FRANCE-BIOIMAGING

Sino-France
2nd Joint Meeting on BioImaging

2024.4.8-10

Zhongguanyuan Global Village, PKU

Hosts Peking University
Institute of Biophysics, Chinese Academy of Sciences
Subcellular Structure and Function subgroup, Biophysical Society of China
Academic Committee of Modern Biophysical Technology and Methods

Co-organizers France-BioImaging

Undertaker National Biomedical Imaging Center, Peking University

Imaging technology, as one of the most crucial research tools in biomedicine, has become a core force driving the development of basic life science research and clinical medical studies. On the occasion of the 60th anniversary of the establishment of diplomatic relations between China and France, to further promote the innovation and development of biomedical imaging technology, strengthen international academic exchanges and cooperation, and leverage the strategic and comprehensive benefits of the national major scientific and technological infrastructure "Multimodal Cross-Scale Biomedical Imaging Facility". We are scheduled to host the "Sino-France 2nd Joint Meeting on BioImaging" from April 8 to 10, 2024, at Peking University. This forum is dedicated to assembling preeminent scientists, researchers, and technical developers from the bioimaging sector to explore the future trajectories of bioimaging technology through the dissemination of the latest research findings, technological advancements, and application cases, and to deliberate on China's participation into Global BioImaging. The forum is hosted by Peking University and jointly organized by France BioImaging.

National Biomedical Imaging Center，Peking University
2024.3

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2024.4.10 PM

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Speakers

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Tao Xu **Institute of Biophysics, CAS**

Research Focus

Super-resolution correlative light-electron microscopy.

Brief Biography

Tao Xu, Member of Chinese Academy of Sciences, Principal Investigator of Guangzhou National Laboratory and Institute of Biophysics, CAS. He obtained his Ph.D. from Huazhong University of Science and Technology in 1996. From 1996 to 2000, he undertook postdoctoral work first with 1991 Nobel prize winner-Erwin Neher at the Max-Planck Institute for Biophysical Chemistry and then with Bertil Hille at the University of Washington. Since 2000, he built his laboratory at Huazhong University of Science and Technology. From 2003, he became a PI and successively served as deputy director, director of the Institute of Biophysics, Chinese Academy of Sciences. He participated in the setting up of Guangzhou Laboratory in 2021 and serve as the deputy director now. His research focuses on the development of super-resolution correlative light-electron microscopy.

Report

Cryogenic super-resolution correlative light and electron microscopy on the frontier of subcellular imaging

Over the past decade, there has been extraordinary advancement in the field of cryogenic superresolution correlative light and electron microscopy. In this presentation, I am excited to showcase cryogenic imaging techniques that deliver outstanding localization precision performances. We have utilized cryogenic super-resolution correlative light and electron microscopy (csCLEM) to accurately delineate the spatial interplay between proteins and their inherent cellular architectures. Our findings reveal that several fluorescent proteins (FPs) exhibit photoswitchable properties and emit significantly more photons under cryogenic conditions. This enhancement leads to a localization precision that rivals that of conventional super-resolution imaging conducted at room temperature. To preserve the near-native state of specimens with enhanced fluorescence retention, we prepared vitrified samples using high-pressure freezing followed by cryo-sectioning. Furthermore, we extended the application of csCLEM to mammalian cells. This allowed us to observe a precise correlation between a mitochondrial protein and the mitochondrial outer membrane with nanometer resolution in three dimensions. Additionally, we developed a cryogenic correlated light, ion, and electron microscopy (cryo-CLIEM) platform capable of preparing cryo-lamellae guided by three-dimensional confocal imaging. Through the successful preparation of cryo-lamellae containing either a single centriole or contact sites between subcellular organelles, we demonstrate the broad applicability of this technique. This innovation paves the way for further applications of cryo-electron tomography (cryo-ET), promising to revolutionize our understanding of cellular structures and functions.

Yves Mely University of Strasbourg

Research Focus

Synergistic development in fluorescence probes and advanced fluorescence microscopy techniques

Brief Biography

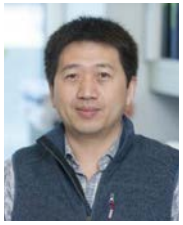
Yves Mély is professor of biophysics at the University of Strasbourg. Pharmacist and holder of a PhD in biophysics from the University in Strasbourg (1988). He was appointed lecturer in 1989, then full professor in 1998. He heads the team "Biophotonics of molecular and cellular interactions" since 1999. He headed the Bioimaging and Pathologies laboratory (UMR 7021) from 2009 to 2023. He heads the Alsace node of France BioImaging (FBI) since 2023 and is deputy director of FBI, since 2024. His research activity can be divided into two research axes. The first relates to the development of fluorescence techniques (quantitative and high resolution microscopy) and innovative fluorescence probes for membranes, proteins and nucleic acids. The second axis deals with the application of these techniques and probes to characterize the properties and functions of viral and epigenetic proteins, in order to discover new therapeutic avenues. He has published more than 360 scientific articles. He is the chair of the international conference series "Methods and Applications in Fluorescence" since 2011. He also co-founded the journal "Methods and Applications in Fluorescence" and is one of its editors since 2012. He has obtained an Adrerus thesis prize in 1990 and a von Humboldt fellowship for a sabbatical in 1997. He has been invited as "Senior Scientist" at Riken, Tokyo, in 2015. He received the "Gregorio Weber Award for Excellence in Fluorescence Theory and Applications" in 2017. He was appointed Doctor Honoris Causa at Taras Shevchenko University in Kiev (2011) and senior member of the Institut Universitaire de France in 2018.

Report

Joint development of fluorescent probes and microscopy: a win-win strategy

The development of new advanced fluorescence microscopy techniques requires close synergy between instrumental developments and those in fluorescent and luminescent probes. This interdependence between the two fields quickly became obvious, since each new modality in imaging could only be fully exploited if there were probes perfectly adapted to this modality. The reverse approach has also given interesting results, since several probes by their particular photophysics have given rise to developments in high-performance imaging, taking advantage of the full potential of these probes. I am going through examples chosen in particular from developments made in the laboratory, to illustrate this synergy and the applications that result from it. These cross developments and their applications will be illustrated in two-photon FLIM and FCS microscopy, in super-resolution microscopy and anti-Stokes microscopy.

Songhai Shi **Tsinghua University University**



Research Focus

Brain development and function

Brief Biography

Song-Hai Shi, Ph.D., is currently the Dean of the School of Life Sciences at Tsinghua University, and the Director of the IDG/McGovern Institute for Brain Research at Tsinghua University.

Dr. Shi has focused his research on identifying the common commodities of brain circuits at both the structural and functional levels, and linking them with animal behaviors, with the ultimate goal of arriving at a circuit- and system-level understanding of brain operation and function under normal and disease conditions. He has published numerous peer-reviewed articles in journals including Nature, Science, Cell, Nature Neuroscience and Neuron.

His expertise and achievement have been recognized by a number of awards, including the Amersham Biosciences and Science Grand Prize for Young Life Scientists, the Blavatnik Award for Young Scientists, the Howard Hughes Medical Institute (HHMI) Faculty Scholar Award, the New Cornerstone Investigator, and the Beijing Scholar.

Report

Imaging neocortical development and function

Development of the neocortex is considered to be the crowning achievement of evolution and likely holds the key to the superior mental prowess of humans. The ability of the neocortex to command all higher-order brain functions depends on its intricate circuitry comprised of a vast number of neurons that are incredibly diverse. Despite the advances in the understanding of the initial specification and the general histology and information flow in the neocortex, the principles governing the precise and effective assembly and operation of neocortical circuits remain largely elusive. Our work focuses on identifying the common commodities of neocortical circuits at both the structural and functional levels, and linking them with animal behaviors under normal and disease conditions. In this presentation, I will discuss our ongoing efforts on applying the latest imaging techniques on underlying the development and function of the neocortex.

Sylvain Gigan Sorbonne University

Research Focus

Sylvain Gigan is Professor of Physics at Sorbonne Université in Paris, and group leader in Laboratoire Kastler-Brossel, at Ecole Normale Supérieure (ENS, Paris). His research interests focuses on light propagation in complex media, and range from fundamental investigations, biomedical imaging, computational imaging, signal processing, to quantum information.

Brief Biography

After graduating from Ecole Polytechnique (Palaiseau France) in 2000, and a Master Specialization in Optics from University Paris XI (Orsay, France), he obtained a PhD in Physics 2004 from University Pierre and Marie Curie (Paris, France) in quantum and non-linear Optics. From 2004 to 2007, he was a postdoctoral researcher in Vienna University (Austria), working on quantum optomechanics, in the group of Markus Aspelmeyer and Anton Zeilinger. from 2007 to 2014, he was at ESPCI ParisTech in Paris, as Associate Professor, and started working on optical imaging in complex media and wavefront shaping techniques, at the Langevin Institute. He was appointed full professor at Sorbonne Université in 2014 and joined the LKB as group leader. He was awarded the Fabry de Gramont Prize of the French Optical Society in 2016, The Joseph Fourier ATOS prize in 2018, the Jean Jerphagnon Prize in 2019. He is the recipient of two ERC grants in 2011 and 2017. He is Optica Fellow. He was a member of the Institut Universitaire de France (2016-2021). He is Editor of Optics Communications, Optica, Intelligent Computing & eLight.

