

Abstract

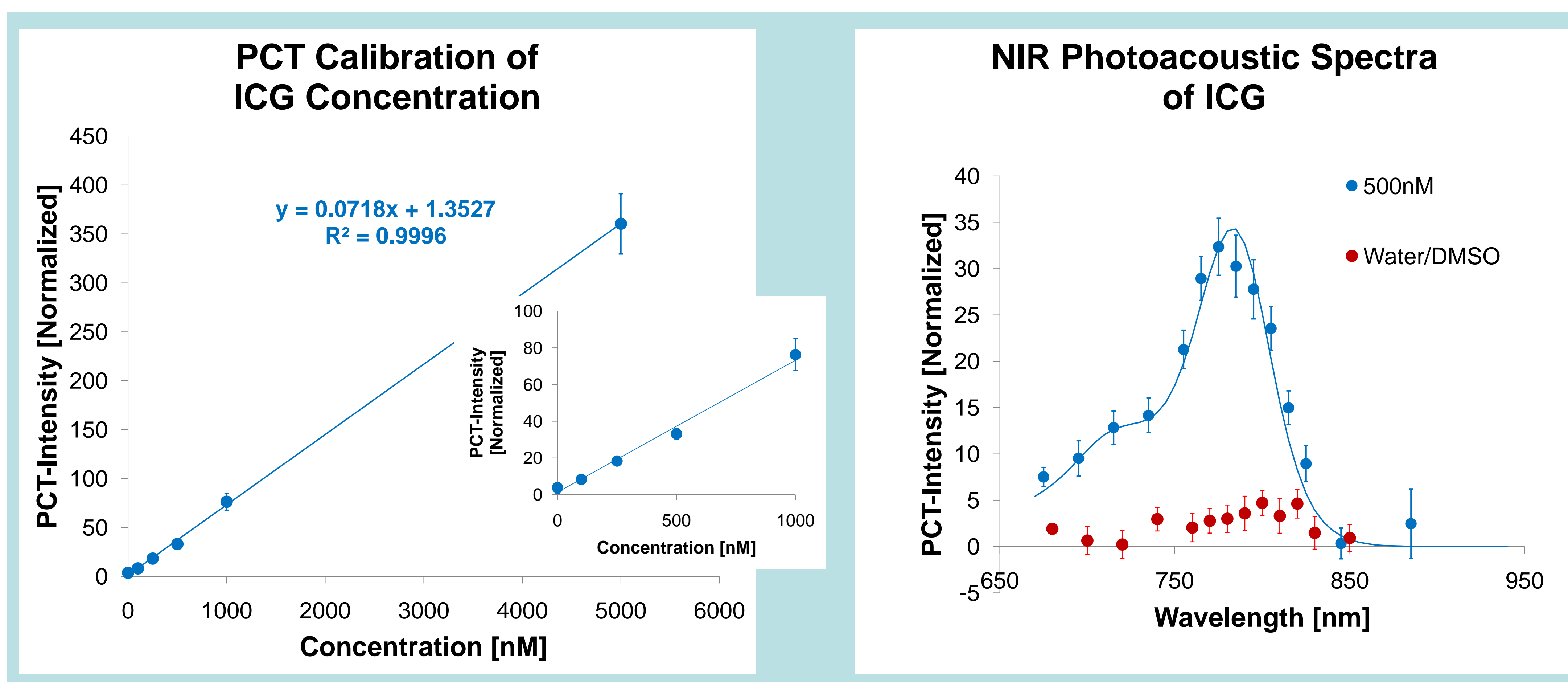
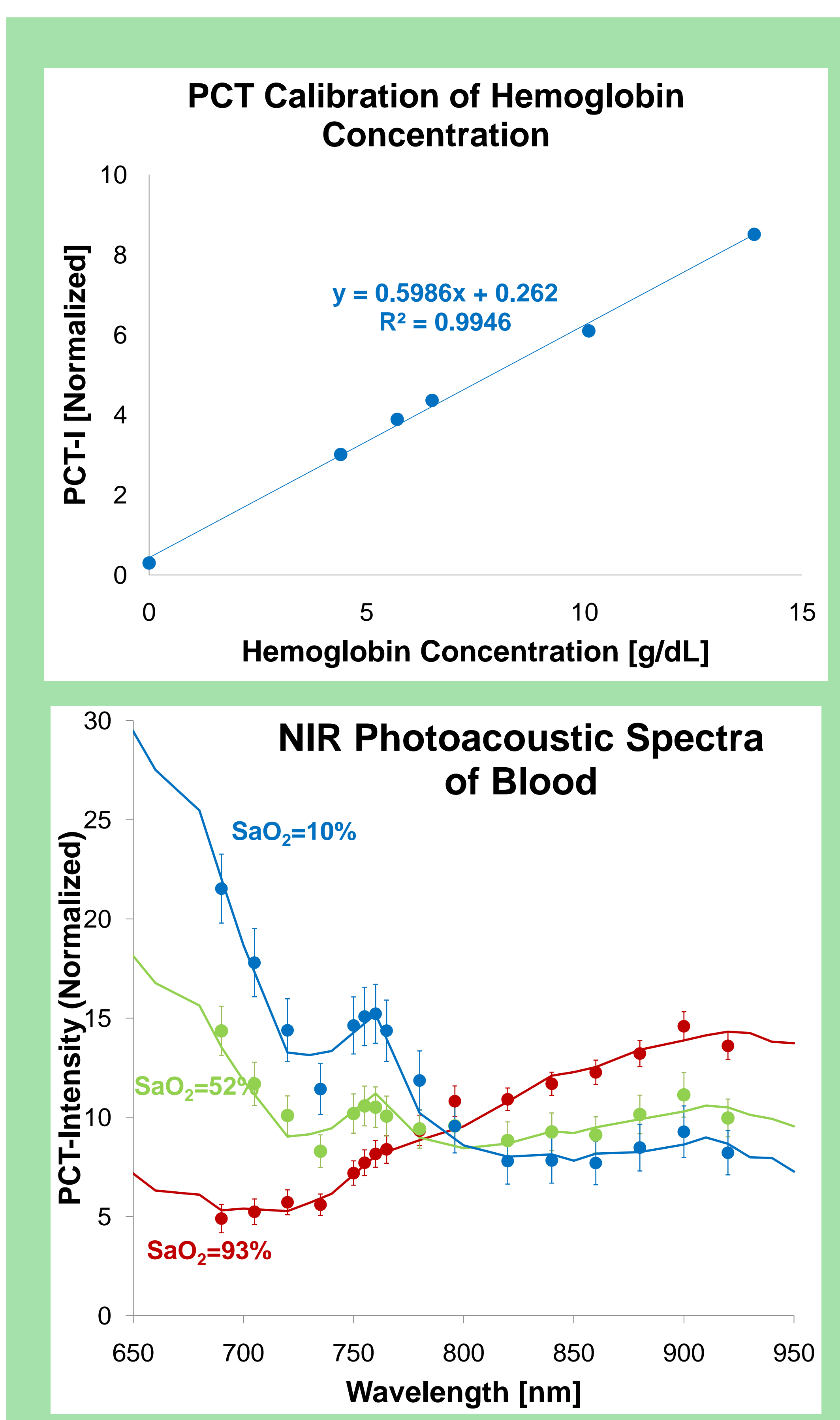
Photoacoustic tomography combines the sensitivity of optical imaging with the resolution of ultrasound, providing a three-dimensional map of molecular or physiological data. Recent technical advances in photoacoustics are enabling practical applications in molecular imaging. We have developed a second generation, volume photoacoustic tomography system, with nanomolar sensitivity and spatial resolution of 280 μm , at depths greater than 20mm, for quantitative molecular imaging in small animals. The spatially resolved concentrations of spectrally distinct chromophores can be determined through the use of multispectral imaging, enabling quantification of endogenous contrast (hemoglobin and oxygenation) and exogenous contrast (blood pool agents and molecularly tagged agents). Over the past five years we have routinely used photoacoustic imaging to study cancer and polycystic kidney-diseased animal models including tissue intra-heterogeneity and labeled antibodies. In this study we will briefly describe the second generation scanner used in this study and the workflow involved in obtaining photoacoustic data. Minimal animal preparation is required and volumetric scan times are as short as one minute, or typically less than 25 minutes for full spectroscopy (14 wavelengths between 690-940nm). We demonstrate applications using endogenous contrast to measure the local concentrations of deoxygenated and oxygenated hemoglobin, providing a quantitative, non-invasive method to track hypoxia in tumors. We use the exogenous contrast agent ICG for blood flow measurements. In addition, we use HER2 antibodies labeled with Licor IRDye800CW to investigate tumor progression. This novel molecular imaging tool is routinely used in our laboratory for a variety of disease and physiology studies without the need for nuclide labeled molecules or specialized siting requirements.

