# Ultrasound resolution beats the diffraction limit

With an acoustic analogue of a Nobel Prize–winning optical technique, researchers can acquire detailed images quickly.

In many ways, ultrasound waves are ideally suited to noninvasive biomedical imaging. They're easy and inexpensive to produce and detect, and they can penetrate deep into tissue without losing their coherence or causing damage. But because of diffraction, conventional ultrasound imaging—like conventional optical microscopy—is limited in resolution to about half a wavelength. In clinical ultrasound applications, which use wavelengths between 200 μm and 1 mm, that limit precludes the imaging of many important structures, including small blood vessels. Shorter wavelengths yield better resolution, but they also penetrate less deeply into tissue.

For optical applications, innovative fluorescence techniques have been devised to overcome the diffraction limit, as recognized by the 2014 Nobel Prize in Chemistry (see PHYSICS TODAY, December 2014, page 18). Inspired by that work, Mickael Tanter and his colleagues at the Langevin Institute (affiliated with ESPCI, Inserm, and CNRS) in Paris have now developed a superresolution ultrasound technique,[1] which they've used to image the blood vessels in a rat's brain with 10-μm resolution, as shown in figure 1. Applying the technique in humans could help to detect cancer and other diseases that alter blood-flow patterns.

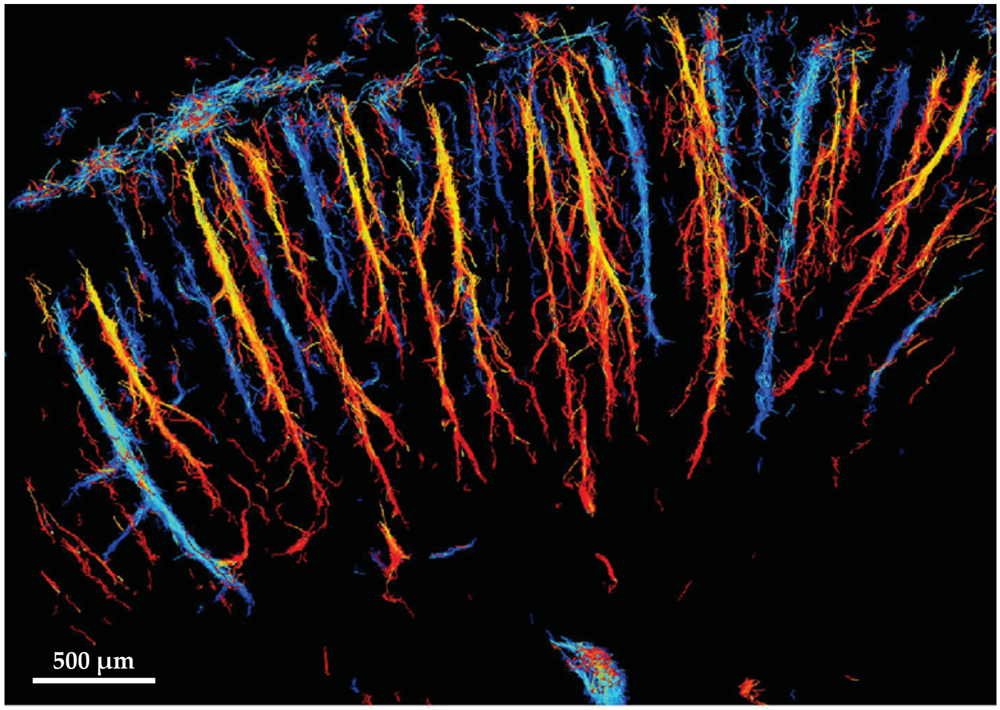


**FIGURE 1. SUPERRESOLUTION ULTRASOUND IMAGE** of the blood vessels in the cortex of a rat's brain. The colors represent velocity: Dark and light blue indicate blood flow in the direction of the skull (toward the top of the image), and red and yellow indicate flow away from the skull. (Courtesy of Mickael Tanter.)