Report

Deep fluorescence imaging in complex media, wavefront shaping meets computational imaging

Wavefront shaping allows manipulation of light in complex media, for focusing and imaging, at depth where no ballistic light is present anymore. It however conventionally requires coherence, in order to manipulate speckle patterns. Fluorescence remains very challenging, both as a contrast mechanism for deep-imaging, and as a guide-star. I will show how the same concepts allows to tackle fluorescence, and even image extended fluorescent objects. I will present results in functional and structural fluorescence imaging at depth, by leveraging both wavefront shaping but also modern computational imaging tools.

Zhixing Chen Peking University

Research Focus

Imaging Probes, Chemical Biology

Brief Biography

Dr. Zhixing Chen is an assistant professor at College of Future Technology, Peking University. He received BS from Tsinghua University (2008) and PhD from Columbia University (2014). He has additional training at Stanford University (Postdoc 2016-2018) and Columbia University (Postdoc 2015). The Chen lab integrate chemistry and biochemistry to create new imaging tools with high brightness, specificity, and biocompatibility to promote advanced bioimaging technologies, aiming to enable new approaches in cell biology, metabolism, and biophysics.

Report

Gentle dyes for 4D fluorescence imaging

4D fluorescence imaging is jointly enabled by new microscopic methods and novel fluorescent markers that are bright, photostable, and biocompatible. Notably, phototoxicity has become a prevailing issue in the super-resolution era when boosted illumination is applied, compromising the physiological relevance of the recorded data. We advocate leveraging chemical approaches to tackle phototoxicity. By exploiting chemical motifs such as triplet state quenchers and biocompatible auxiliaries, we systematically upgrade the commonly used fluorescent markers toward alleviated phototoxicity. These gentle dyes can be directed to various cellular targets spanning mitochondria, DNA, cytoskeleton, insulin granule, and specific proteins, enabling time-lapse super-resolution imaging with minimal photodamage.

Emmanuel Beaurepaire **Institut Polytechnique de ParisParis**

Research Focus

Multiphoton microscopy of tissues

Brief Biography

Emmanuel Beaurepaire (EB) is a specialist of multiphoton microscopy of biological tissues. Trained in engineering physics and informatics in INP Grenoble (France), he worked two years as a visiting scientist at Cornell University (NY,USA), and prepared a PhD in optics at ESPCI in Paris (France). He was hired by the CNRS in 2001 and joined the laboratory for optics and biosciences at Ecole polytechnique (Palaiseau, France), where he initiated the advanced microscopy group (now 20 people). EB pioneered technological fields such as third-harmonic generation microscopy and multicolor 2-photon microscopy, and their applications to live embryo imaging and neurodevelopment.

Report

Color and multicontrast multiphoton imaging of developing tissues

Emmanuel Beaurepaire
Laboratory for Optics and Biosciences, Institut Polytechnique de Paris, Palaiseau, France
https://lob.ip-paris.fr/en

The study of brain tissue development requires imaging approaches that ideally provide micrometer resolution, millimeter imaging depth, and multi-contrast capability. Multiphoton microscopy is well established in neuroscience for its ability to provide high-contrast fluorescence imaging of neurons at depths of a few hundred microns. Over the past two decades, many techniques have been developed to extend the capabilities of two-photon microscopy, either in terms of depth, speed, or contrast modalities. In this talk, we will discuss some recent approaches for large-scale imaging of uncleared tissues, such as color serial two-photon imaging [1] and three-photon (3P) microscopy. We will also discuss label-free contrast modalities that can provide complementary information to fluorescence, such as third-harmonic generation (THG) and third-order sum frequency generation (TSFG) [2].

References:

[1] Abdeladim et al, Nat Commun (2019), https://doi.org/10.1038/s41467-019-09552-9.
[2] Ferrer Ortas et al, Light Sci App (2023), https://doi.org/10.1038/s41377-022-01064-4.

Ke Si **Zhejiang University**

Research Focus

Deep tissue imaging, microscopy, neurophotonics

Brief Biography

Dr. Ke Si is a professor in the School of Brain Science and Brain Medicine, Zhejiang University, He is also a joined professor in the College of Optical Science and Engineering, Zhejiang University. He received his Bachelor degree in Zhejiang University, and his PhD degree in NUS, respectively. Then he was trained as a Research Fellow in Singapore-MIT Alliance for Research and Technology (SMART), and later as a Research Associate at Janelia Farm Research Campus Howard Hughes Medical Institute (HHMI), respectively. Hsi main research interest is to understand and control the neuroscience circuit with custom-designed optical tools. 1) Non-invasive deep tissue optical imaging, 2) non-invasive deep tissue optogenetics with sub cellular resolution. His papers have been published on “Nature Photonics”, “PNAS”, “Molecular Psychiatry” and so on.

Report

Multi-scale mapping from mouse to human brain with integrative multimodal ultra-platform (iMap)

Light-sheet fluorescence microscopy (LSFM) provides high-throughput imaging at micrometer resolution. However, its versatility is limited to specific specimens, either small organs or thin tissue slices, making it a great challenge to be widely used in biological science. Here we develop an integrative multimodal ultra-platform, termed iMap, with angle switchable LSFM (MUSIA), hybrid three-dimensional deconvolution (Hy3D), custom-designed tissue preparation procedure and automatic reconstruction software (CoFFeR). With iMap, we can achieve high-throughput micrometer-resolution imaging for multi-scale samples from tissue slices to entire organs of mouse and human. For entire organs, we integrate the orthogonal-plane configuration of MUSIA with whole-organ clearing and Hy3D for one-shot holistic organ imaging. Then we first reveal the 3D fluorescence distribution of human retinal vasculature separated from an entire human eyeball (~7000 mm3 in volume). For tissue slices, we integrate the diagonal configuration of MUSIA with fast slice clearing (e.g. 2-minute FOCM) for rapid imaging. Further, with a custom-designed human brain tissue preparation procedure and CoFFeR, we successfully reconstruct a ~5-mm thick brain tissue from 14 consecutive human brain slices (400 μm in thickness) with micrometer-level 3D structural connectivity. Our results demonstrate that iMap is a powerful, yet accessible platform for high-throughput volumetric imaging of multi-scale samples in biological and clinical applications.

Valentin Nägerl University of Bordeaux

Research Focus

Super-resolution microscopy of brain microstructures

Brief Biography

Valentin Nägerl is a Professor of Neuroscience at the University of Bordeaux. His research is centered on the development and application of super-resolution microscopy to study the nanoscale mechanisms of synaptic plasticity and brain physiology using the mouse brain as a model system. In particular, his team explores the structure and function of the extracellular space of the brain.

After studies in physics and medicine in Göttingen, Germany, he got a PhD in neuroscience from UCLA with Istvan Mody and did a postdoc at the Max Planck Institute of Neurobiology with Tobias Bonhoeffer, spending research visits at the Max Planck Institute for Biophysical Chemistry with the Nobel laureate Stefan Hell. In 2009, he obtained the habilitation for neuroscience from the Technical University of Munich under the Brain Prize winner Arthur Konnerth, before setting up his own lab in Bordeaux. In 2017, he became senior member of the 'Institut Universitaire de France', and his work has been recognized by an 'Equipe FRM' award (2016), a Research Prize from the French Academy of Sciences (2018) and the Maxime Dahan Prize from the Curie Institute. He has received two HFSP program awards (2010 and 2020), an ERC synergy grant (2020) and a Discovery Award by the Wellcome Trust (2023). In 2021, he was promoted to the rank of 'Professeur classe exceptionelle'.

Report

STED-SUSHI imaging of dynamic brain microstructures in vivo

Progress in microscopy has a long history of triggering major advances in neuroscience. STED microscopy, famous for shattering the diffraction barrier of light microscopy, is a case in point. It gives access to anatomical designs and dynamics of nano-structures, which are impossible to resolve using conventional light microscopy, from the elaborate anatomy of neurons and glial cells, to the organelles and molecules inside of them. Brain cells such as neurons and astrocytes exhibit an extremely elaborate morphology, and their functional specializations like synapses and glial processes often fall below the resolution limit of conventional light microscopy. This is a huge obstacle for neurobiologists because the nanoarchitecture critically shapes fundamental functions like synaptic transmission and Ca2+ signaling. Overcoming this problem, STED offers the chance to visualize the structural and molecular organization of brain cells in a living and dynamic tissue context, unlike traditional methods like electron microscopy or atomic force microscopy.

In the first part of my talk, I will review our contributions to developing live-cell STED approaches and their application to interesting problems in cellular neurobiology concerning the structure and function of neuronal synapses, and their interactions with glial partners. I will then introduce the topic of the extracellular space (ECS), which is increasingly attracting the attention of neuroscientists after yielding to novel labeling and imaging approches. I will lay out the case for “shadow imaging” to visualize the convoluted physical structure of the ECS and, by implication, the dense and comprehensive microanatomical organisation of living brain tissue. I will show published and recent data, illustrating the potential of the inverse labeling strategy for unbiased imaging of brain microstructure and dynamics in a panoramic and multi-scale way.

