

Researchers at the Texas A&M University have developed a new method for using the photoacoustic effect to create images. The technique allows for functional imaging of oxy and deoxyhemoglobin with an axial resolution of about 15 μm , a lateral resolution of 45 μm , and an imaging depth of 3 mm.

"This technique measures optical contrast based on physiological parameters, such as the total hemoglobin concentration and the oxygen saturation of hemoglobin," explained Lihong Wang, of Texas A&M's Optical Imaging Laboratory in College Station, Tex.

The photoacoustic effect is an ultrasonic wave created when tissue is irradiated by a short-pulse laser. Some of the laser light is absorbed and converted partially into heat, which then creates a rise in pressure through thermo-elastic expansion. The pressure rise moves through the tissue as a sound wave that can be detected by ultrasonic transducers.

"Since ultrasonic waves are much less scattered than optical waves, photoacoustic imaging combines the contrast of optical absorption with the spatial resolution of ultrasound," Wang explained. The technique is really a cross between absorption spectroscopy and ultrasound. In essence, the group picks the absorption band of hemoglobin and shines the laser light on the tissue. Then, they listen to the ultrasonic wave and use that to create an image. "Ultrasonic scattering is 100 to 1000 times less than optical scattering," Wang continued. "As a result, ultrasound can provide much better resolution than light can for structures located deeper than 1 mm below a tissue surface."

To create their functional photoacoustic microscope, the group used a tunable dye laser generating pulses of 6 ns. The dye laser is pumped by an Nd:YAG laser. The group transmits the laser through an optical fiber to the scanning head and uses a photodiode to calibrate the laser energy. The laser beam passes from the fiber through a conical lens that makes a ring-shaped illumination pattern. The illumination ring is focused into the tissue and the focal area overlaps with the ultrasonic focal area, although the optical focus is wider. The ring shaped pattern is intended to reduce the photoacoustic effect that is generated within the field of view of the ultrasonic transducer. The system is set up to detect the laser-generated photoacoustic signal in reflection mode.

The focal diameter of the ultrasonic transducer determines the system's lateral resolution. "If the laser pulse is sufficiently short," Wang said, "a high numerical aperture acoustic lens and a high-center-frequency ultrasonic transducer provides high lateral resolution. A wide-band ultrasonic transducer provides high axial resolution."

According to Wang's published research on the topic, when the center frequency of the transducer exceeds 10 MHz, the penetration depth of the ultrasonic wave determines the maximum imaging depth – not the penetration of the excitation light. In their July 2006 report, Wang and his other researcher reported using a Panametrics 6 mm ultrasonic detector with a 50 MHz center frequency and a 70 percent bandwidth. Coupled with a homemade spherically focusing lens with an NA of .44, a focal zone of .3 mm and a focal length of 6.7 mm the group was able to create images with an axial resolution of 15 μm , a lateral resolution of 45 μm at a depth of up to 3 mm in living tissue.

Hemoglobin and melanin are responsible for most of the optical absorption that creates the photoacoustic effect. As a result, by choosing the correct excitation wavelength, the group can detect blood, which they say allows for high contrast, specific images of the microvasculature. According to their published research, by tuning the laser to 584-nm, they are able to select the wavelength where oxygenated and deoxygenated hemoglobin have the same molecular extinction coefficient. As a result, the image contrast becomes dependent on the total concentration of hemoglobin, but does not reflect changes in oxygenation levels. This could be useful in imaging the blood vessels that characterize rapidly growing tumors and thereby estimate the effectiveness of therapies that target angiogenesis.

The group also demonstrated same technique with dual-wavelength imaging using 584-nm light to image blood vessel proliferation in a tumor and then re-imaging the same section with 764-nm light that misses the absorption peaks of hemoglobin and melanin and penetrates deeply into the tumor to provide information about the thickness of a particular tumor. Combining the two images creates a high contrast image capable of resolving individual microvessels roughly 50 μm in diameter. The group also can use the dual wavelength capability to measure oxygen saturation in individual blood vessels.

Wang's group first tested the oxygen saturation measurements using bovine blood ex vivo and compared their results with a standard optical oxygen saturation measurement. When the results proved to be similar, they used the system to measure normal oxygen saturation in live human tissue, detecting a saturation level of 97 ± 2 percent for arterial blood, and 77 ± 2 percent for venous blood. They also demonstrated that the system can respond accurately to a shift from normal oxygenation to hypoxic conditions. However, the system not only created an accurate measurement of overall oxygen saturation, it also can map the saturation levels of individual blood vessels. This, the group thinks, may be useful in conducting functional brain imaging.

At present, however, the system is somewhat slow. A one-dimensional scan in both the depth and the transverse directions takes about 10 seconds to produce. It takes a full 18 minutes to get a two-dimensional image of a 64 square mm area, and more than 2 hours for the oxygen saturation measurements. A single one dimensional depth image takes only about 2 seconds. Wang and his colleagues acknowledge this and posit that by increasing the laser repetition rate and using an array of ultrasonic transducers, they can make the system faster.

Wang said they plan to begin studying the new method in clinical applications such as melanoma imaging.

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