632.19 / EEE14 - Simulating the light propagation in rodent brain tissues using ray tracing with a 3D microvasculature model

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Abstract
The advent of two-photon fluorescence microscopy has opened unprecedented opportunities in revealing neuronal firing, hemodynamic changes, and metabolic activity on microscopic level *in vivo*. However, data interpretation of two-photon fluorescence microscopy on dyes with small signal change such as *β-nicotinamide adenine dinucleotide* (NADH), a key metabolic marker, faces enormous challenge because the measured signal change is often highly distorted by hemodynamic changes. Prior work [Baraghis et al, Journal of Biomedical Optics 16, 106003 (2011) and Sauer et al, SFN 2014] modeled two-photon NADH fluorescence with precise maps of cortical microvasculature and corrected for the measured NADH signal change by using the fluorescence change of Sulforhodamine 101 (SR101), a functionally inert dye topically applied to the cortex. However, we only simulated for one animal model (a rat) and one dye, NADH. Here, we extend the prior work to systematically calculate the point to point correction factor using real 3D microvasculatures in two animal models (rats and mice) and two different dyes (NADH and Oregon Green 488 BAPTA-1, OGB). We have found that for both animal models and dyes: 1) the correction factors vary significantly with the appearance of large vessels near the pial surface while they tend to be more homogeneous at deeper depths (Figure: 1st three columns show the correction factor at 40, 60, 80, 100, 120, 140, 200, 300 and 400 microns. 4th column shows the corresponding vasculatures). 2) a single-value correction factor may be used to effectively correct the hemodynamic distortion in layer II or deeper depths. The residual error after the correction is comparable to the experimental error. Our study may help quantify cellular NADH signal change and is also applicable to two-photon fluorescent measurements where changes of tissue optical properties affect measured signals, thus allowing more accurate interpretation of functional imaging studies.