Dinggang Shen ShanghaiTech University

Research Focus

Brief Biography

Dinggang Shen is a Professor and a Founding Dean with School of Biomedical Engineering, ShanghaiTech University, Shanghai, China, and also a Co-CEO of United Imaging Intelligence (UII), Shanghai. He is a Fellow of IEEE, AIMBE, IAPR, and MICCAI. He was also a recipient of the Distinguished Investigator Award from The Academy for Radiological & Biomedical Imaging Research, USA (2019). He was Jeffrey Houpt Distinguished Investigator and a Full Professor (Tenured) with The University of North Carolina at Chapel Hill (UNC-CH), Chapel Hill, NC, USA, directing The Center of Image Analysis and Informatics, The Image Display, Enhancement, and Analysis (IDEA) Lab, and The Medical Image Analysis Core. Before that, he was also a tenure-track assistant professor in the University of Pennsylvanian (UPenn), and an Instructor in the Johns Hopkins University. His research interests include medical image analysis, computer vision, and pattern recognition. He has published more than 1600 peer-reviewed papers in the international journals and conference proceedings, with H-index 140 and over 85K citations. He serves as an Editor-in-Chief for Frontiers in Radiology, as well as an editorial board member for eight international journals. Also, he has served in the Board of Directors for MICCAI Society in 2012-2015, and was General Chair for MICCAI 2019.

Report

Fast Development and Deployment of AI Techniques for Medical Imaging

I will introduce our developed full-stack, full-spectrum Artificial Intelligence (AI, or deep learning) techniques for whole clinical workflow, from data acquisition to disease detection, follow-up, diagnosis, therapy, and outcome prediction. In particular, I will demonstrate some innovative technical development and implementation in scanners and clinical workflows, i.e., serving for fast MR, low-dose CT/PET acquisition, and clinical diagnosis/therapy.

Sam Li **University of California, San Francisco**

Research Focus

Elucidating the function of the microtubule organizing center (MTOC).

Brief Biography

The MTOC is an important and complex cellular organelle in eukaryotes. It regulates cell motility and determines cell polarity in interphase and is required for normal cell division during mitosis and meiosis. The MTOC is highly dynamic and tightly regulated during the cell cycle. Defects in centrosomes, one of the MTOCs in mammalian cells, are linked to many human diseases such as cancer, Huntington's disease and genetic disorders. Currently I am focusing on two eukaryotic MTOCs, the spindle pole body (SPB) from the budding yeast Saccharomyces cerevisiae and the centriole/basal body from the low plants and mammals. The objectives of the research are; 1) biochemical characterization and structure determination of major compositions of the MTOC; 2) elucidation of the supermolecular assembly the MTOC; 3) combining structure, biochemistry and cell biology information to obtain insights into the molecular mechanism and the cellular functions.

Report

Stepwise assembly of cilium revealed by electron cryo-tomography

The cilium, comprising both the basal body and the axoneme, serves as a microtubule-based organelle critical for numerous cellular functions. Its assembly is intricately regulated, and mutations in cilium components or disruption of its formation can lead to various human diseases, including cancers and ciliopathies. By using electron cryo-tomography and subtomogram averaging techniques on the basal body and axoneme from the unicellular model organism Tetrahymena thermophila, we obtained high-resolution structures of the cilium at distinct regions: the probasal body, the central core region, and the axoneme. Leveraging AI-based protein modeling, we identified several basal body proteins, some distributed throughout the entire organelle, while others were specific to particular cilium locations. These findings establish a molecular model for cilium assembly, underscoring its precise spatial and temporal regulation.

Ignacio Izeddin ESPCI Paris

Research Focus

Single-molecule localization microscopy; transcription regulation, DNA repair

Brief Biography

Ignacio Izeddin is a physicist renowned for his expertise in single-molecule microscopy, aiming to elucidate molecular dynamics and organization within cells. He earned his Ph.D. in Physics from the University of Amsterdam and worked as a postdoctoral researcher at the École Normale Supérieure (ENS) and the Institut Curie in Paris with Maxime Dahan and Xavier Darzacq. In 2015, Ignacio established his own research group at the Institut Langevin, ESPCI Paris.

Throughout his career, Ignacio has been invested in deciphering the intricate mechanisms that govern cellular processes, leveraging advanced fluorescence microscopy techniques to shed light on these phenomena. His lab develops cutting-edge single-molecule techniques for biological imaging, particularly in super-resolution imaging and single-particle tracking. His work is characterized by pioneering instrumental advancements and original data treatment that are subsequently applied to biological systems, producing insights of biophysical importance. Ignacio's significant contributions to the fields of transcription regulation and DNA repair processes have helped improve our understanding of the spatial and temporal organization of cell nuclei at the molecular level.

Report

Enhanced single-molecule localization microscopy with event-based sensors

Although single-molecule localization microscopy (SMLM) gives access to spatial resolutions down to the scale of protein size, a number of applications in live-cell imaging remain challenging due to the trade-off between temporal and spatial samplings. This is particularly problematic whenever the system studied displays heterogeneous protein densities and/or dynamic processes at different temporal scales, as scientific cameras (EMCCD/sCMOS) are poorly suited to these purposes. We recently introduced a new approach to SMLM (Eve-SMLM) using event-based sensors (EBS) instead of scientific cameras. EBSs are commercially-available matrices of independent, asynchronous (i.e. they do not use an exposure time) pixels that are sensitive to optical intensity variations rather than to the absolute number of photons integrated over a set exposure time. We were able to successfully detect single molecules to perform SMLM bioimaging with spatial resolutions comparable to those achieved with scientific cameras. We furthermore used the intrinsic sparsity of the data to efficiently deal with the challenge of high density SMLM imaging, where the overlap of the Point Spread Functions (PSF) cause traditional strategies to under-perform, allowing the reconstruction of dynamic SMLM images on living samples with a temporal resolution around 1 minute [1].

Building upon Eve-SMLM, we exploit the EBS' asynchronous detection and fast response to extract single molecules' temporal emission profile without the need of oversampling, providing information about the photophysical behavior of single molecules without undermining their localization precision. Furthermore, we use the improved time resolution for modulated excitation techniques. Since Eve-SMLM provides the temporal profile of the emitted signal at the single-molecule level, it is possible to encode information about the absorption spectrum of each molecule in the retrieved signal frequency by modulating the excitation laser. We used this in the context of multicolor SMLM imaging, allowing identification of different fluorophores in a simultaneous acquisition. This simple approach furthermore can improve the spatial resolution by increasing the number of events generated by each molecule as the modulation frequency increases.

In this talk, I will present the particularities and advantages of event-based single-molecule localization microscopy (Eve-SMLM), show our results in high-density conditions, multicolor imaging, and preliminary results on multi-scale single-particle tracking with EBS.

[1] C. Cabriel, T. Monfort, C.G. Specht, and I. Izeddin. Event-based vision sensor for fast and dense single-molecule localization microscopy. Nat. Photon. 17, 1105–1113 (2023). https://doi.org/10.1038/s41566-023-01308-8

Yu-Hui Zhang **Huazhong University of Science and Technology**

Research Focus

•Cell-permeable organic fluorescent probes for live-cell super-resolution imaging
•Dynamic interactions among subcellular structures

Brief Biography

Yu-Hui Zhang, PI, full professor of Wuhan National Lab for Optoelectronics, Huazhong University of Science and Technology. She received her B.S. degree from the University of Science and Technology of China and her Ph.D. degree from the University of Hong Kong. She did postdoctoral research in Harvard Medical School and joined Wuhan National Lab for Optoelectronics in Huazhong University of Science and Technology since 2006. Dr. Zhang's research interests focus on developing cell-permeable organic fluorescent probes for live-cell super-resolution imaging and their applications. The findings have been published on Nat Methods, Light-Sci&Appl, Nat Commun, Biomaterials, J Control Release, and J Proteome Res, etc.

Report

Cell-permeable organic fluorescent probes for live-cell super-resolution imaging

Thanks to recently developed super-resolution fluorescence microscopy, imaging subcellular structures with a spatial resolution beyond the diffraction limit has been achieved. However, limitations of the existing fluorescent probes for subcellular structures, such as cell-impermeable, photo-bleaching, and a nonspecific background, hinder the characterization of dynamic interactions between organelles by super-resolution imaging in live cells. We have developed more than 20 novel cell-permeable organic fluorescent probes for various subcellular structures, including microtubules, nuclei, mitochondria, lysosomes, the ER, F-actin, intermediate filaments, early endosomes, late endosomes, and peroxisomes, with high specificity and excellent photo-stability. Our results not only greatly expand the applications of super-resolution imaging in various live-cell dynamic studies but also open up new avenues in the design of fluorescent probes for live-cell super-resolution imaging.

Vincent Villette **Ecole Normale Superieure**

Research Focus

The function of cerebellar inhibitory microcircuits during postural control and learning.

Brief Biography

My scientific aim, since my PhD obtained in 2010, is to understand the role of inhibitory microcircuits in brain computations in both health and disease. I have always worked in awake behaving animals, to relate neuronal activity to behavioral features, first with electrodes during my PhD and then using optical recordings during my postdocs, and to date.

During the first stages of my career I worked on septo-hippocampal and hippocampal circuits. During my first postdoctoral fellow done at INMED in Marseilles, I contributed to reveal network assemblies, which are building blocks of the sequential activities that fit to the spatial and or temporal representations of the mice environment. After a short postdoc in Canada, I joined the IBENS in PARIS and get a permanent position in 2017 by the CNRS.

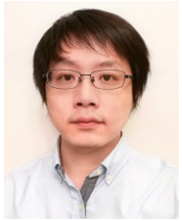
In the group of Stéphane Dieudonné, since my arrival, we study how cerebellar microcircuits are wired and coordinated during postural control. To this end, we implemented in the last past years, optical methods to record either subthreshold and spiking signals or calcium signals of population of neurons in 2D and 3D in the behaving animal. Particularly, using acousto-optic random-access two-photon microscopy (2P) and holographic pattern of fluorescence excitation together with genetically encoded voltage indicators, we pioneered, in 2P, chronic recordings of multiple neuronal activities in depth and for dozens of minutes, enabling to probe microcircuits functions.