Calibration Spectra

Blood and ICG

Right panel: A calibration study employs a circulation system to change the oxygen partial pressure (PO_2) values of blood through a 1.1mm diameter tube while being scanned. The circulation system consists of a tonometer, an air/nitrogen gas supply, a pump and an oxygen electrode detector. The tonometer consists of a gas-permeable membrane which allows gases to blend with the blood sample. An oxygen electrode detector measures the percent dissolved oxygen (DO) in the blood. Expired donated blood obtained from blood bank (University Hospital, Indiana University School of Medicine) was circulated through the tonometer mixing with the gas and its DO monitored by the oxygen electrode. The amount of DO was controlled and maintained by continuously bubbling air and nitrogen into the reservoir of blood within the tonometer. Upon reaching equilibrium (1-2% variation in DO), a sample was extracted, and scanned in the preclinical PCT-1 scanner (Endra, Inc.) and co-oximeter. The top plot shows a linear relationship of PCT intensity as a function of hemoglobin concentration; the bottom plot demonstrates the ability of the photoacoustic scanner to measure different oxygen saturations levels by fitting NIR spectra to a database of hemoglobin absorption spectra.

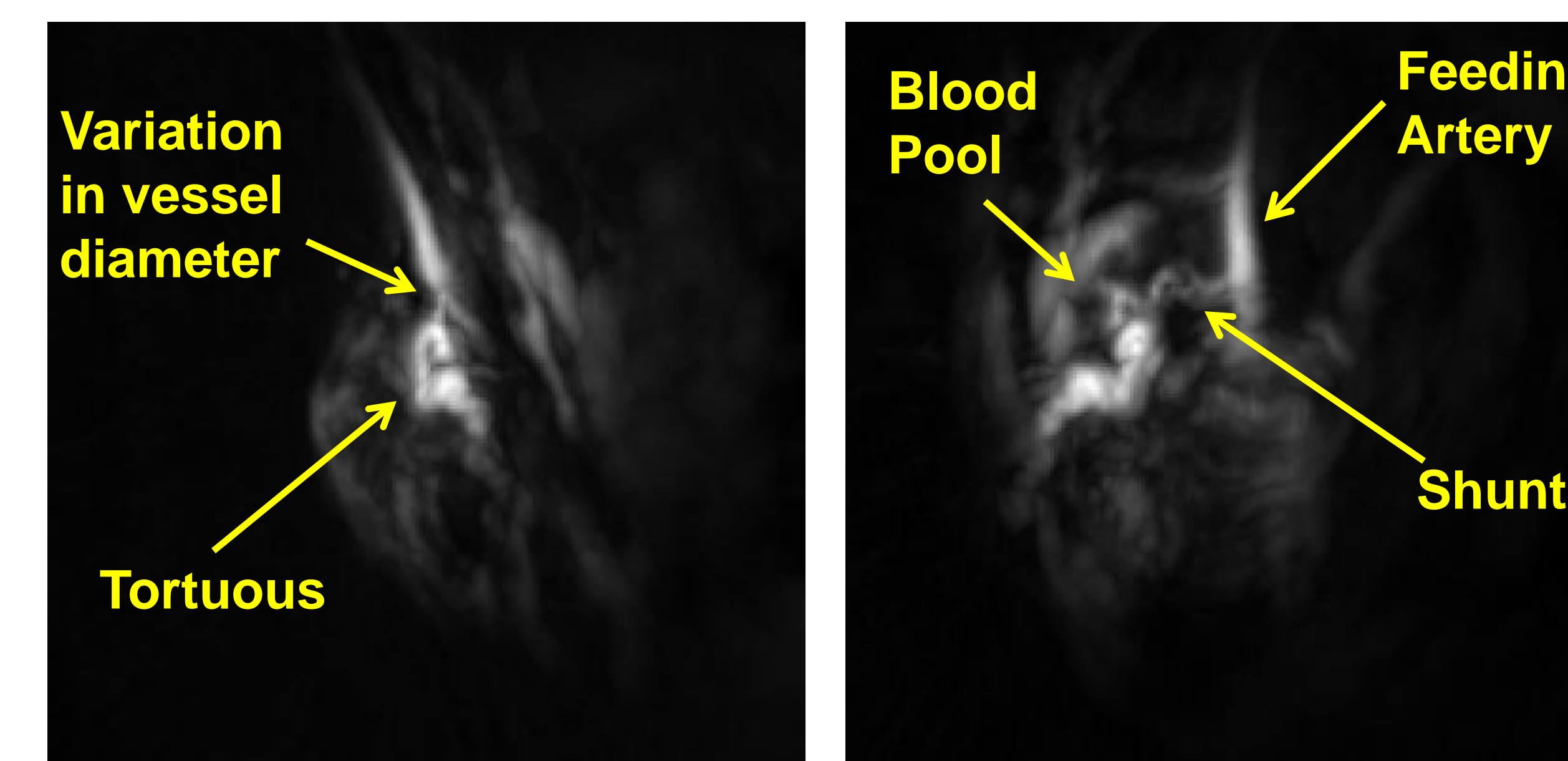
Bottom panel: Indocyanine green (ICG) is an FDA approved contrast agent. ICG contrast is proportional to photoacoustic intensity at 780 nm (left plot), and its spectra is consistent with photospectrometer measurements of its absorbance (right plot)..



