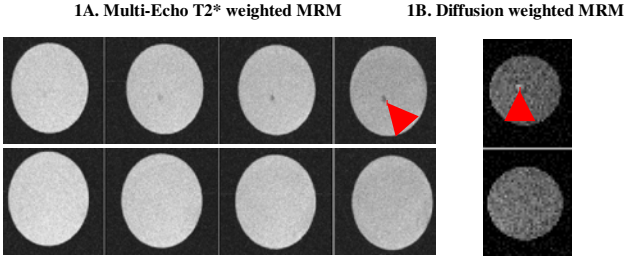
Magnetic Resonance Microscopy of a Glioma Spheriod in a Collagen I Matrix

S. Huang[1], D. Vader[2], D. A. Weitz[2], G. Dai[3], B. R. Rosen[4], T. S. Deisboeck[1,2]

[1]MIT/MGH, Cambridge, MA, United States, [2]Harvard University, Cambridge, MA, United States, [3]MGH, Cambridge, MA, United States, [4]Radiology, MIT/MGH, Cambridge, MA, United States

Introduction: Amongst their distinct characteristics, human high-grade gliomas, particularly glioblastomas, exhibit not only rapid volumetric growth dynamics but also extensive local infiltration into the adjacent brain parenchyma. It is this characteristic local cell invasion that renders these tumors surgically non-curable and motivates ongoing research to better understand the crucial relationship between tumor and its microenvironment. Employing a multicellular tumor spheroid (MTS) model that focuses on glioma MTS evolving within extracellular matrix environments, we have previously investigated the spatio-temporal expansion dynamics *in vitro* [Refs [1-3]] and have started to model them *in silico* [Refs [4-6]]. However, relevance for and applicability of the insights gained from these intriguing interdisciplinary studies to the clinical area will also depend on evidence that the experimental system can be analyzed with methods and techniques that are utilized in clinics as well. As such, NMR-techniques, that currently build the backbone of clinical neuro-imaging, will have to be employed and the results have to be compared with light and fluorescence microscopy methods that are commonly used to monitor MTS growth and invasion in vitro. Here, we have applied magnetic resonance microscopy (MRM) to the glioma MTS model and were able to show that MRM can easily distinguish a multicellular tumor spheroid of roughly 250 µm in diameter in a collagen I matrix, even in the absence of any labeling. It is therefore feasible to use MRM to monitor tumor cell expansion in vitro at a resolution comparable to light microscopy.

Method and Results: Using standard procedures [Ref 7] a human U87MG glioma MTS was allowed to grow in tissue culture medium at 5% CO2 to about 250 µm in diameter before it was placed into a collagen I gel matrix. The MTS was then imaged using MR microscopy at 14T (Bruker Biospin System). Specifically, we acquired multi-gradient echo (TR /TE = 3 8 13 18ms, FOV=0.95cm, 256x256, slice thickness = 400 µm, α=30) images with an in-plane resolution of 37x37 µm2. In addition, we then acquired diffusion weighted spin echo images (TR/TE: 500/15ms, 128x128, FOV: 0.5cm, slice thickness: 400 µm, b-value: 85s mm-2) to depict the different diffusion properties of water in collagen I gel and MTS, respectively. The sample was imaged before and after the MR-procedure using standard light microscopy to ensure proper positioning of the MTS within the assay, and for comparison with the MRM data (data not shown). We observed that the intrinsic MRI property, i.e., the T2 value, was relatively different between the surrounding matrix, i.e. collagen gel, and the MTS (Figure 1A). Furthermore, the diffusion weighted images showed relative restricted diffusion in MTS compared with that in collagen I gel although the diffusion in both contents is comparably fast as reflected by diffusion weighting (Figure 1B).



1A. Multi-Echo T2* weighted MRM 1B. Diffusion weighted MRM

Conclusion: In summary, (1) this preliminary result provides proof-of-feasibility that using a 14T setting MR-microscopy is capable of imaging a microscopic tumor of this size range even without any contrast agent. (2) We therefore argue that such MR-microscopy can serve as a link to the optical imaging level, hence facilitating the transition of results gained with conventional light and fluorescence microscopy toward a more clinically relevant imaging modality for tracking tumor expansion. Lastly (3), this result further warrants the continued use of the MTS model in interdisciplinary brain cancer research. Future work will be directed toward multimodality high-resolution imaging, combining the strengths of molecular imaging techniques with MR-microscopy.

References:

1. Deisboeck T.S., Berens M.E., Kansal A.R., Torquato S., Stemmer-Rachamimov A.O., and Chiocca E.A.: Pattern of self-organization in tumor systems: complex growth dynamics in a novel brain tumor spheroid model. **Cell Prolif.** 34: 115-134, 2001.
2. Gordon V.D., Valentine M.T., Gardel M.L., Andor D., Dennison S., Bogdanov A.A., Weitz D.A., and Deisboeck T.S.: Measuring the mechanical stress induced by an expanding multicellular tumor system: a case study. **Exp. Cell Res.** 289 (1): 58-66, 2003.
3. Kaufman L.J., Brangwynne C.P., Kasza K.E., Filippidi E., Gordon V.D., Deisboeck T.S., and Weitz D.A. Glioma expansion in collagen I matrices: analyzing collagen concentration-dependent growth and motility patterns. **Biophys. J.** 89:635-650, 2005.
4. Mojsilovic A., Rogowitz B., Gomez J., and Deisboeck T.S.: Analysis, reconstruction and visualization of malignant brain tumors: a case study in data synthesis. **Analyt. Quant. Cytol. Histol.** 24: 125-133, 2002.
5. Sander L.M. and Deisboeck T.S.: Growth patterns of microscopic brain tumors. **Phys. Rev. E** 66: 051901, 2002.
6. Delsanto, P.P., Guiot, C., Degiorgis, P.G., Condat, C.A., Mansury, Y., & Deisboeck, T.S.: A growth model for multicellular tumor spheroids. **Appl. Phys. Lett.** 85: 4225-4227, 2004.
7. Kelm, J.M., Timmins, N.E., Brown, C.J., Fussenegger, M., & Nielsen, L.K.: Method for generation of homogeneous multicellular tumor spheroids applicable to a wide variety of cell types. **Biotechnol. Bioeng. 83: 173-180, 2003.**