Report

Ultrafast Optical recording of neuronal activities with voltage indicators in behaving mice and applications in the cerebellum.

The aim of our research project is to understand the rules that govern the coordination and activities of the cerebellar inhibitory microcircuitry during motor control, sensorimotor anticipation and its adaptation to novelty. This requires the study of genetically and morphologically identified neuronal ensembles in behaving rodents. Given the sustained firing of most cerebellar cell types and the lack of accuracy of indirect but standardly used calcium indicators, we have pushed the use of genetically encoded voltage indicators (GEVI) with two-photon technologies to directly monitor spikes and subthreshold activities. In this presentation, I will guide the audience through the path we have taken to achieve chronic, long duration optical voltage recordings in behaving mice. I will also present unpublished results on various GEVIs, cell types and applications.

Hanqing Xiong **Peking University**



Research Focus

Molecular spectroscopy and imaging

Brief Biography

Dr. Hanqing Xiong’ s research is focused on nonlinear molecular spectroscopy and its application in chemical and biomedical imaging. He is now an assistant professor at the College of Future Technology, Peking University. In recent years, he has published systematic results in the fields of (i) mesoscale connectomics and (ii) vibrational spectroscopy & imaging.

Report

Time-domain stimulated Raman scattering imaging for biomedical applications

As an intrinsic contrast of chemical bonds and molecular structure, Raman spectroscopy has been widely used as an essential tool to probe molecular distributions, dynamics, and environment coupling in biomedicine, chemistry, and material science. Featured by the high gain of stimulated emission, the non-distorted line shape, and its linear concentration dependence, stimulated Raman scattering (SRS) has emerged as one of the main-stream Raman imaging modalities for biomedical imaging in recent years. However, current mainstream SRS imaging methods are all based on frequency-domain excitation strategies, whose intrinsic properties hinder their applications for hyperspectral imaging with high spectral resolution. In this talk, we will discuss our newly developed time-domain SRS method that optimized for biomedical applications. Its advantages and drawbacks will be discussed in detail.

Takahiro Deguchi European Molecular Biology Laboratory

Research Focus

Development and applications of fluorescence microscopy in cell biology

Brief Biography

Takahiro Deguchi studied biophysics and received his Ph.D. in 2015 from the University of Turku, Finland. Afterwards, he moved to Genoa, Italy for his postdoc research in the Diaspro group at Istituto Italiano di Tecnologia, where he learned optical engineering for confocal and STED fluorescence microscopy, including adaptive optics technology. In 2020, he joined the Ries group at EMBL and since then has been working on a biophysical project using MINFLUX nanoscopy to monitor protein dynamics in living cells. His research accomplishments include 27 peer-reviewed scientific publications, of which 8 were first-authored, 2 patents (1 filed and 1 applied), 2 postdoc and 3 PhD fellowships including the prestigious Marie Skłodowska-Curie fellowship, and close industrial collaborations, contributing in a commercialization of Nikon NSPARC microscope. His main scientific interest is a development of super-resolution fluorescence microscopy for studies of dynamics of protein complexes in living cells.

Report

MINFLUX tracking of motor protein dynamics in live cells

Super-resolution fluorescence microscopy is a powerful tool for structural cell biology to reveal detailed spatial arrangements of biomolecules. However, it has been a challenge to study structural changes of protein machines in living cells due to their small size and fast dynamics. Among all super-resolution techniques, MINFLUX utilizes the limited photon budget of fluorescent markers most efficiently. It can achieve nanometer resolution and was shown to reach a sub-millisecond temporal resolution for tracking single fluorophores in vitro.

In this study, we focus on the motor protein kinesin-1, which is an intracellular cargo transporter and exhibits processive motility along microtubules. Driven by ATP hydrolysis, each motor domain moves with 16 nm steps in a hand-over-hand manner. Due to the small step size and fast stepping rate, previous studies of kinesin stepping behavior were limited to in vitro work with purified proteins. Here, we use MINFLUX to track fluorescently-labeled kinesin as it walks on cellular microtubules. Using motor-PAINT with purified and labelled kinesin in fixed cells, we could precisely track the path of microtubules and reveal kinesin side-stepping between neighboring protofilaments. In living cells, we were able to clearly resolve the 16 nm steps of the kinesin motor domain at ~2 nm localization precision with sub-millisecond temporal resolution.

Our results will facilitate further in vivo studies of precise motor protein kinetics, enable the super-resolution mapping of complex microtubule arrays, and pave the way towards monitoring functional conformational changes of protein machines at high spatiotemporal resolution in living systems.

Xin Zhou **Innovation Academy for Precision Measurement Science and Technology**

Research Focus

Cutting-edge MRI technology and its clinical applications with ultra-sensitive techniques

Brief Biography

Dr. Zhou is the President of the Innovation Academy for Precision Measurement Science and Technology (APM), CAS. He holds a PhD from the Graduate School of the Chinese Academy of Sciences, and subsequently engaged in three years of postdoctoral research at Harvard University followed by two years as a research fellow at UC Berkeley. In 2009, he joined the APM, as a full professor.

Dr. Zhou pioneered the first clinically approved Human Lung Gas MRI Instrument, which enables sensitive regional ventilation, microstructure, and gas exchange from the alveoli to the blood without radiation or invasion. Additionally, he has made significant contributions in the field of multi-nuclei MRI technologies and high-sensitivity MRI contrast agents. These advancements are anticipated to offer robust technical support, fresh insights, and innovative strategies for research in critical lung and brain diseases.

The Lung gas MRI system, developed by Zhou's group won the CIIF award in the 22nd session China International Industry Fair, and was presented at the National "13th Five-Year" Science and Technology Innovation Achievement Exhibition. Additionally, Dr. Zhou has been honored with several prestigious awards, including the Top Ten National Science and Technology Innovative Person accolade, the inaugural Xplorer Prize, and the National Innovation Award. His prolific research output includes over 170 publications in academic journals, and he holds more than 100 China and PCT international patents.

Report

Hyperpolarized 129Xe multinuclear MRI and molecular imaging

With the technique of spin-exchange optical pumping (SEOP), the spin polarization of laser-enhanced xenon atoms can be enhanced by four or five orders of magnitude, which makes it feasible to image the lung. Our group successfully developed the first human multi-nuclear MRI instrument in China, boosting 129Xe signal intensity more than 70,000 folds. Hyperpolarized 129Xe enables us to image the structure and function of the lung, which cannot be executed by traditional MRI. We acquired the first batch of multi-nuclear human lung images with hyperpolarized 129Xe MRI in China, and the technique has been used in Wuhan Jinyintan Hospital and Tongji Hospital for comprehensively evaluating lung microstructure and function changes in COVID-19 patients. Moreover, we have also quantitatively assessed the lung morphology and physiology changes caused by various lung diseases, such as PM2.5-induced lung injuries, radiation-induced lung injury and chronic obstructive pulmonary disease. Furthermore, utilizing the compressed sensing and AI techniques can accelerate 129Xe ventilation imaging and diffusion-weighted imaging.

Moreover, the integration of hyperpolarized xenon with multimodal molecular imaging technology has emerged as a powerful tool for swiftly identifying lesions in both laboratory and clinical settings. Our team has pioneered the development of the inaugural high-field Chemical Exchange Saturation Transfer (CEST) contrast agent, dubbed "hyper-same" (Hyperpolarized Xe signal Advanced by Metal Organic Framework), engineered to amplify gas magnetic resonance signals intensity within aqueous solutions. This innovation not only enriches ultrasensitive MRI with adaptable metal-organic frameworks but also introduces a novel avenue for crafting ultra-high sensitivity magnetic resonance molecular images. Such advancements hold promise for future clinical applications, facilitating the precise and quantitative detection of various diseases. Additionally, we have devised a multimodal imaging probe tailored to target specific markers within the tumor microenvironment, facilitating real-time imaging and accurate tumor localization during surgical resections. Furthermore, our pioneering cancer theranostic technology enables early cancer detection while concurrently eradicating cancer cells and reshaping the tumor microenvironment. This innovative approach has proven effective in halting the progression of solid tumors, thwarting cancer metastasis, and preventing recurrence, underscoring its potential in advancing cancer treatment paradigms.

Laurent Bourdieu Ecole Normale Superieure

Research Focus

I study cortical dynamics using innovative in vivo two-photon microscopy methods, including adaptive optics, wavefront shaping and fast random-access scanning using acousto-optic deflectors.

Brief Biography

Laurent Bourdieu is a CNRS researcher and head of the "Cortical Dynamics and Coding Mechanisms" team at IBENS, the Biology Institute of the Ecole Normale Supérieure, Paris.
He was trained in physics, biophysics and optics at the Ecole Normale Supérieure. After his PhD at the Institut Curie, Paris, and his postdoc in the laboratory of Albert Libchaber at Princeton University, he joined the CNRS at the University of Strasbourg to work in the fields of soft condensed matter and biophysics. In 2004 he moved to the Ecole Normale Supérieure to study cortical dynamics using two-photon microscopy in vivo. There he pioneered the development of new ultrafast scanning strategies based on acousto-optic deflectors, which he has since constantly improved. He also studies the coding of sensory information in the rodent cortex and its modulation by top-down processes.

