Researchers improve optical tissue clearing method to diagnose cancer

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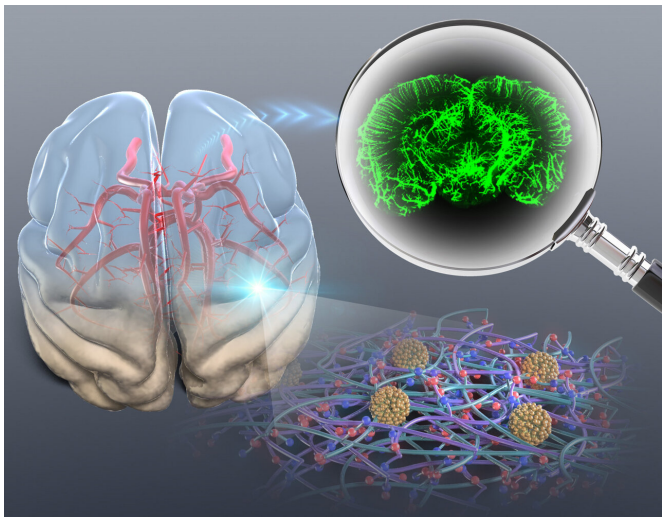

Optical clearing process of a brain tissue to visualize the 3D vascular network in fluorescence imaging. Zwitterionic polymer hydrogels including polymer hydrogels that mimic fatty molecules on the tissue promoted the optical tissue clearing process by discharging fats. Credit: ©Chie Kojima, Takayuki Koda, Tetsuro Nariai, Junji Ichihara, Kikuya Sugiura, and Akikazu Matsumoto, Osaka Prefecture University

When it comes to cancer, clarity is key. The ability to visualize cancerous tumors and metastatic tissue three-dimensionally (3D) can help clinicians diagnose the precise type and stage of cancer, while also informing the best treatment methods. To obtain an even clearer tissue for imaging, a research team based in Japan has tested the effectiveness of specialized hydrogels. Acting as a 3D molecular network, these hydrogels can rapidly remove fats from tissues, which are a factor in tissue opacification, without losing their structure. The material is used in several biomedical devices, including contact lenses.

They published their results online on June 21 in *Macromolecular Bioscience* with the print edition issued on Sep. 16.

"Since 1981, the leading cause of death in Japan has been cancer," said first author Chie Kojima, associate professor in the Department of Applied Chemistry in the Graduate School of Engineering at Osaka Prefecture University. "We need new treatment methods and diagnostic techniques. 3D fluorescence imaging is one such approach that could prove indispensable for understanding multicellular systems on the scale of an organ, as it can give us more information than traditional 2D imaging. This could be useful for personalized medicine in diagnosis, as well as elucidating biological phenomena."

This type of imaging involves tagging certain molecular machines, such as proteins, so they fluoresce with different colors depending on what they are. The glowing signals can be viewed in a variety of samples, from whole organisms down to the cellular level. Most tissues are opaque, though, blocking the ability to see these signals. In 2D imaging, the samples are sliced thinly, which makes the signals easy to see but removes the ability to visualize the full system in 3D.

Previously, researchers have used an approach known as CLARITY, in which the tissues are embedded in polyacrylamide hydrogels. The fats are removed from the tissues and the refractive index of the media is adjusted. The tagged glowing signals can be visualized in 3D, but it takes a month for the cancerous tissue to clear—far too long for a patient waiting for a diagnosis, according to Kojima. In that time, the tumor would have likely spread.

"The optical clearing process time in the CLARITY method needs to be shortened for practical applications," Kojima said.

To cut this time, the researchers used zwitterionic hydrogels, which are balanced in their charged

molecules and hold the structure of tissue samples. (AMED)

Of several zwitterionic hydrogel combinations, the team found that polymer hydrogels that mimic fatty molecules on the tissue appear to optically clear tumor tissues the quickest. According to Kojima, the hydrogels are highly osmotic, which may help pull other fatty acids from the tissue.

"Blood vascular networks in murine brain tissues, as well as metastatic tumor tissues could be visualized in 3D using our system," Kojima said.

And they could visualize the tumor tissues quicker than in their previous attempts: What previously took a month could be achieved in a week with the improved approach.

The researchers are continuing to explore the technique and how to apply it for diagnosing cancer in humans.

"We are attempting to apply our system for pathological diagnostics," Kojima said. "We expect that it will be possible to diagnose a whole biopsy sample—instead of thin slices—which could prevent the oversight of small cancers.

Other contributors include Takayuki Koda and Akikazu Matsumoto, Department of Applied Chemistry, Graduate School of Engineering, Osaka Prefecture University; Tetsuro Nariai, and Junji Ichihara, Bioscience Research Laboratory, Sumitomo Chemical Company, Ltd.; and Kikuya Sugiura, Department of Advanced Pathobiology, Graduate School of Life and Environmental Sciences, Osaka Prefecture University.

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