## Fluorophores and microbubbles

In conventional fluorescence microscopy, one decorates a specimen of interest with fluorescent molecules, or fluorophores, that emit photons at a characteristic wavelength when they're optically excited. Each fluorophore produces a diffraction-limited blur of light, several hundred nanometers in diameter; the blurs from all the fluorophores overlap and combine to yield a low-resolution image.

But if the blur from a single fluorophore can be somehow isolated (for example, by rendering all the surrounding fluorophores temporarily unable to fluoresce), then its center—the fluorophore's position—can be pinpointed precisely. Within a few months of one another in 2006, three groups—led, respectively, by Eric Betzig of Janelia Farm, Xiaowei Zhuang of Harvard University, and Samuel Hess of the University of Maine—published imaging techniques based on that principle.[2] In each case, the researchers repeatedly imaged the specimen, with a different set of fluorophores activated each time, to build up a high-resolution picture.

Tanter learned about the new superresolution optical techniques from Hess in 2009, when the two researchers were invited lecturers at a summer course at Cold Spring Harbor Laboratory in New York. Tanter got the idea for a similar technique based on ultrasound and worked with his Langevin Institute colleagues, including Olivier Couture and PhD student Claudia Errico, to bring it to fruition.

In the role of the fluorophores the ultrasound technique uses micron-sized bubbles of inert gas. Because they're strong scatterers of ultrasound and safe when injected into the bloodstream, microbubbles are an established means of enhancing acoustic contrast in medical imaging. But unlike fluorophores, their scattering can't readily be turned off and on. Another approach would have to be found to make the bubbles behave as nonoverlapping point sources—the key requirement for superresolution imaging.

One possible solution is simply to use fewer of them: If the bubbles themselves are hundreds of microns apart, their ultrasound signals don't overlap. That approach has recently been tried by researchers at Imperial College London and Kings College London.[3] They imaged the blood vessels in a mouse's ear with subwavelength resolution, but it took a long time—on the order of an hour—to build up the composite images.

From the start, Tanter and colleagues took a different tack: using ultrafast ultrasound to image the rat's brain at 500 frames per second and looking at the differences between successive images. Most of the imaged region changes little in the short time between frames, so its contribution cancels out. But the bubbles that move appreciably or disintegrate show up as wavelength-sized blobs. The Paris researchers could then fit those blobs to Gaussian profiles, as shown in figure 2, and locate their centers—the bubbles' positions. By superposing the bubble positions from thousands of difference images taken over 2.5 minutes, they obtained high-resolution composite images of the rat's blood vessels. And because they can track individual bubbles from image to image, they can also deduce the velocity of blood flow in each vessel, as indicated by the colors in figure 1.

## Twisty vessels

Several disease conditions that disrupt blood circulation could potentially be diagnosed with the new technique. Some, such as stroke, are characterized by changes in blood flow through existing vessels; others, such as cancer, change the structure of the vessels themselves. To supply their cores with nutrients, large tumors must grow their own blood vessels, which differ in many ways from those in healthy tissue. But even small, early-stage tumors influence the architecture of the surrounding blood-vessel network: Instead of being straight and direct, the vessels become winding and tortuous.

Paul Dayton and his group at the University of North Carolina, Chapel Hill, have been developing quantitative methods to distinguish tumor-bearing from healthy tissues based on ultrasound images of blood vessels.[4] The images they used were subject to the tradeoff between diffraction-limited resolution and penetration. The new high-resolution imaging technique could make their methods