Report

Recording neuronal activity in 3D within a cortical column with millisecond resolution using Custom-Address Serial Holography

We have designed a new optical technique referred as Custom-Address Serial Holography (CASH) that allows recording the spiking activity of hundreds of cells in 3D at a sampling rate of several hundreds of Hz in the head-fixed behaving rodent [1]. It provides an unprecedent 3D sampling speed of neuronal activity with an optical method. CASH allows indeed random address sampling of 20 cells at 1 kHz up to 200 cells at 0.1 kHz in head-fixed behaving mice across a volume of (500 µm)3. Using fast acousto-optic modulation, every laser pulse of a 40 kHz regenerative amplifier is individually patterned [2,3] to serially address the target cells with a 5x5 spot volume covering the cell body and the surrounding space to prevent motion artefacts [1]. The recorded activity is corrected for neuropil contamination by subtraction of a neuropil reference signal [1]. We validated the performance of 3D-CASH by analyzing the spatio-temporal organization of GCaMP6f expressing neuron activity in layer 2/3 and 5 of mouse primary visual cortex in response to moving contrast gratings [1]. Recently we also obtained preliminary experiments using genetically-encoded voltage indicators. Finally, we took advantage of the possibility with CASH to shape the laser wavefront at tens of kHz rate to perform trans-skull imaging: by updating the correction of aberration and scattering at the scanning rate, images could be obtained over fields of view that extend beyond the scattering memory effect [4].

References

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Ming Lei Xi'an Jiaotong University

Research Focus

Super-resolution microscopy, Optical trapping

Brief Biography

Dr. Ming Lei is a professor at Xian Jiaotong University. He received his BE degree from the School of Physics and Optoelectronic Engineering, Xidian University in 2000, and his PhD from Xian Institute of Optics and Precision Mechanics, Chinese Academy of Sciences, in 2007. He was trained as a postdoctoral research fellow in the Department of Chemistry, University of Konstanz, Germany, from 2008 to 2010. His research is focused on super-resolution microscopy and optical trapping technologies.

Report

High speed image reconstruction for structured illumination microscopy

Super-resolution structured illumination microscopy (SR-SIM) is an outstanding method for visualizing the subcellular dynamics in living cells. To date, by using elaborately designed systems and algorithms, SR-SIM can achieve rapid, optically sectioned, SR observation with hundreds to thousands of time points. However, real-time observation is still out of reach for most SIM setups as conventional algorithms for image reconstruction involve a heavy computing burden. To address this limitation, an accelerated reconstruction algorithm was developed by implementing a simplified workflow for SR-SIM, termed joint space and frequency reconstruction. This algorithm results in hundreds fold improvement in reconstruction speed relative to the widely used Wiener-SIM. Critically, the increased processing speed does not come at the expense of spatial resolution or sectioning capability, as demonstrated by live imaging of microtubule dynamics and mitochondrial tubulation.

Alexandra Fragola **Paris Saclay University**

Research Focus

Adaptive optics fluorescence microscopy for fast 3D in vivo imaging

Brief Biography

I was an assistant professor in the physics department for 17 years at Sorbonne University, and developed my research in the Synthesis and Imaging of Inorganic Nanoparticles team at ESPCI (Paris). I am now professor of physics at the University of Paris Saclay, in the laboratory Institut des Sciences Moléculaires d' Orsay. I have a long experience in the development and the optimization of advanced microscopy setups for high-resolution dynamic imaging of living samples. In recent years, my research has focused on adaptive optics in fluorescence microscopy for fast, high-resolution 3D imaging deep inside living specimens, such as mice or zebra fish. I participate in or lead numerous multidisciplinary projects with researchers in biology and chemistry, and I work in close collaboration with the company Imagine Optic (Orsay).

Report

Adaptive optics fluorescence microscopy for high-resolution in-depth imaging in vivo

The study of fast biological phenomena at the cellular level has become possible even in depth in biological tissues in several animal models and rodents, thanks to efficient 3D microscopy techniques. Light sheet fluorescence microscopy (LSFM) is particularly well suited to imaging relatively transparent specimens such as zebrafish, while 2-photon excited fluorescence microscopy (TPEF) remains the technique of choice for more scattering tissues like mammalian brain. But image quality deep inside biological tissues remains limited by scattering and optical aberrations due to the refractive index inhomogeneity, which limit both resolution and sensitivity in all microscopy modalities. To overcome this difficulty, in particular regarding aberrations, we proposed a few years ago an innovative strategy of adaptive optics for neuroimaging, by adapting pioneer work from astronomy to the constraints of fluorescence microscopy. We performed extended-based adaptive optics integration into TPEF and LSFM microscopy, in order to demonstrate the performance of this new, fast closed-loop approach and the ability to distinguish individual cells even at great depths. This is key to many studies, as a major biomarker is the quantification of cell amount/density (segmentation) in many fields (developmental biology, neuro, cardio).

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Jianfeng Liu **Huazhong University of Science and Technology**

Research Focus

G protein coupled receptor (GPCR) and ion channel

Brief Biography

Prof Jianfeng LIU is a Principal Investigator at the College of Life Science and Technology (CLST), Huazhong University of Science and Technology. He also serves as the dean of CLST and the chair of Biophysical Society of China in Hubei Branch. He has received the National Science Fund for Distinguished Young Scholar, Chang Jiang Scholars Program and Tan Jiazhen Life Science Award.
His research focuses on the structure and function of G protein coupled receptor (GPCR) dimers. His work has found the asymmetric activation of class C GPCRs and illustrated the dynamic allosteric modulation for signaling integration in these GPCR dimers. He also established sensitive sensors and assays to monitor GPCR dimer activation and identified new function of GPCRs in aging and aging-related diseases regulation. He has published over 80 articles in high-level journals such as Cell (x2), Nature (x2), Nature Metabolism; Nature Aging, Nature Chemical Biology (x2), Nature Communications (x5), Science Advances, Science Signaling as the co-responding author, with more than 4500 citations.

Report

Structural snapshot of class C GPCRs dimer activation

Most GPCRs are cell surface proteins that can signal through G proteins in a monomeric state. These proteins can however influence each other through possible allosteric interaction within dimeric or even oligomeric entities. The discovery of the mandatory homodimeric or heterodimeric nature of the class C GPCR, stimulated research on the allosteric interaction and signal integration through GPCR dimerization.
We first investigated the dynamic rearrangement of the dimer interfaces during activation of the two representative class C GPCR dimers: mGlu and GABAB receptors. We identified TM4 and TM5 as the interface of the inactive state, while TM6-TM6 is indeed the main component of the active interface. These findings were confirmed by the cryo-EM structures of either the mGlu2, mGlu3, mGlu4, mGlu5, GABAB or CaSR dimers in different states. Further, we found no outward shift of TM6 was observed in GABAB, when coupled to G protein, but relied on a shallow pocket formed by intracellular loops for G protein recognition, which is different from GPCR monomers. It was confirmed later in mGlu2, mGlu4 and CaSR dimers, indication a distinct asymmetric activation in GPCR dimers. Additionally, we found a high proportion of mGlu2-4 heterodimer in native tissues in brain using specific nanobody tools, indicating different properties might generate through GPCR dimerization. We also observed differences in G protein responses in neurons triggered some endogenous neurontransmitter GPCR dimers, which was different from that in recombinated cell line.

Perrine Paul-Gilloteaux **Nantes University**

Research Focus

Image analysis in microscopy , in particular for correlative microscopies

Brief Biography

After electrical engineering studies, P. Paul-Gilloteaux defended her PhD in 2006 in Rennes, about deformations of the brain during surgery. She did a first postdoc in Italy to work on augmented reality in surgery, and then moved to Ireland in the Hamilton Institute Systems Biology group, where she co-developed an intelligent fluorescent microscope. In 2010, she joined the CNRS as research Engineer in the microscopy facility in Institut Curie, Paris, France. Her job was to support the biologists in image analysis, by adapting existing software or developing new methods.
In June 2015, she joined the federative structure of research in biology and health of Nantes, France. She is now leading R&D activities in multimodal image analysis and heading MicroPICell, a microscopy facility performing services and R& D in histology, photonic microscopy and image analysis, part of the France BioImaging national infrastructure. She is data mission officer for France BioImaging, one of the co-funder of NEUBIAS (network of European Bio Image Analysts) and the COMULIS (Correlated Multimodal Imaging in Life Sciences) networks.

Report

Image analysis methods for multimodal correlative microscopies: from segmentation using AI to information fusion

Correlative microscopy is a set of methods that allows imaging the same sample under different modalities, without necessarily having a multimodal hybrid microscope. The simplest acquisition workflow is to image a sample under a first microscope, then move it and find the same area to image it under another microscope that will provide different and complementary information. Accurate registration of imaging modalities is one of the key steps of a correlative imaging workflow. For this, we have developed EcClem, a plugin under the free and open source software platform ICY, aiming to ease the registration process in correlative microscopies workflows, whatever the modalities in use. We present here EcClem and EcClemAutofinder, which allow automatic registration based on segmentations of common features. We also provide a new method of error estimation in registration, assessing the co-localization of pixels from different registered modalities. In this presentation, we exemplify its usage on several examples of correlative microscopies, light to electron microscopy correlation but also with a broader range of modalities.

