Feasibility of monitoring liver metastases from breast cancer in a mouse model using single transducer-harmonic motion imaging *in vivo*, Md Murad Hossain[1], Xiaoyue Li[1], Saurabh Singh[2], Indranil Basu[2], Chandan Guha[2], and Elisa E. Konofagou[1], [1]*Department of Biomedical Engineering, Columbia University, New York, NY and* [2]*Department of Radiation Oncology, Albert Einstein College of Medicine, New York, NY,* mh4051@columbia.edu

Background: Breast cancer is the most frequently diagnosed cancer and 50% of all breast cancer patients develop metastatic disease. Out of all metastatic breast cancer patients, liver metastases develop in approximately 50% of them. If left untreated, liver metastases are associated with poor survival ranging from 4 to 8 months. Early diagnosis or therapy monitoring could help to tailor treatment for individual patients. Change in liver metastases can be monitored via measuring the mechanical properties of the liver. To interrogate the mechanical properties at the "on-axis" to acoustic radiation force, single transducer-harmonic motion imaging (ST-HMI) transmits tracking pulses in-between the discrete excitation pulses. The objective of this study is to test the feasibility of monitoring liver metastases from breast cancer in a mouse model using ST-HMI-induced displacement at multiple frequencies *in vivo*.

Methods: The orthotropic, 4T1 breast cancer mouse model (N=6) was generated by injecting 1 x 10^5 4T1 breast cancer cells in the 4th inguinal mammary fat pad. ST-HMI of both primary cancer and liver was performed at a 7, 10, 13, 20, 26, and 32 days post-injection of tumor cells using Verasonics research system (Vantage 256, Verasonics Inc., Kirkland, WA, USA) with L22-14vXLF (Vermon, Tours, France). ST-HMI was implemented with 13 discrete excitation pulses per period after sampling a continuous excitation pulse composed of the sum of sinusoids with 100:100:1000 Hz to interleave tracking pulses with a PRF of 15 kHz. The center frequency of the excitation and tracking pulse was 15.63 and 20.8 MHz respectively. The displacements with respect to the reference frame were estimated using 1-D normalized cross-correlation with a kernel length of 0.77 mm. For each pixel, estimated displacements were filtered out using the 2nd order Butterworth filter followed by the generation of a peak-to-peak displacement (P2PD) image at 100:100:1000 Hz obtained by averaging P2PD values over 5 cycles. Median P2PD values over the region of interest in the primary tumor and liver were computed.

Results and Discussion: In primary tumors, P2PDs were decreased over time which indicates stiffening, and P2PD across time points were statistically different for 100-500 Hz ($p < 0.05$, Kruskal-Wallis). P2PD at 100-500 Hz detected changes in tumor stiffness as early as between 7 versus 10 days. In the liver, P2PD was also decreased over time but the rate was slower than primary tumor which was expected. The Kruskal-Wallis test indicates that the P2PD was different across time-points for 100-200 and 400-700 Hz and P2PD at 400 Hz detected changes in tumor stiffness as early as between 7 versus 10 days. A good correlation was found between the change in P2PD in the primary tumor versus the liver metastases with the highest R^2 of 0.72 observed at 100 Hz.

Conclusion: This initial study demonstrates the feasibility of using ST-HMI-derived displacements at multiple frequencies to monitor the progression of both primary tumors and metastases in the liver. Future studies will compare P2PD in the liver between the control group versus the tumor group with histopathological validation.