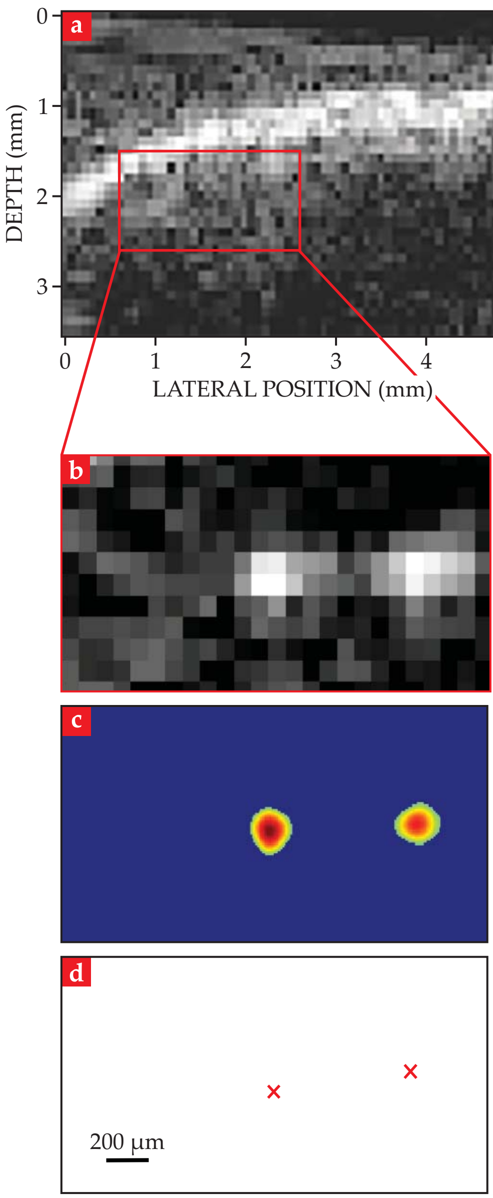


**FIGURE 2. RESOLUTION, POINT BY POINT.** Even when injected with inert gas microbubbles to enhance acoustic contrast, the blood vessels in a rat's brain are not visible in a conventional ultrasound image **(a)**; the prominent white streak is the skull. But the difference between two rapidly acquired frames **(b)** shows a sparse set of diffuse blurs, each arising from a bubble that moved or disintegrated. Fitting the blurs to two-dimensional Gaussian profiles **(c)** allows the bubbles to be located precisely **(d)**. (Adapted from ref. 1.)

even more powerful. "It is pretty exciting technology," says Dayton, "and very relevant to our work."

To obtain the crisp image in figure 1, Tanter and colleagues had to thin down the rat's skull from 700 μm to 100 μm; images taken through an intact skull still had superresolution features, but they were significantly less detailed in deep regions. That's because bone is a strong scatterer of ultrasound. Thinning a human skull—normally 7 mm thick—is clearly out of the question. Still, Tanter notes that other tumor-prone organs, such as the liver and breast, are not surrounded by bone, so they could be more easily imaged. And he plans to try imaging through an intact human skull using a longer ultrasound wavelength.

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## References


1. C. Errico et al., *Nature* **527**, 499 (2015).
2. E. Betzig et al., *Science* **313**, 1642 (2006); M. J. Rust, M. Bates, X. Zhuang, *Nat. Meth.* **3**, 793 (2006); S. T. Hess, T. P. Girirajan, M. D. Mason, *Biophys. J.* **91**, 4258 (2006).
3. O. M. Viessmann et al., *Phys. Med. Biol.* **58**, 6447 (2013); K. Christensen-Jeffries et al., *IEEE Trans. Med. Imag.* **34**, 433 (2015).
4. R. C. Gessner, S. R. Aylward, P. A. Dayton, *Radiology* **264**, 733 (2012); S. E. Shelton et al., *Ultrasound Med. Biol.* **41**, 1896 (2015).


# A quantum cascade laser gets a geometric makeover

A change in the cross section and orientation of the laser's optical cavity collimates an otherwise highly divergent terahertz-frequency beam.

Achieving a high-quality beam—one whose profile is near Gaussian and as narrow as diffraction allows—can be a challenge in semiconductor laser design. That's especially true in the few-terahertz frequency regime, an elusive part of the electromagnetic spectrum just below the reach of most optical technologies and just above the reach of electronics.

No natural material has an open bandgap small enough to emit coherent photons below tens of terahertz. The quantum cascade laser (QCL), one of the few devices that can operate at a few terahertz, relies on stacked, two-dimensional layers of semiconductors, which create quantum wells that act as gain material. The spatial confinement of electrons in the wells splits the conduction band into discrete subbands, separated by energies as small as a few meV, depending on the semiconductor-layer thickness (1 THz corresponds to about 4 meV).

Transitions between the subbands are at the heart of the QCL (see the article by Federico Capasso, Claire Gmachl,