

Molecular Imaging Advance Watches Tumors Grow, Shrink

HAVING USED bioluminescence tomography (BLT) to identify adrenal tumors in living mice, researchers at Virginia Tech said they have brought BLT imaging a step closer to potential use in preclinical drug and therapy trials.

Ge Wang, Ph.D., is an endowed engineering professor and director of the newly established Biomedical Imaging Division at the Virginia Tech-Wake Forest University School of Biomedical Engineering & Sciences in Blacksburg, Va. Dr. Wang was instrumental in inventing the BLT process in 2002 while on faculty at the University of Iowa in Iowa City.

BLT is a form of optical molecular imaging in which a naturally occurring enzyme, luciferase, is used to tag specific cells in small animals such as mice. The organic light from the enzyme—the same light that causes fireflies to glow—can then be measured from surface readings of the animal in order to achieve in vivo mapping.

Michael Henry, Ph.D., an associate professor in the Department of Molecular Physiology and Biophysics at the University of Iowa, who worked with Dr. Wang on past projects, explained the process. “The gene that makes the enzyme is cloned and put into cancer cells or microbial cells, which are then injected into animals,” said Dr. Henry. “The animal then expresses that gene in some tissue or in some particular cell. To visualize those cells, you give the animal an injection of luciferin, a substrate of luciferase.”



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The appeal of this approach is that it allows researchers to monitor tumor growth in a living animal, said Dr. Henry. “You can watch the tumor grow, and if you put the animal on an experi-

mental therapy, you can watch whether the tumor responds by monitoring the change in light output from that particular tumor,” he said.

The emitted bioluminescent light is captured by a cooled charge-coupled device (CCD)-based camera. When mapping gene expressions, BLT imaging is more sensitive and specific than standard X-ray and MR imaging techniques, researchers said.

In addition to monitoring tumors, the nature of BLT imaging also lends itself to measuring specific cell behav-

ior. “People often want to study how various signaling processes work within cells,” noted Dr. Henry. “You can set up the system so that the luciferase only gets turned on when a particular pathway is activated. Therefore, you know that the particular signaling event is happening in that cell.”

Said Dr. Wang: “The introduction of BLT relative to planar bioluminescent imaging can be compared to the development of computed tomography based on radiography. The advantage of BLT is that you can not only target specific cell structures or cell behaviors but also localize and quantify them within a living mouse volumetrically and dynamically.”

Diffusion is the Catch

The catch to using bioluminescence for imaging, the researchers said, is that as the light signal is emitted from the animal’s internal organs, it is instantly diffused by the living tissue around it,

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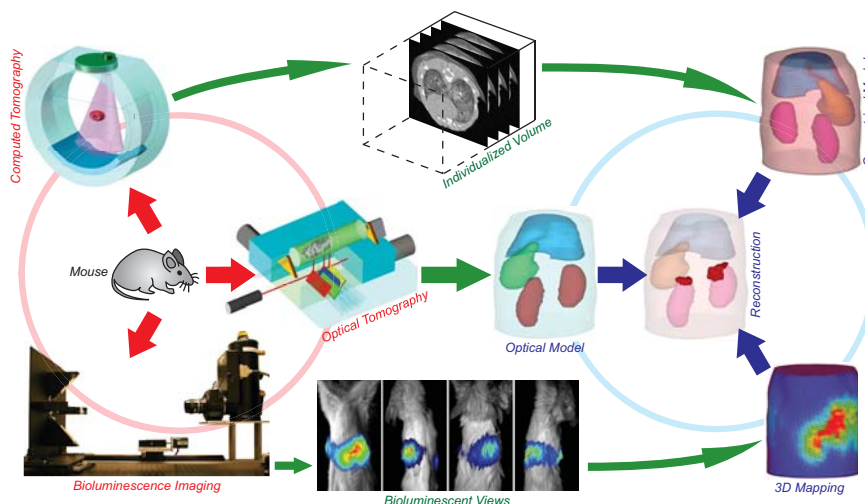
making it difficult to derive useful information. Noted Dr. Wang: “You have this beautiful bioluminescent light coming out from around the mouse, but the question is, what is the source? Where is the underlying bioluminescence? How strong is it? Where is the tumor? How many cancer cells are there?”

“When you see the signal coming from the surface of the animal, you can’t tell exactly where inside the animal the source of that signal is, and you can’t tell other things about it, like how large or intense the source is,” added Dr. Henry. “The signal is significantly impacted by its interaction with tissues, so it gets scattered and absorbed by the various tissues that it passes through. You can’t tell whether the signal is emanating from a very weak source that’s very near the surface of the animal, or a very strong source that is much deeper in tissues.”

Dr. Wang said he and his collaborators were able to render BLT imaging practical by developing a series of computer algorithms that work from 3D models of mouse anatomy and its optical properties. “The light diffuses in a zig-zag pattern, and the algorithms help decode that pattern,” he said. As a result, Dr. Wang and his team were able to successfully use BLT to find and measure the bioluminescent sources inside mice and therefore define internal tumors.

Two problems still make clinical use of BLT imaging a relatively distant goal, the researchers said. Noted Dr. Henry, “One problem with applying this technology clinically is that humans don’t make luciferase—in order to use BLT, you would have to introduce the enzyme into the human in some way, and that is complicated.” He added that while the depth through which the light diffuses in a mouse is small, the attenuation of light through much deeper tissues—like those of humans—is a significant barrier to extracting any signal or any information.

“For those two reasons—that you need to introduce the luciferase gene in



Flowchart for Bioluminescence Tomography.

A bioluminescent source active mouse is imaged in a bioluminescent mode to capture bioluminescent views around the mouse, in a tomographic mode (CT or MR) to reconstruct an anatomical volume and in an optical mode to estimate optical parameters keyed to each structural component. A linear imaging model is then built between the bioluminescent measurement and source distribution and inverted for 3D localization and quantitative analysis of the underlying molecular/cellular activities.

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some way and the depth problem—there is probably no immediate application in patients,” said Dr. Henry.

Critical Role to Play in Drug Development

Both Drs. Wang and Henry, however, underscored the immediate use of BLT imaging in nonhumans. “Right now bioluminescence tomography is very important for preclinical study using animal models,” said Dr. Wang. “It’s a tool that can be useful for small animal studies, especially when testing drugs or gene therapy.”

Added Dr. Henry: “A lot of drug development involves creating drugs to block or enhance the function of certain signaling pathways. With this system, luciferase is your reporter on whether or not that particular pathway is active and whether or not it responds to the drug you’re developing to block or enhance one of those pathways.”

Dr. Wang said he is also enthusiastic about future research into using temperature modulation techniques to improve BLT imaging. The emission spectra of various bioluminescent

probes are temperature dependent, so manipulating the temperature of the mouse’s body, using techniques similar to ultrasound thermal therapy, can change the spectrum of the BLT light emissions. “This is something we are actively working on so we can achieve better results,” said Dr. Wang.

He also expressed a strong interest in finding more collaborators and partners who need BLT imaging to answer their important biomedical questions. “With an increasing number of successful pre-clinical applications of BLT, we hope we will demonstrate that this technology is critically useful and becoming more and more popular,” he said. □

Learn More

■ More information on the Bioluminescence Tomography Laboratory at the Virginia Tech-Wake Forest University School of Biomedical Engineering & Sciences can be found at www.imaging.sbes.vt.edu/bltlab/bltlab.html. Researchers interested in collaborating with Dr. Wang on uses for BLT imaging can contact him wangg@vt.edu.