3D MR Microscopy of Ex Vivo Retina Using Quantitative Susceptibility Mapping

Wei Li[1,2], Eric R. Muir[1], Bryan H. De La Garza[1], and Timothy Q Duong[1,2]

[1]*Research Imaging Institute, University of Texas Health Science Center at San Antonio, San Antonio, TX, United States,* [2]*Ophthalmology, University of Texas Health Science Center at San Antonio, San Antonio, TX, United States*

Audience: Researchers interested in neuronal retina and Quantitative Susceptibility Mapping (QSM)

Introduction The neural retina is characterized by distinct and highly stratified layers (1), including the ganglion cell layer (GCL), inner plexiform layer (IPL), inner nuclear layer (INL), outer plexiform layer (OPL), outer nuclear layer (ONL) and inner and outer segment (IS/OS). External to neural retina, the *choroidal* vasculature constituted another layer, which is sandwiched between the retinal pigment epithelium (RPE) and the sclera. The imaging of these highly stratified layers in retina and surrounding tissues is nontrivial due to the requirements of both contrast and spatial resolution. The recently developed QSM using gradient echo phase is a promising candidate for this purpose. Previous QSM studies have demonstrated its excellent contrast and its unique chemical sensitivity to molecular and cellular components in the brain, especially at higher magnetic field. In this study, we explored the use of ultra high-resolution 3D QSM to delineate the stratified layers of rat retina *ex vivo* at a high field of 11.7T.

Methods An adult Sprague Dawley rat (250-300 g) was euthanized and eyes were enucleated. Extracted eyes were fixed in 10% neutral buffered formalin overnight before imaging (2). MRI was performed on a Bruker 11.7T scanner using a custom-built, small circular surface coil (id~1cm). The eye was scanned using a 3D FLASH sequence with the following parameters: flip angle =25°, TE = 9 ms, TR = 39 ms, 10 averages, FOV = 7.5 x 7.5 x 7.5 mm^3, data matrix = 320 x 320 x 128, yielding an in-plane resolution of 23 x 23 x 57 μm^3. After MRI, the eyes were paraffin embedded and sectioned at 10 µm. H&E staining was performed.

The phase from the gradient echo data was unwrapped using Laplacian-based unwrapping, a modified SHARP method for background phase removal. The magnetic susceptibility was then calculated using the LSQR method (3).

Results and Discussion

Fig. 1 shows a slide of the magnitude, phase and susceptibility images with a matching histology slide and layer assignments. The magnitude only shows weak contrast between different layers. In contrast, both phase and magnetic susceptibility show excellent contrast between different retinal layers around the optic nerve. Interestingly, the intensities of magnitude are similar between different layers, while the corresponding phase and susceptibility values are significantly different across layers (**Fig. 2**). From both the image and the line profiles, the convergences of various retinal layers are evident.

While the biophysical underpinnings of phase and susceptibility contrast among different retinal layers are unknown, it is likely that these layers have characteristic molecular and cellular components with different magnetic properties. The detailed knowledge of the contrast origins may allow us to gain novel information regarding the molecular and cellular components of the retinal layers, and even their microstructures. It should also be noted that though phase also offers excellent contrast, its nonlocal properties should be considered when interpreting the findings. In contrast, the magnetic susceptibility has similar image quality, and is an intrinsic property of tissue, and thus provides a better candidate for quantitative assessment.

Conclusion We applied high-resolution QSM to study the retina ex vivo. Comparing to the low contrast in the magnitude, both gradient echo signal phase and magnetic susceptibility showed excellent contrast between the different layers in the rat retina with high spatial resolution. These layers are consistent with histological findings. Though the contrast mechanisms are still not clear, these results suggest that high resolution QSM at ultra-high magnetic field hold great potential for retina research. Future studies will apply this approach to study the in vivo retina.

References: [1] Wassle and Boycott, Physiol Rev 1991; 1:447. [2] Petiet et al, PNAS 2008; 105: 12331. [3] Li et al, NeuroImage 2011; 55: 1645.

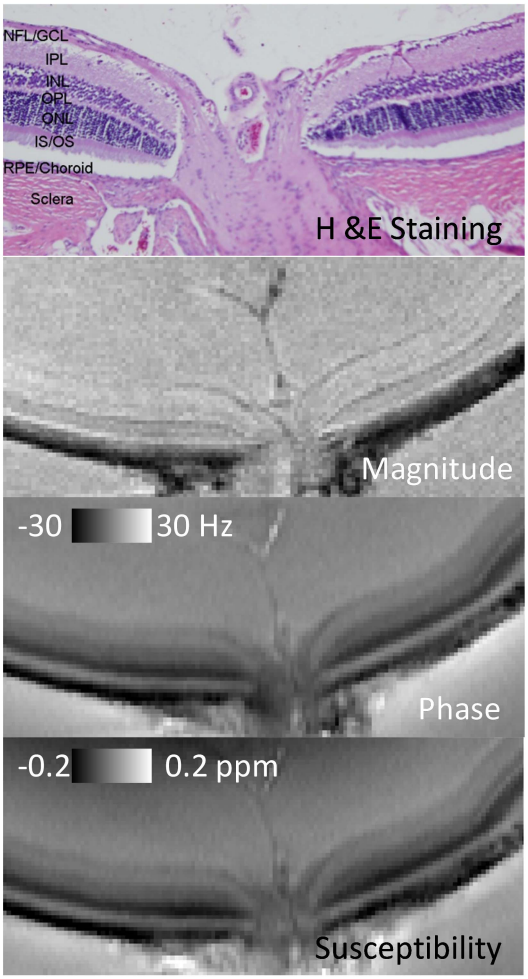

Fig.1. The magnitude, phase and susceptibility images of a rat retina with a matching histological slide.

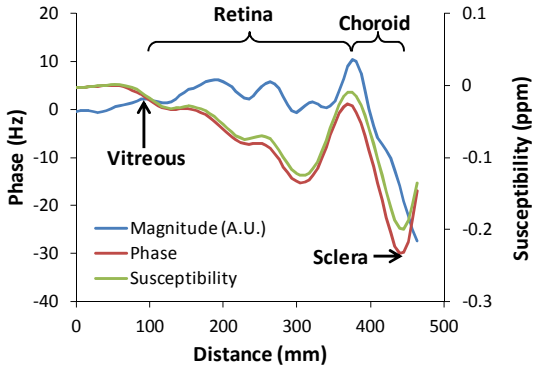

Fig.2. The profile of magnitude, phase and susceptibility across different layers.