Yiqian Wu **Peking University**

Research Focus

Synthetic biology, immunotherapy

Brief Biography

Dr. Yiqian Wu received her PhD in Bioengineering from UCSD in 2020 under the supervision of Dr. Yingxiao Wang and Dr. Shu Chien. Dr. Wu continued her research at UCSD as a postdoctoral fellow and later an assistant project scientist. She joined Peking University as an assistant professor in 2023. Her main research interests include optogenetics, sonogenetics, and gene and cell therapy.

Report

Sonogenetics-based reprogramming of cellular functions

Ultrasound, owing to its strong penetration power in biological tissues and great clinical compatibility, has emerged as a new tool for manipulating genetic and cellular functions, especially for therapeutic purposes. Here we have developed sonogenetics-based approaches to control T cell functions for immunotherapy, i.e., ultrasound-controllable chimeric antigen receptor (CAR)-T cell therapy. We have also engineered an ultrasound-controllable CRISPR-based toolbox for genetic and epigenetic reprogramming in vivo.

Rémi Galland **University of Bordeaux**

Research Focus

Fluorescence Microscopy for Biology

Brief Biography

After a diploma from the engineer school Institut d' Optique Graduate School in 2004, R. Galland obtained his PhD in physics in 2008 at the University Joseph Fourier in Grenoble. He then developed several advanced optical tools to probe or control biological systems through two Post-Doc at the University Joseph Fourier and within the CEA-Grenoble. In 2013, he joined the team "Quantitative Imaging of the Cell" at IINS headed by JB. Sibarita where he developed the soSPIM technology in collaboration with the group of V. Viasnoff (MBI, Singapore). In 2015, R. Galland obtained a CNRS position (Chargé de Recherche – CRCN) in the team "Quantitative Imaging of the Cell" at IINS in Bordeaux where he continued to develop the soSPIM technology and actively participate to its valorization its dissemination. He also participated to the development of several advanced super-resolution imaging methods aiming at improving their content and automation. He also co-leads a working group on Light-Sheet Microscopy techniques within the France BioImaging (FBI) infrastructure. He owns 4 patents (3 being about the be licensed to the industry) and participate to 5 Material Transfer Agreements.

Report

Multi-scale imaging using the soSPIM technology - From in depth single molecule imaging up to 3D cell cultures screening

The combination of fluorescents probes and light microcopy offers the unique capacity to monitor the organization and dynamics of proteins of interest within living samples. Recently, new and extremely promising 3D in vitro biological models (spheroids, organoids, ⋯) have emerged, making it possible to better recapitulate the physiology and functions of human organ and/or pathologies.

In the meantime, 3D fluorescence microscopy for biology is undergoing an in-depth revolution with the advent of Light-Sheet Fluorescence Microscopy (LSFM or SPIM) technics. They enable to tackle many limitations of standard 3D imaging approaches in term of photo-toxicity, speed and sensitivity, but remain limited in their imaging throughput or spatial resolution.

Within the team, we developed an innovative SPIM architecture, called soSPIM (single-objective Selective Plane Illumination Microscope), which relies on the integration part of the optics into the imaging devices in place of the excitation objective. I will discuss the multiscale imaging capability of this technology which allows to image down to the nanoscale organization of proteins up to the development of living 3D organoids. Especially, I will present the recently engineered multiwell devices, named JeWells, that enable to standardize and parallelize both the culture and the imaging of 3D cell models. Combined with a tunable analysis framework, it allows to turn the soSPIM system into a versatile high-content screening platform of living 3D cellular models.

Pingyong Xu **Institute of Biophysics, CAS**

Research Focus

Super-resolution microscopy probes, methods and visualization studies of important biological processes

Brief Biography

Dr. Pingyong Xu is a tenured professor of Institute of Biophysics, Chinese Academy of Science and a professor of college of life sciences at University of Chinese Academy of Sciences. He received his Ph.D from Huazhong University of Science and Technology in 2004. His laboratory combines spectroscopy, biophysical microscopy techniques, and protein design and engineering for the development of novel optical imaging tools, especially photo-controllable fluorescent proteins and functional probes. Dr. Xu has developed the first fluorescent protein mEosEM for the correlative light and transmission electron microscopy. He has developed many excellent fluorescent protein probes for PALM/STORM, SOFI, RESOLFT, NL-SIM and other super-resolution technologies to greatly improve their temporal and spatial resolutions. Among them, mEos3.2 and Skylan-S are star probes for PALM and SOFI imaging, respectively. Dr. Xu has also developed SIMBA and Quick-SIMBA live-cell super-resolution technique based on single-molecule localization.

Report

Photo-controllable fluorescent proteins and derived super-resolution technologies

Christophe Leterrier Aix Marseille University

Research Focus

Super-resolution microscopy for the study of the neuronal cytoskeleton

Brief Biography

An engineer by training, Dr Leterrier turned to cell biology and neurobiology for his PhD. He since aims at understanding how neurons are organized at the cellular level: how do they differentiate, then build and maintain their incredibly complex arborization? Since 2017, he's leading the NeuroCyto lab in Marseille that applies advanced microscopy techniques to directly observe molecular assemblies at the nanoscale inside neurons, revealing how they organize and shape neuronal physiology.

Report

The axonal cytoskeleton down to the nanoscale

The intricate arborization and molecular identity of axons is maintained for decades, but must also continuously adapt to changes in the environment and modulate the activity of neurons. Axons fulfill these paradoxical demands thanks to a unique cytoskeletal organization that ensures the coordinated transport, anchoring and assembly of axonal components. In our lab, we use super-resolution microscopy to delineate and map the nanoscale architecture of cytoskeletal structures within the axon: the periodic actin/spectrin submembrane scaffold, intra-axonal hotspots and trails, and presynaptic actin assemblies. We are exploring their molecular organization and functions by combining versatile labeling approaches, correlative live-cell/super-resolution/electron microscopy and quantitative analysis that allow for high-content, nanoscale interrogation of the axonal architecture.

Fazhan Shi **University of Science and Technology of China**

Research Focus

Quantum metrology and applications

Brief Biography

Fazhan Shi, professor in School of Physics, USTC. Researches on quantum metrology based on Nitrogen-Vacancy single spin in diamond and its applications on life science, such as the quantum measurement technology and methods, single-molecule spectroscopy and nanoscale imaging.
So far he has published more than 70 scientific papers, and received the XPlorer Prize, Natural Science Award from the Ministry of Education of China (the first class prize, the second position), the Young Investigator Award of the International EPR/ESR Society etc.

Report

Towards in-vivo single-molecule magnetic resonance with diamond quantum sensing

Magnetic resonance (MR) is one of the most important techniques for characterizing compositions, structure and dynamics of molecules. However, current methods need billions of uniform units on centimeter-scale to accumulate large enough signal-to-noise ratio. High sensitivity MR techniques are urgently needed for new applications on single molecule. A quantum sensor to accomplish single molecule detection is the nitrogen-vacancy (NV) defect center in diamond. By combining the quantum controls and long coherence time of NV, we have experimentally realized single molecule scale nuclear MR and electron spin resonance. This talk introduces the major works we achieved along this line. (I) Single molecule MR spectroscopy. We obtained the first single-protein spin resonance spectroscopy under ambient conditions [Science 347, 1135 (2015)], the electron spin resonance spectroscopy of single molecules under physiological conditions [Nature Methods 15, 697 (2018)], and developed the zero-field electron spin resonance spectroscopy on nanoscale [Nature Communications 9, 1563 (2018); Science Advances 6, eaaz8244 (2020)]. Using nanodiamond sensor, we realized in-situ electron spin resonance spectroscopy [Nature Communications 14, 6278 (2023)]. (II) Microscale MR imaging. Using NV sensor, we realized nanoscale MRI of ferritins in a single cell [Science Advances 5, eaau8038 (2019)], and the Immunomagnetic microscopy of tumor tissues with micrometer-resolution [PNAS 119, e2118876119 (2022)]. These results, together with the relation works in the field, open a door to nanoscale/single molecule MR and will be potentially used as a new tool on a broad range of scientific areas from life science to physics and chemistry.

Sandrine Lévêque-Fort **Paris-Saclay University**

Research Focus

Design of new optical microscope

Brief Biography

Sandrine Lévêque-Fort is a CNRS Researcher Director at the Institute of molecular science (ISMO) in Paris Saclay University.
She obtained her PhD on the development of a new acousto-optic imaging approach for imaging through scattering media in the Optical Lab of ESPCI in Paris. She then became a postdoctoral fellow at Imperial College, where she started to develop time resolved fluorescence microscopy but also structured illumination strategy. She joined the CNRS in 2001 to develop different strategies to improve spatial and temporal resolution for fluorescence microscopy, by implementing new configurations or by introducing plasmonics substrates to engineer fluorescence emission. Since 2009, she has proposed various approaches to bypass the diffraction limit, taking take advantage of supercritical angle fluorescence (SAF) emission as an alternative intrinsic tool given by the fluorophore itself to access axial information, or by introducing a time signature within the localization process. Her current developments aimed at offering both structural and functional information at nanoscale in living cells. She' s the recipient of the Irène Joliot Curie prize in 2020 (Women and company) and Knight of the national order of merit.

