Novel insights into plant biology with electron microscopy

From ultrastructural visualization to atomic-resolution analysis

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Pu Qian, PhD
Scientist III, Research & Development
Thermo Fisher Scientific

Rebecca Thompson, PhD
Director, Sales Development
Thermo Fisher Scientific

Foreword

Structural biology techniques, with their ability to unravel the 3D architecture of macromolecules, have played a crucial role in deciphering various physiological activities of plants at the molecular level.

Cryo-electron microscopy, in particular, has emerged as a powerful approach that enables the study of many previously intractable targets such as membrane proteins, heterogeneous protein complexes, and megacomplex assemblies. It has been used in almost all major areas of the plant sciences, offering a glimpse into the hidden molecular machinery within plant cells. Electron microscopy is one of the only techniques that can provide biological insight across orders of magnitude in length scale; from high-resolution structures that reveal the molecular organization of macromolecules, to volume electron microscopy techniques that unlock fine details of ultrastructural organization in cells, tissues, and even whole organisms.

Photosynthesis is one of the most important reactions that occur on Earth. This intricate process involves a series of proteins and pigment-protein complexes, many of which have been investigated at unprecedented resolution with cryo-EM, giving us a greater understanding of the mechanisms that drive photosynthesis. Cryo-EM has also been used to study other key plant proteins that are involved in a range of physiological activities including nutrient uptake and distribution, cell signaling, and responses to environmental stimuli and stressors. Even for more sophisticated biological systems, including virus-plant interactions and organelle structures, cryo-EM is providing a variety of pivotal and irreplaceable structural information.

In structural biology, new hardware and software are constantly improving both the quality and quantity of data that can be collected. The wide range of novel electron microscopy techniques, including single particle analysis and cryo-electron tomography, presents a unique opportunity to bring together structural data with cellular- and tissue-scale molecular organization. We are excited to support the plant sciences community with these novel techniques and beyond, helping you reach remarkable insights into plant physiology and biology.

Cover image:
PDB 7ZCU

About Thermo Fisher Scientific

As the world leader in serving science, our innovative microscopy solutions and application expertise help scientists find meaningful answers that accelerate breakthrough discoveries, increase productivity, and ultimately change the world.

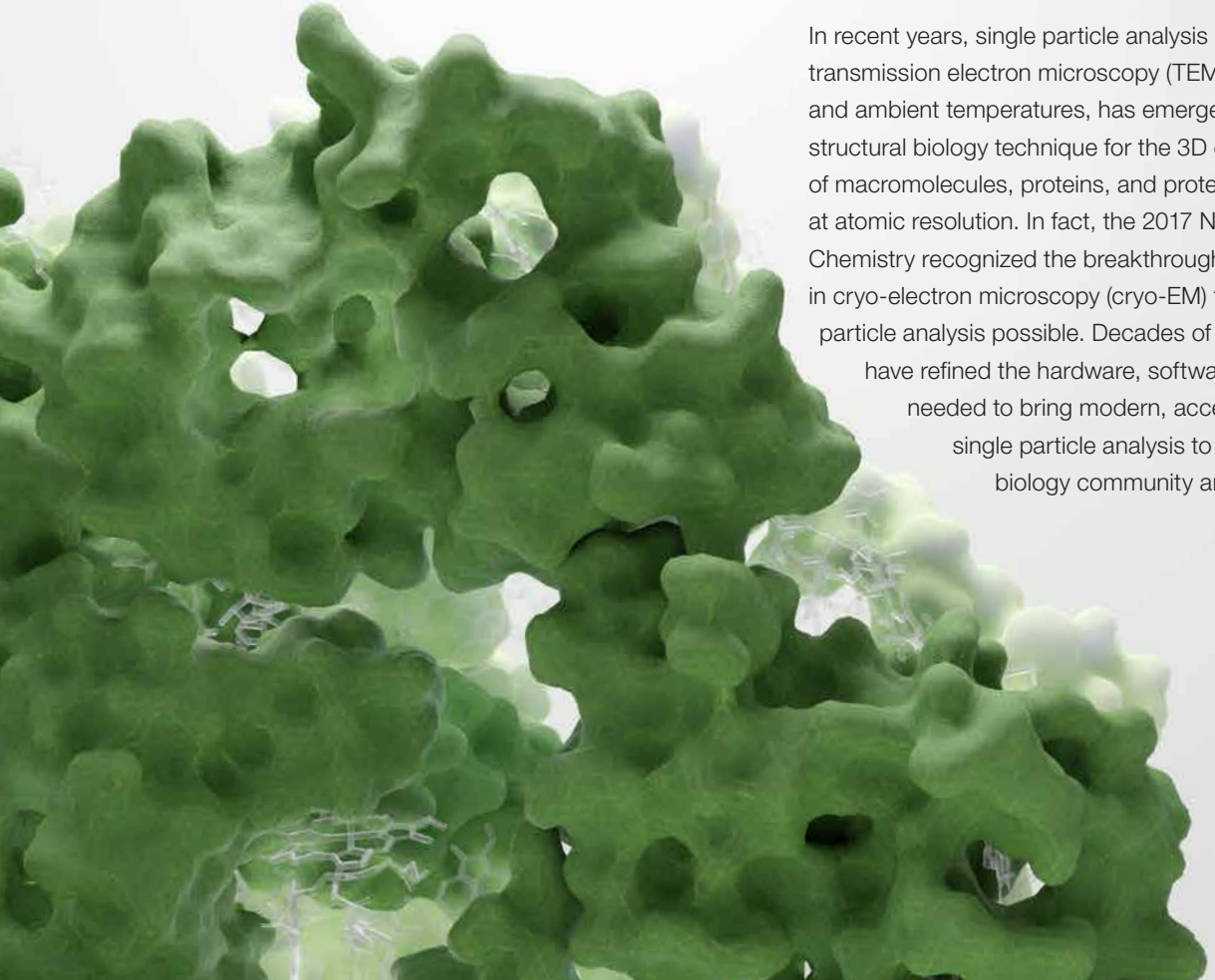
We develop high-end electron microscopes, with key sites located in Eindhoven (NL), Brno (CZ), and Hillsboro, Oregon (US). At these locations, R&D engineers and scientists are trained in all specialties needed to develop electron microscopes and workflows, including physics, mechatronics, electronics, software, and biochemistry. By continually expanding our capabilities and driving innovation, we are helping to advance electron microscopy for the life sciences, enabling unique biological insights, from fundamental research to drug discovery. Notably, recent advances in technology, automation, and artificial intelligence, have made cryo-electron microscopes increasingly easier to use, more affordable, and accessible to the wider scientific community.

