KYUSHU UNIVERSITY

Public Relations Initiative, Kyushu University
744 Motooka Nishi-ku Fukuoka, 819-0395
TEL: +81-92-802-2443 FAX: +81-92-802-2139
E-MAIL: koho@jimu.kyushu-u.ac.jp
URL: https://www.kyushu-u.ac.jp

PRESS RELEASE (2026/03/12)

A new reagent makes living brains transparent for deeper, non-invasive imaging

Researchers at Kyushu University have developed a new tissue-clearing reagent, SeeDB-Live, which enables repeated, reversible, and real-time imaging of the living brain at greater depth and clarity

Fukuoka, Japan—Making a living brain transparent and watching its neurons fire without disturbing their function—sounds like science fiction, doesn't it? Yet the solution may already exist within our own bodies.

In a paper published in *Nature Methods* on March 12, a research team led by Kyushu University introduces a new reagent called SeeDB-Live. It uses albumin—a common protein in blood serum—to clear tissue while preserving cellular function. The technique allows scientists to see deeper, brighter structures in both brain slices in a dish and living mice, reaching neural activity that was previously out of sight.

"This is the first time tissue clearing has been achieved without altering its biology," says Takeshi Imai, Professor at Kyushu University's Faculty of Medical Sciences and the study's senior author.

"SeeDB-Live can pave the way for deep-tissue live imaging, both *ex vivo* and *in vivo*," adds the study's first author, Assistant Professor Shigenori Inagaki of the same faculty.

How to see deeper into the living brain?

Complex functions like memory and thought arise from real-time communication between cells deep in the brain. Although slices preserve some activity, understanding normal brain dynamics requires imaging the living brain.

Making the opaque brain transparent is one solution, and it begins with optics.

Consider glass marbles: clearly visible in air but nearly disappear in oil. This is because light refracts and scatters when passing between materials with different refractive indices, and brain tissue behaves the same way. Lipids and other cellular components create tiny mismatches, scattering light, hiding deeper structures. Reduce them, and light travels uniformly.

Through systematic experiments, Imai's team found that living cells become most transparent when the refractive index of the extracellular solution is adjusted to 1.36–1.37.

With a precise target in hand, the team needed a non-toxic way to reach it while maintaining osmotic balance, so that cells neither swell nor shrink. They previously tried natural substances such as sugar, but these required high concentrations that increased osmotic pressure and dehydrated cells.

As osmotic pressure depends on the number of molecules, the team turned to large spherical polymers. Their greater size means fewer are required to raise the refractive index, which adjusts optical performance without overwhelming the cells. However, despite screening nearly 100 compounds, the answer refused to come.

A blood protein is the surprising key to brain transparency

The turning point came unexpectedly.

Late one night, Inagaki returned to a simple idea: proteins are polymers. He grabbed a bottle of bovine serum albumin (BSA), a common blood-derived laboratory reagent, which, to his surprise, showed the lowest osmotic pressure at the desired refractive index.

"I tested it three or four times before I believed it," Inagaki recalled. Alone in the lab that night, he let out a shout of excitement. "Of all things, we never expected it would come down to this."

By adding albumin to the culture medium to match the refractive index inside cells, the team developed a live-tissue clearing solution, which they named SeeDB-Live.

"During the development of SeeDB-Live, we found that neurons are extremely sensitive to ion concentrations, and it took us enormous effort to get the formulation right. Thanks to that fortunate night alone in the lab, I helped myself to an expensive, high-purity BSA I wouldn't normally dare use," Inagaki adds with a laugh.

SeeDB-Live renders mouse brain slices transparent within one hour of immersion. When combined with a calcium indicator, the normal neuronal firing deep inside the tissue was illuminated in the transparent brain slice. When applied to living mouse brains, fluorescence signals from deep neurons became three times brighter.

This opens up clear views of layer 5 of the cerebral cortex, where richly branched neurons help reveal how the brain processes information and translates neural activity into action. Before SeeDB-Live, crisp images at this depth were difficult to obtain with conventional strategies.

Moreover, as the extracellular fluid washes out SeeDB-Live within hours, the tissue transparency returns to its original state. Because the method causes no permanent changes, the same mouse can be imaged repeatedly to track brain activity over time.

"Albumin is abundant in blood and highly soluble, which makes it well-suited for clearing," notes Imai. "It was an accidental discovery, but looking back, it feels almost natural. What evolution has shaped over millions of years is truly impressive."