Report

Time-Modulated illumination for enhanced single molecule localization microscopy

In Single Molecule Localization Microscopy (SMLM), the positions of the fluorophores are obtained from a fitted Point Spread Function. This spatially based localization precision will then strongly depend on the PSF shape, which can be degraded by defocusing and aberrations and affect both lateral and axial localization precision, but also the capability to image in depth in complex samples. We proposed an alternative localization method called ModLoc where the uniform excitation is replaced by a time-varying structured illumination over the entire field of view. The illuminated fluorophores have a modulated emission where the phase encodes their position. The demodulation of the fluorescence emission requires extracting four intensities values for each single molecule event, and as emitters can exhibit fast ON-time, we have developed various demodulation strategies based on the introduction of active optical elements that samples the modulated emission in distinct subarrays of the camera to take into account all events. We will present recent developments in particular a new implementation which offers an increased field of view (~50 x 50 µm2) with a flexible demodulation speed and the capability to reveal multiple dyes based on a specific implementation of spectral demixing. We will in particular present the unique performances of ModLoc to retrieve 3D images of multiple targets at various depths but also in complex samples.

Yongdeng Zhang Westlake University

Research Focus

Super-resolution microscopy

Brief Biography

Dr. Yongdeng Zhang received his Ph.D. degree in Biophysics from Huazhong University of Science & Technology with joint training at the Institute of Biophysics in 2013. He then joined Yale University School of Medicine as a Postdoctoral Associate in 2014. He joined the School of Life Sciences at Westlake University as a Principal Investigator in 2020.

Report

Seeing the Unseen: One Molecule at a Time

Super-resolution microscopy now plays a crucial role in exploring the complex molecular mechanisms underlying a wide range of biological processes. Here, we present the development of a highly efficient multicolor 4Pi-SMS technique, enabling the acquisition of 3D super-resolution images of entire mammalian cells at a resolution of 5-10 nm. Additionally, notable progress has been achieved in attaining high-quality 3D super-resolution imaging of biological tissue samples. These developments hold great potential to enhance our comprehension of biological processes at the nanoscale level.

Jiamin Wu **Tsinghua University**

Research Focus

Computational imaging, Intravital fluorescence microscopy

Brief Biography

Jiamin Wu is an associate professor in the Department of Automation at Tsinghua University, and PI at the IDG/McGovern Institute for Brain Research, Tsinghua University. His current research interests focus on computational imaging and system biology, with a particular emphasis on developing mesoscale optical setups for observing large-scale biological dynamics in vivo. He has proposed a series of new microscopic imaging framework including camera-array-based gigapixel mesoscale microscopy, scanning light field microscopy, digital adaptive optics, and two-photon synthetic aperture microscopy to overcome the barriers of intravital imaging, with orders of magnitude improvement in spatiotemporal resolution, imaging data throughput, and orders of magnitude reduction in phototoxicity for long-term observations. His work has been published in more than 40 journal papers such as Nature, Cell, Nature Photonics, Nature Biotechnology, Nature Methods, etc, opening up a new horizon for the study of large-scale intercellular interactions in mammals. He has served as the Associate Editor of PhotoniX and IEEE Transactions on Circuits and Systems for Video Technology, and Guest Editor in Chief of Light: Science & Applications.

Report

Scanning light-field imaging for mesoscale intravital fluorescence microscopy

Long-term mesoscale intravital 3D imaging in mammals is vital to study diverse intercellular behaviors and organelle functions at the organ level during native patho-physiological processes. However, optical heterogeneity, tissue opacity, and phototoxicity pose great challenges, leading to the tradeoff between the field of view, resolution, speed, and sample health. In this talk, I will discuss our recent work in mesoscale intravital fluorescence microscopy based on computational imaging methods and artificial intelligence algorithms. I will show several typical examples for broad biomedical applications, including brain-wide neural recoding in mice at single resolution, 3D voltage propagations in Drosophila larval neurons, membrane dynamics in zebrafish embryos, and large-scale cell migrations during immune response and tumor metastasis in mice. We believe these novel technological advancements enable simultaneous in vivo studies of large-scale morphological and functional cellular dynamics, opening up a new horizon for the systemic understanding of intercellular interactions and collective cellular behaviors.

Chunzhu Zhao Peking University

Research Focus

Multi-photon microscopy for brain imaging

Brief Biography

Dr. Zhao is a research assistant professor at College of Future Technology of Peking University. He obtained his doctorate degree in optical engineering from Changchun University of Science and Technology, followed by postdoctoral training at Peking University. His research focuses on multi-photon microscopy for brain imaging, and developed a miniature three-photon microscope for deep brain imaging and a miniature two-photon microscope with a millimeter field of view. His work has been published in journals such as Nature Methods, Optics Express, and Applied Optics. In recognition of his achievements in deep brain imaging, he was honored as one of the "Innovators under 35" in the Asia-Pacific Region by MIT Technology Review in 2023.

Report

Miniature multi-photon microscopy for deep brain imaging

Developments in miniature fluorescence microscopes have enabled visualization of brain activities and structural dynamics in animals engaging in self-determined behaviors. However, achieving deep brain imaging in freely behaving animals remains a challenge due to the optical scattering properties of brain tissue. This talk will discuss the methods for deep brain imaging with miniature two-photon and three-photon microscopes, as well as the imaging tests and brain imaging data obtained by the newly developed miniature multi-photon microscopes. The miniature two-photon microscope achieved the neuronal calcium imaging at a depth of 0.8 mm, and the miniature three-photon microscope extended the depth to 1.2 mm. In addition, the use of miniature multi-photon microscopes for multi-color imaging and large field of view imaging will also be discussed.

Institution

National Multimode Trans-scale Biomedical Imaging Facility

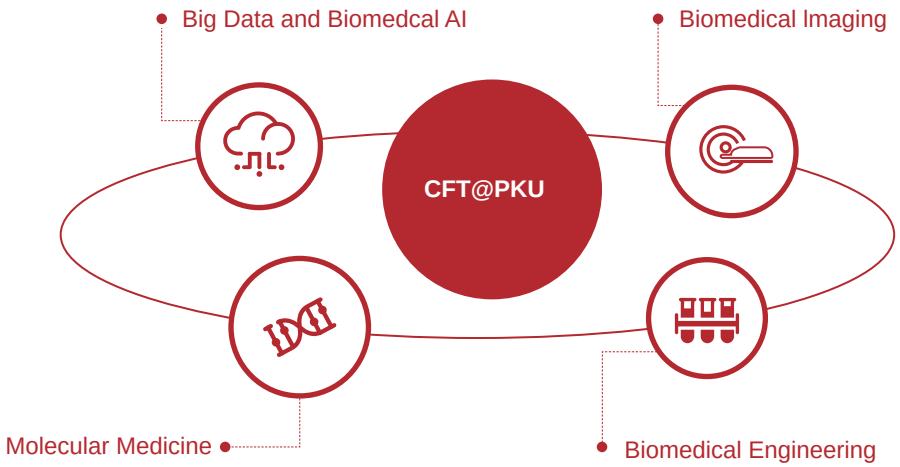
Multimode Trans-scale Biomedical Imaging Facilities is a construction project identified in the "Thirteenth Five-Year Plan" for the Construction of National Major Scientific and Technological Infrastructure, and it is a large-scale scientific facility in the field of biomedical imaging initiated by Chinese scientists. As the project's legal entity construction unit, Peking University, in collaboration with the Institute of Institute of Biophysics， Chinese Academy of Sciences, Harbin Institute of Technology, University of Science and Technology of China, Institute of Physics CAS，and The Institute of High Energy Physics of the Chinese Academy of Sciences, jointly undertake the construction task. The total investment in the project is 1.717 billion yuan. The construction site is located in the core area of Huairou Science City in Beijing, with a construction land of 100 acres and a newly added construction area of 72,000 square meters. The project is expected to be completed in 2024.

The goal of the imaging facility is to break through the scale barrier, with principle innovation and independent integration as the main focus. Through research and development and organic integration of different imaging modalities, a seamless, integrated biomedical imaging technology cluster from molecules to humans is created in terms of time and space scales and modalities. The facility consists of four core devices and one auxiliary platform, including Multimode Medical Imaging Platform, Multimode Live System Imaging Platform, Multimode High-Resolution Molecular Imaging Platform, Multimode Trans-scale Image Data Integration System, and auxiliary platforms such as Animal Model Platform and supporting facilities. Through interconnection and intercommunication, the imaging facility will cover a scale range spanning nine orders of magnitude, forming a trans-scale, multimode, automated, and high-throughput biomedical imaging research platform, becoming a world-class biomedical imaging facility in comprehensive capabilities.

The imaging facility will implement an open, flow, and selection mechanism, open and shared to the whole country, and will also establish an international alliance with biomedical imaging platforms in the United States, the European Union, and other places. The imaging facility will provide imaging omics research methods for complex life science problems and major diseases, conduct panoramic research and analysis of major biomedical scientific problems, and promote the paradigm shift of biomedical research. At the same time, the imaging facility will become a national base for leading and driving technological and methodological innovations. It will establish an industrial innovation alliance with universities, research institutes, biomedical imaging equipment, software, and probe companies to create an innovative ecosystem for biomedical imaging technology "industry-university-research-application." This will provide strategic support and guarantees for realizing "Created in China" of high-end biomedical imaging equipment.

College of Future Technology, Peking University

In the historic intersection of the new round of scientific and technological revolution and industrial reform around the world and the transformation of our country's development pattern, it is the historical mission of higher education to train leading talents of science and technology innovation with forward-looking and inter-disciplinary thinking, to seize the opportunity of future scientific and technological development. In order to fully implement the General Secretary Xi Jinping's important discussion on education, science and technology, steadily promote the construction of new engineering discipline, and strengthen the research of original, leading as well as groundbreaking future-oriented science and technology, Peking University established the College of Future Technology on September 23, 2020.