Oncological Studies

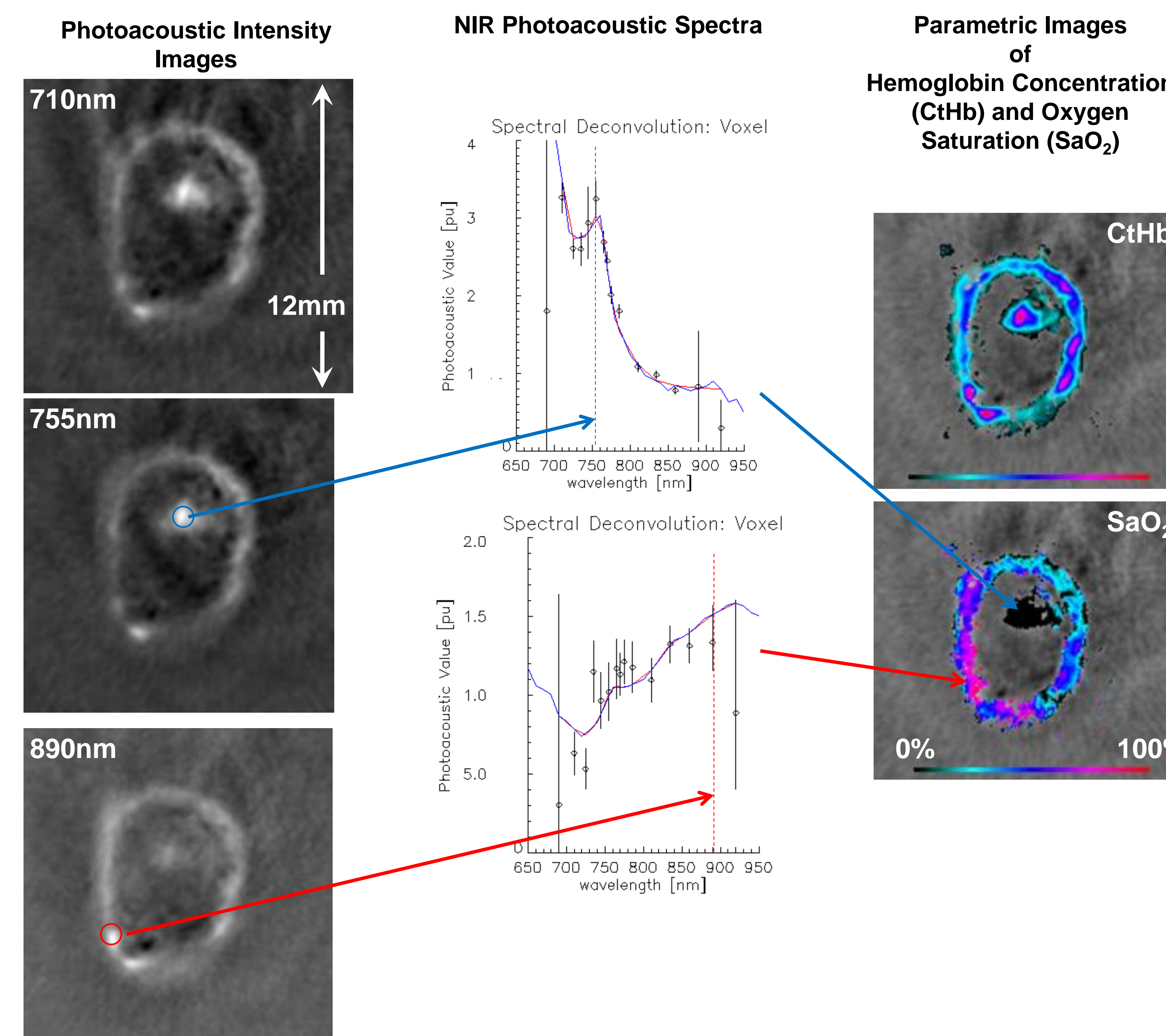
Vascular Morphology

Tumors initiated with MCF7 breast cancer cells transfected with the VEGF165 gene is imaged using the a preclinical photoacoustic CT scanner (PCT-1; Endra, Inc). The above figures display maximum intensity projections (MIPs) at two different angles, where image intensity is proportional to hemoglobin concentration (C_{thb}). From these images, key structural features of the vasculature can be identified, such as variations in vessel diameter, tortuous vascular structure, potential blood pools, feeding arteries, and vascular shunts.



In Vivo NIR Photoacoustic Spectroscopy

Photoacoustic images of tumors initiated with MCF7 breast cancer cells transfected with the HER2 were taken at wavelength ranging from 690 to 920nm. A cross-sectional image of a tumor is displayed below (left) for optical wavelengths 710, 755, and 890 nm. From these images, a clear change in photoacoustic intensity is observed indicative of variations in oxygen saturation levels. ROIs are drawn within two vascular features, one on vasculature interior (blue circle) to the tumor and a second at the periphery of the tumor (red circle). Spectral plots clearly identify blood with negligible oxygen content (top plot, middle column) and relatively high oxygen levels (bottom plot, middle column). Each voxel is fit a linear combination of oxy- and deoxygenated hemoglobin spectra to obtain parametric maps of hemoglobin concentration (top plot, right column) and oxygen saturation (bottom plot, right column).



Conclusion

The preclinical scanner PCT-1 (Endra, Inc.) provides volumetric spectral data capable of non invasively visualizing tumor vascular morphology and assessing intra-tumor heterogeneity in hemoglobin concentration and oxygen saturation.

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