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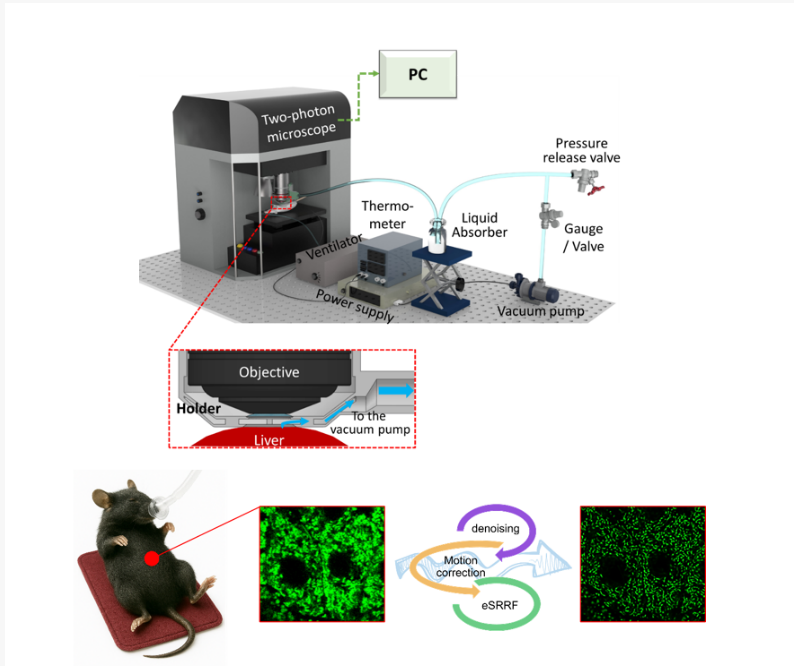
Multiphoton intravital microscopy in small animals of long-term mitochondrial dynamics

To accomplish this, a conventional two-photon microscope was equipped with a 3D-printed holders, which stabilize the tissue surface.



Schematic of suction-based stabilization for super-resolution radial fluctuations (SRRF) intravital imaging and its results.

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Studying how cells work inside a living body is one of the most powerful ways to understand health and disease. However, looking deep inside live tissue is extremely challenging, especially when trying to see very small structures like mitochondria—the tiny engines inside cells that produce energy and help regulate many important biological functions. These structures are constantly moving and changing, so scientists need imaging tools that can capture them in action, clearly and without harming the animal.

In this study, researcher developed a new imaging approach that combines two powerful techniques to solve this problem. First, a special type of microscope called a two-photon microscope is used, which can look deep into live tissues with minimal damage. To improve how steady the tissue remains during imaging, they designed a custom-made, 3D-printed holder that gently uses suction to hold the tissue in place. This suction-based stabilization helps keep the tissue still within the camera's focus, which is essential when trying to capture high-resolution images of tiny moving structures.

Next, the images are improved further using advanced computer algorithms. These included techniques to reduce "noise" (unwanted blurry spots in images), correct for tiny shifts or drifts in the picture, and enhance image sharpness using a method called Super-Resolution Radial Fluctuations (SRRF). Together, these improvements doubled the resolution, allowing the researchers to clearly see details smaller than 250 nanometers, far beyond what a typical microscope can do.

To demonstrate the power of this approach, they used a genetically modified mouse called the Mito-Dendra2 model, where mitochondria glow green under the microscope. With this model, the team was able to watch, in real time, how mitochondria split, merge, move, and respond to different health conditions. For example, it is observed how mitochondria behave in a model of alcohol-induced liver disease, and how a natural compound called berberine can help restore mitochondrial health during recovery. These insights would be nearly impossible to gain without being able to see the mitochondria directly inside a living animal.

This work is important because it allows scientists to study the smallest building blocks of life in their most natural environment, inside living and functioning tissue. The combination of gentle physical stabilization, powerful microscopy, and advanced image processing offers a new standard for intravital imaging within live organisms. It opens the door for new discoveries in how cells respond to stress, how diseases develop, and how treatments work at the organelle level.
In short, this breakthrough provides researchers with a valuable tool to explore the hidden world inside living tissues, making it easier to understand life at the cellular level and develop better ways to diagnose and treat disease.

The research group of Prof. Jun Ki Kim from Asan Medical Center, the largest hospital in Korea, and University of Ulsan, College of Medicine, introduces a groundbreaking technology that provides super-resolution imaging inside the cells of living animals.
Located at the intersection of engineering, medicine, and optics, the work conducted in this optics laboratory within a biomedical engineering department and affiliated medical center plays a vital role in shaping the future of healthcare and scientific discovery. This interdisciplinary environment brings together physicists, engineers, biologists, and clinicians to address some of the most pressing challenges in modern medicine—diagnosing diseases earlier, treating them more precisely, and understanding human biology at a deeper level.

The importance of this work lies in its focus on translating light-based technologies—such as advanced imaging systems, diagnostic tools, and therapeutic devices, into real-world clinical solutions. In hospitals and clinics, doctors often rely on indirect indicators of disease, like blood tests or tissue biopsies, which can be invasive or limited in detail. Optical technologies offer a different approach: they provide real-time, non-invasive insight into the human body, revealing structures and molecular changes that are invisible to the naked eye.

In this lab, researchers develop tools that can image cells inside a living body, detect early signs of disease, or guide surgeons during operations with light-based visualization techniques. Working hand-in-hand with medical professionals, engineers transform scientific principles into devices that are practical, safe, and effective for patient care.

Moreover, the lab’s presence within a medical center fosters rapid collaboration and translation. Research does not remain confined to the lab bench; instead, it moves efficiently toward patient trials and clinical use. This dynamic setting ensures that innovations are not only technically advanced but also medically relevant.
In the broader context, this work contributes to a global effort to make medicine more personalized, less invasive, and more data-driven and ultimately improving outcomes and quality of life for patients around the world.

The research group specializes in the development and application of advanced bio-optical imaging systems, focusing on cutting-edge technologies that enable the visualization and analysis of biological tissues at cellular and subcellular levels. Their work in in vivo microscopy allows for high-resolution imaging of living tissues, providing critical insights into dynamic biological processes in real-time. This technique is particularly valuable in studying tissue structures and identifying disease markers, offering significant potential for improving diagnostics and therapeutic interventions.

Another major area of focus for the group is medical device development, where they design and create innovative tools that integrate optical and imaging technologies for clinical applications. Their efforts are aimed at enhancing the precision, reliability, and accessibility of diagnostic devices, ensuring that they meet the needs of both healthcare providers and patients. In parallel, the group is heavily involved in the development of optical probes that can be used for in-depth, non-invasive tissue analysis. These probes are designed to interact with biological tissues at the molecular level, enabling detailed, real-time assessments of cellular processes.

A key area of the group's research is diagnosis and therapy, particularly leveraging the power of Raman spectroscopy. By utilizing the unique vibrational properties of molecules, Raman spectroscopy provides a powerful tool for identifying chemical signatures associated with various diseases, enabling early detection and precise monitoring of therapeutic responses. This approach holds great promise for improving disease diagnosis, particularly in oncology and other fields where early intervention is critical.
Through their multidisciplinary work, the research group is advancing the frontiers of medical diagnostics, offering new solutions for in vivo imaging, disease detection, and personalized therapy.

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