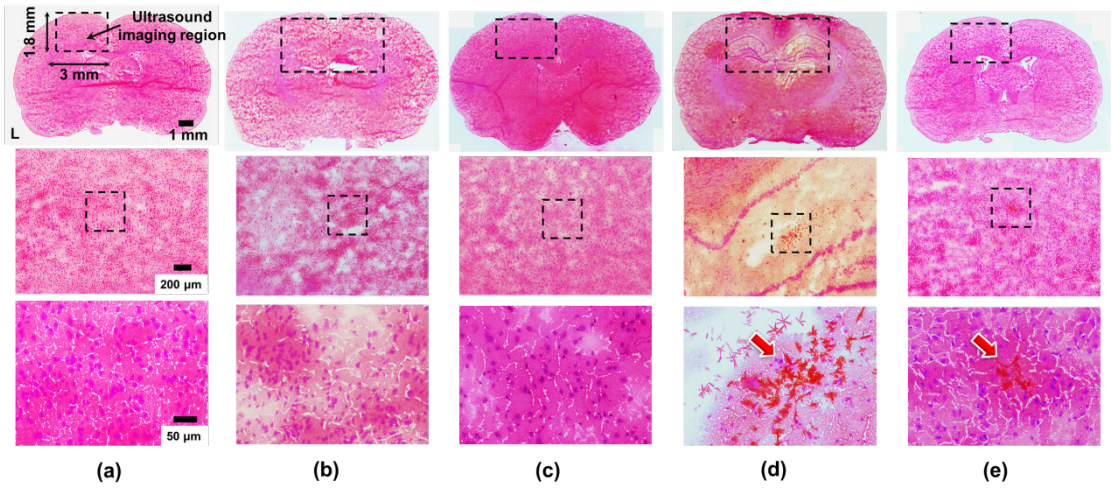
Supplemental Figure S7. (a) TIC response at different perfusion flow rates ranging from 0.53 to 2.63 mm/s. (b) TIC response at different concentrations of MBs ranging from 1000 to 3000 fold dilution, equivalent to 10^5 to 10^6 bubbles/mL in circulation. (c) Estimation of α value at different concentrations of MBs ranging from 1000 to 3000 fold dilution, equivalent to 10^5 to 10^6 bubbles/mL in circulation.



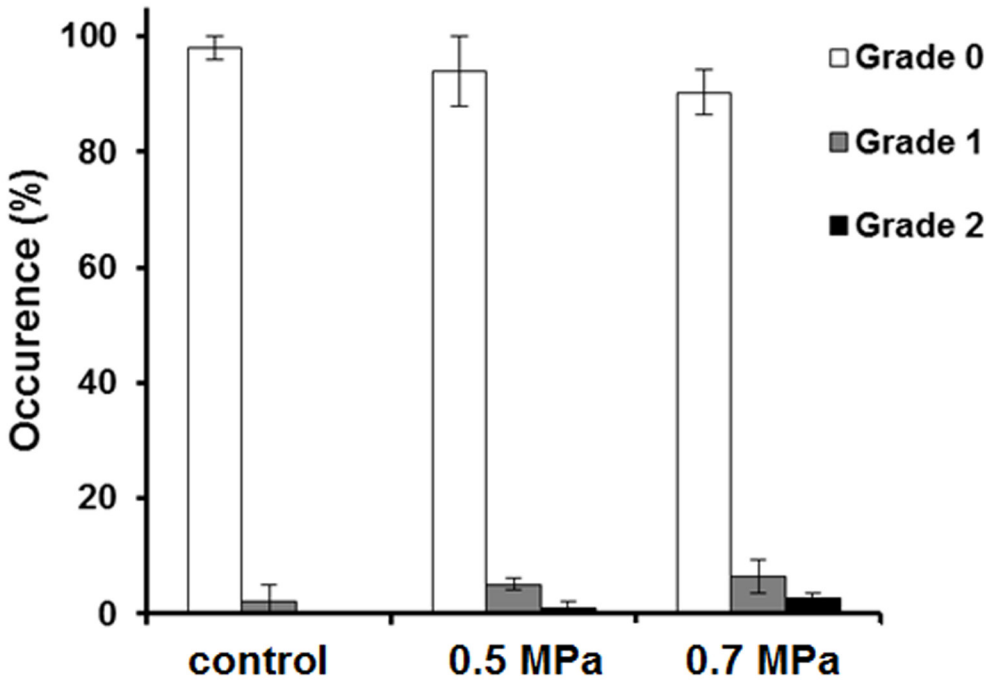
Supplemental Figure S8. EB dye stained brain hemisphere to show the level of FUS-induced BBB opening at different FUS pressures: (a) 0.5 MPa; (b) 0.6 MPa; (c) 0.7 MPa; (d) 1 MPa; (e) 1.5 MPa. Bar = 1 mm.