The College of Future Technology of Peking University has integrated talents from the Institute of Molecular Medicine, the Department of Biomedical Engineering, and the National Center for Biomedical Imaging Science, and set out to create a department of Big Data and Biomedical Artificial Intelligence. Since its establishment, the College has made gratifying progress in refining the discipline direction, strengthening team building, innovating teaching system, promoting the construction of national imaging facilities, as well as deepening the integration of industry and education and so on. In May 2021, it was selected into the list of the first batch of colleges of future technology constructed by the Ministry of Education.

Taking the future health and disease prevention technology as its main direction, the College of Future Technology, acting on the spiritual tradition of "inclusiveness and excellence" and adhering to the pursuit of "World class, Chinese characteristics, Peking University style", aims at the technological development direction with significant social and economic benefits. Relying on industry-university-research innovation projects, national major engineering tasks and talent needs of emerging industries, it transforms comprehensive resources such as research bases and high-tech enterprises jointly, exploring the formation of an industry-university-research integrated technology innovation system and a new training model for engineering talents to cultivate leading talents who are "innovative, versatile and interdisciplinary".

National Biomedical Imaging Center, Peking University

National Biomedical Imaging Center (NBIC) was established in September 2020, which is an important strategic measure of Peking University for the construction of national major science and technology infrastructure and the construction of "double first-class". NBIC is an independent entity for scientific research institutions. Peking University has made full use of its multidisciplinary advantages to build a national major science and technology infrastructure project of the 13th Five-Year Plan -- multi-modal and multi-scale biomedical imaging facility, and is committed to exploring frontier scientific issues and tackling key technologies. As the main body of imaging facility construction and operation, NBIC focuses on frontier exploration and technological innovation in the field of biomedical imaging science, provides technical support for basic life science and medical research, and provides solutions to major issues of life and health. At the same time, we will promote the innovative development of biomedical imaging technology, and promote the early realization of "created in China" in high-end scientific research and medical equipment.

Guided by national facilities, NBIC takes into account both free exploration and organized innovation, vigorously develops scientific research based on big science facilities, and strives to realize more "from 0 to 1" source innovation. Relying on big science facilities, it promotes the construction of frontier interdisciplinary disciplines of "imaging omics", promotes the change of scientific research paradigm in the field of life science, and cultivates future innovative talents in "big science" of life science and compound innovative talents in "new engineering" and "new medical science".

Institute of Biophysics, Chinese Academy of Sciences

The Institute of Biophysics, Chinese Academy of Sciences (hereinafter referred to as the "IBP") is a national basic research institute dedicated to fundamental life sciences, established in 1958. The mission of the institute is to give full play to the comprehensive advantages of multi-disciplinary and achieve basic, forward-looking and strategic breakthroughs in the frontier fields of protein sciences, brain and cognitive sciences, epigenetics, infection and immunity, and nucleic acid biology. The institute also strives to strengthen innovative research and development of key equipment in the field of life sciences, achieve key breakthroughs in key technologies and experimental methods, build a transformational research system focusing on biopharmaceuticals and in vitro diagnostics, and lead the innovation in the national innovation system. It will continue to produce major leading scientific research achievements and strive to enter the ranks of world-class research institutes in the fields of life and health sciences.

State Key Laboratories

Biomacromolecules
Brain and Cognitive Sciences

Key Laboratories of CAS

Biomacromolecules
Epigenetic Regulation and Intervention

CAS Engineering Laboratory

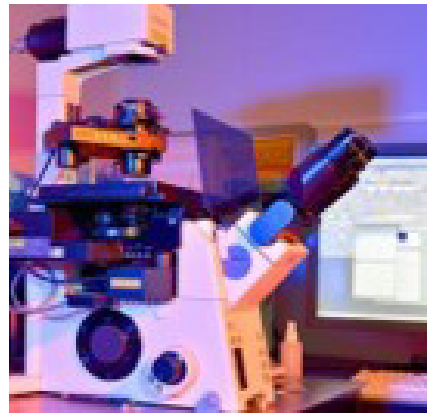
Nanozyme

Beijing Engineering Technology Research Center

Biomacromolecules Drug Transformation
Biomedical-Molecular Detection

IBP has two state key laboratories: the National Laboratory of Biomacromolecules and the State Key Laboratory of Brain and Cognitive Science; Two key laboratories of the Chinese Academy of Sciences (CAS): the Key Laboratory of Biomacromolecules, Chinese Academy of Sciences, and the Key Laboratory of Epigenetic Regulation and Intervention, Chinese Academy of Sciences. In addition, it hosts the Beijing Engineering Technology Research Center of Biomacromolecule Drug Transformation, the Beijing Engineering Technology Research Center of Biomedical-Molecular Detection, and the CAS Engineering Laboratory for Nanozyme.

IBP actively engaged in the development of one facility and two platforms in Huairou Science City (HSC). It plays a collaborative role in the construction of National Multimode Trans-Scale Biomedical Imaging Center (NBIC), one of the key projects listed in the "13th Five-year Plan for national major scientific and technological infrastructure". IBP also takes the lead in establishing CAS-Beijing Interdisciplinary Research Platform for Brain, Cognition and BCI. Additionally, it is a key participant in the collaborative construction of the Transdisciplinary Platform of Brain Functional Connectome and Brain-inspired Intelligence, one of the Science and Education Infrastructure of CAS.



Relying on the institute, the Core Facility for Protein Research, Chinese Academy of Sciences and the Beijing MRI Center for Brain Research help provide a complete public technical support for scientific research. At the same time, they carry out experimental methodology and experimental technology innovation research, and are open to the public.

IBP has accumulated extensive experience in the open operation of large-scale instrument platforms. It has received accolades for excellence in the open sharing assessment and evaluation of major scientific research infrastructure and large scientific research instruments at central-level universities and scientific research institutes, overseen by the Ministry of Science and Technology, the Ministry of Finance, and the Ministry of Education of the People’ s Republic of China. Impressively, IBP has consistently a top-ranking position for five consecutive years.

2
National Natural Science Award First prize

14
National Natural Science Award Second prize

5
National Award for Science and Technology Progress Second prize

IBP has garnered numerous prestigious awards, including 2 first prizes and 14 second prizes from the National Natural Science Award, as well as 5 second prizes from the National Award for Science and Technology Progress. Furthermore, IBP has also been recognized with important accolades at the provincial, ministerial, and CAS levels.

IBP's research achievements in various projects have been recognized as significant contributions. "Total Synthesis of Yeast Alanine Transfer RNA", "Quantitative Relation Between Modification of Functional Groups of Proteins and Their Biological Activities" ,"Crystal Structure Determination of the Major Light-Harvesting Complex (LHC-II) of Spinach" ,and "Crystal Structure Determination of Porcine Insulin" were included in the selection of one hundred significant achievements during the CAS' 60th anniversary celebration; "Structure-Function Relationship of Eukaryotic Membrane Proteins and Protein Complexes" was acknowledged as part of the 25 major scientific and technological achievements and landmark progress during the 12th Five-Year Plan of CAS.

In the summary evaluation of the first-stage of the goals and tasks of the CAS "Leading Action Plan", the "Structure, Function, and Regulation of Supra-Molecular Complexes in Photosynthesis" led by IBP, was selected as a major scientific and technological achievement and a landmark progress. The major breakthrough in "Structure, Function, and Regulation of Supra-Molecular Complexes", and three key cultivation directions including "Functions and Applications of Non-Coding RNA", "Innovation and Transformation in Biological Macromolecular Drugs", and "Biophysical New Technologies and Methods", have been rated excellent.

France-BioImaging-NIBH, led by the CNRS, is the national research infrastructure for biological imaging and the French node of the ERIC Euro-BioImaging. The infrastructure is organised around 8 regional nodes bringing together 23 central facilities associated with specialised R&D teams and a cross-cutting node dedicated to computing for imaging data processing. Two categories of technologies are available within France BioImaging: advanced imaging systems using optical microscopy and sophisticated and innovative imaging systems and methods. They are provided by more than 200 expert imaging scientists. In addition to operating technologies, FBI members offer training (FBI-AT) and data analysis (FBI-AS) services, participate in technology transfer between R&D laboratories and imaging facilities, and develop an integrated imaging data management and continuous improvement strategy, in collaboration with other national research infrastructures. The FBI is an active member of the Global BioImaging initiative.

Euro-BioImaging ERIC offers international researchers access to state-of-the-art biological and biomedical imaging technologies and expertise via 41 Nodes, which are renowned imaging centres hosted by national research institutions and universities across 18 European countries (member states) and by one intergovernmental organization, the European Molecular Biology Laboratory (EMBL). These Nodes comprise 192 facilities and represent a wide range of expertise and innovative nano-, micro- and macroscopic imaging techniques, spanning more than 100 different biological and biomedical imaging technologies. Euro-BioImaging’ s mission is to provide researchers with imaging services that bridge biological and biomedical imaging and facilitate innovative and world-class research.

Global BioImaging is an international bottom-up initiative, bringing together imaging professionals and facility staff. GBI advocates imaging technologies and research infrastructures' critical role in advancing life sciences and addressing societal challenges. GBI's impact extends beyond its network, inspiring the formation of national and crossnational imaging networks globally. By supporting national networks in securing funding and build ing sustainability for imaging infrastructures, GBI has democratised access to highquality imaging facilities and boosted open science.

FRANCE-BIOIMAGING

Sino-France 2nd Joint Meeting on BioImaging