In this on-demand webinar "Insights into plant respiration using cryo-EM," Drs. Letts and Maldonado at UC-Davis discussed how they obtained high-resolution cryo-EM structures of respiratory complexes from plants using the Glacios Cryo-TEM, expanding the understanding of respiration across species and generating new mechanistic hypotheses for this core metabolic process. Watch now>

Learn how instruments like the Thermo Scientific Tundra Cryo-TEM broaden the accessibility of cryo-electron microscopy. **Hear from fellow scientists about the Tundra Cryo-TEM.**

Answer your critical questions in the plant sciences with powerful analytical technologies

Plants, algae, and cyanobacteria have complex and unique molecular components compared with other eukaryotic systems. Although gene expression profiling and other techniques have provided useful insights into these structures, advanced microscopy platforms have only recently begun to be utilized to identify 3D cell structures and the function of their related protein, RNA, and DNA components.



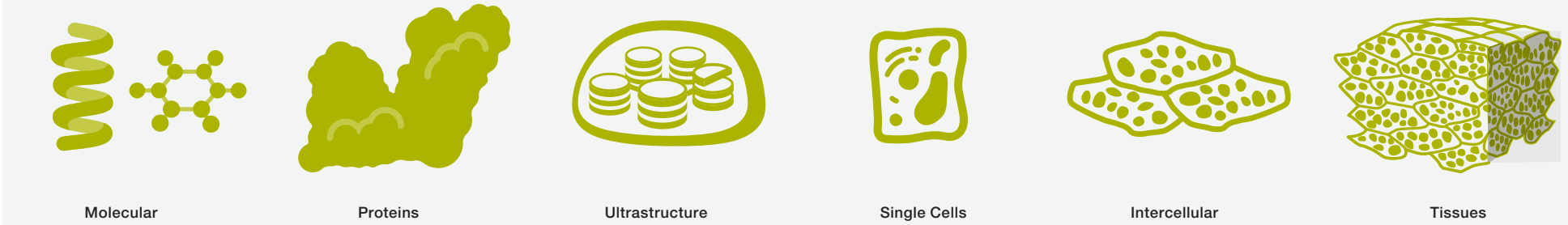
In recent years, single particle analysis (SPA) using transmission electron microscopy (TEM) at both cryogenic and ambient temperatures, has emerged as a mainstream structural biology technique for the 3D characterization of macromolecules, proteins, and protein complexes at atomic resolution. In fact, the 2017 Nobel Prize in Chemistry recognized the breakthrough developments in cryo-electron microscopy (cryo-EM) that made single particle analysis possible. Decades of dedicated work have refined the hardware, software, and automation needed to bring modern, accessible, and reliable single particle analysis to the structural biology community and beyond.