A decade after saying "impossible"

SeeDB-Live demonstrates the first non-invasive optical clearing that greatly increases imaging depth and enables observation of tissue-wide dynamics.

Researchers expect it to enhance deep fluorescence imaging for understanding brain integrative functions. It may also help evaluate 3D tissues and brain organoids for drug discovery research.

The team notes that although SeeDB-Live works well for brain tissue, biological barriers limit delivery to other organs, and accessing the brain still requires a surgical window that can cause stress and reduce efficiency.

"I feel we have not yet fully materialized its potential," Inagaki says, adding that future efforts will focus on less invasive delivery methods to improve penetration for deeper imaging and better functional analysis of brain activity.

For Imai, the achievement marks the culmination of more than a decade of work. After developing SeeDB in 2013 and SeeDB2 in 2016 for fixed tissue, he was repeatedly asked whether live tissue clearing was possible.

"That question came to me about a hundred times, and each time I answered 'impossible,'" Imai reflects. "But ten years later, here we are. When something seems unachievable, if you keep thinking about it, you may eventually find a way."

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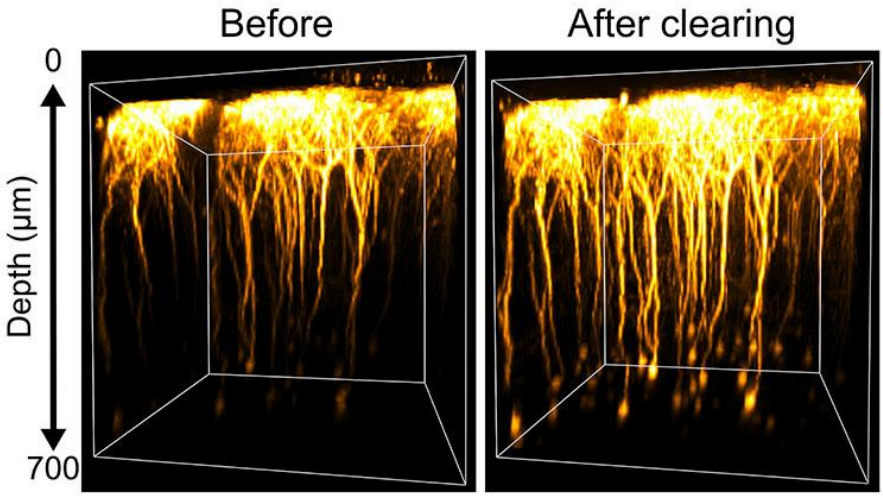
For more information about this research, see " Isotonic and minimally invasive optical clearing media for live cell imaging ex vivo and in vivo," Shigenori Inagaki, Nao Nakagawa-Tamagawa, Nathan Zechen Huynh, Yuki Kambe, Rei Yagasaki, Satoshi Manita, Satoshi Fujimoto, Takahiro Noda, Misato Mori, Aki Teranishi, Hikari Takeshima, Koki Ishikawa, Yuki Naitou, Tatsushi Yokoyama, Masayuki Sakamoto, Katsuhiko Hayashi, Kazuo Kitamura, Yoshiaki Tagawa, Satoru Okuda, Tatsuo K. Sato, Takeshi Imai, *Nature Methods,* https://doi.org/10.1038/s41592-026-03023-y

About Kyushu University

Founded in 1911, Kyushu University is one of Japan's leading research-oriented institutions of higher education, consistently ranking as one of the top ten Japanese universities in the Times Higher Education World University Rankings and the QS World Rankings. Located in Fukuoka, on the island of Kyushu—the most southwestern of Japan's four main islands—Kyushu U sits in a coastal metropolis frequently ranked among the world's most livable cities and historically known as Japan's gateway to Asia. Its multiple campuses are home to around 19,000 students and 8,000 faculty and staff. Through its VISION 2030, Kyushu U will "drive social change with integrative knowledge." By fusing the spectrum of knowledge, from the humanities and arts to engineering and medical sciences, Kyushu U will strengthen its research in the key areas of decarbonization, medicine and health, and environment and food, to tackle society's most pressing issues.



Three-dimensional fluorescence imaging of neurons in Layer 5 of the cerebral cortex, captured by two-photon microscopy in mice expressing a fluorescent protein (Thy1-EYFP-H). SeeDB-Live clearing improved fluorescence brightness in deep brain regions compared to uncleared tissue.

[Contact]
Takeshi Imai, Professor
Faculty of Medical Science
Tel: +81-92-642-6090
E-mail: imai.takeshi.457@m.kyushu-u.ac.jp