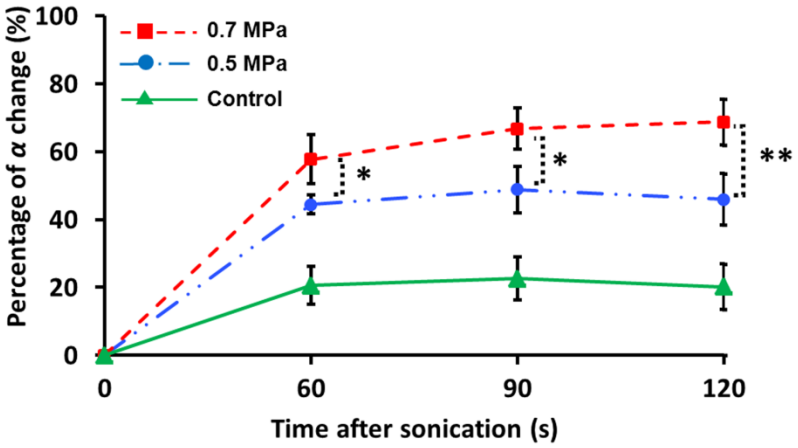
Supplemental Figure S9. Effect of MB-destroying ultrasonic exposure on the animal brain. (a) In-house built 40 MHz ultrasound imaging with 2 MHz MB destruction at 0.6 MPa. (b) Commercial 21 MHz ultrasound imaging (Vevo 2100) with 21 MHz MB destruction at a power of 100%.



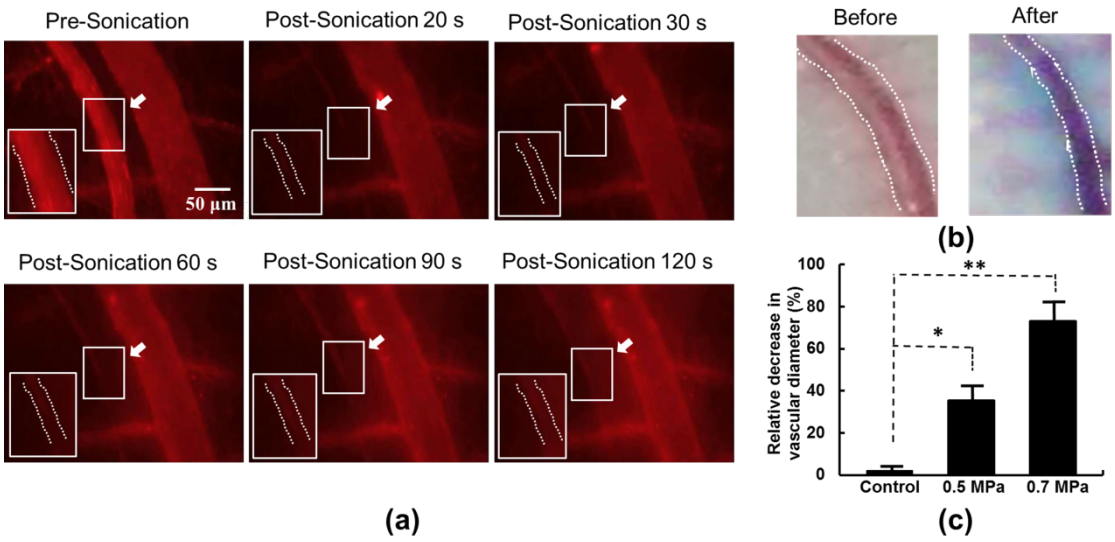
Supplemental Figure S10. H&E staining of the FUS exposed tissues corresponding to Figure 6. Upper: whole brain, bar = 1 mm; middle: 40×, bar = 200 μm; lower: 200×, bar = 50 μm. (arrow: scattered RBC extravasations at the FUS exposure site)



Supplemental Figure S11. Occurrence of brain damage according to sonication at different acoustic pressures characterized by erythrocyte extravasation grading. Grade 0: no detectable extravasations been observed; grade 1: scattered but few erythrocyte cells (less than 10 in microscopic observation) been observed; grade 2: grouped erythrocyte extravasations been observed (larger than 10 but less than 100); grade 3: large-scale extravasations been observed (larger than 100). No grade-3 extravasations been observed either in 0.5- or 0.7-MPa exposure samples.



Supplemental Figure S12. Variance of α-change value after FUS sonication at different time point. Single asterisk, $p<0.05$; double asterisk, $p<0.01$.



Supplemental Figure S13. (a) Subcutaneous fluorescent light microscopic observation of FUS exposure (pressure = 0.5 MPa) in the presence of MBs to cause vasoconstriction under mice dorsal skin window chamber setup. (b) Bright field optical imaging of vessel before and after FUS exposure in the presence of MBs (pressure = 0.5 MPa). (c) Relative decrease in vascular diameter (%) after FUS exposure in the presence of MBs at 0.5 MPa and 0.7 MPa.