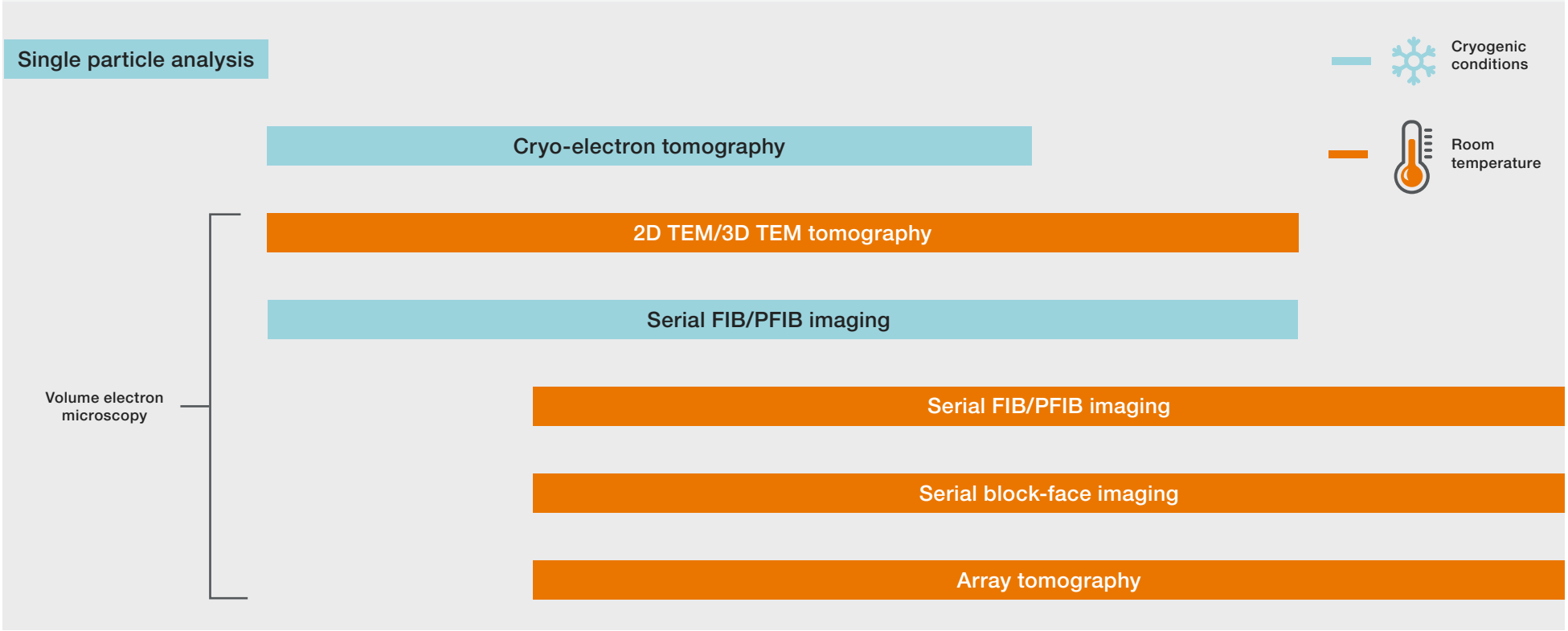
Additionally, cryo-electron tomography has revealed remarkable spatial details of plant tissues, along with their cellular functions. New technologies and improved sample preparation techniques offer a deeper understanding of plant ultrastructure and function. Other recently applied methods include plasma focused ion beam scanning electron microscopy (plasma FIB-SEM), tomography with TEM or scanning transmission electron microscopy (STEM), as well as volume electron microscopy techniques such as serial-section TEM, serial block-face SEM (SBF-SEM), array tomography, etc. This eBook will highlight the impact of these various cutting-edge technologies on our understanding of key areas in the plant sciences, including photosynthesis, respiration, protein translation, host-pathogen interactions, crop sciences, and more.

Learn more:

1. **Plant Structural Biology** https://www.mdpi.com/journal/plants/sections/Plant_Structural_Biology
2. Czymmek, KJ, *et al*. **Realizing the Full Potential of Advanced Microscopy Approaches for Interrogating Plant-Microbe Interactions.** *MPMI* 36:4 p245-255, 2023. doi: 10.1094/MPMI-10-22-0208-FI

EM techniques span multiple length scales





Photosynthesis

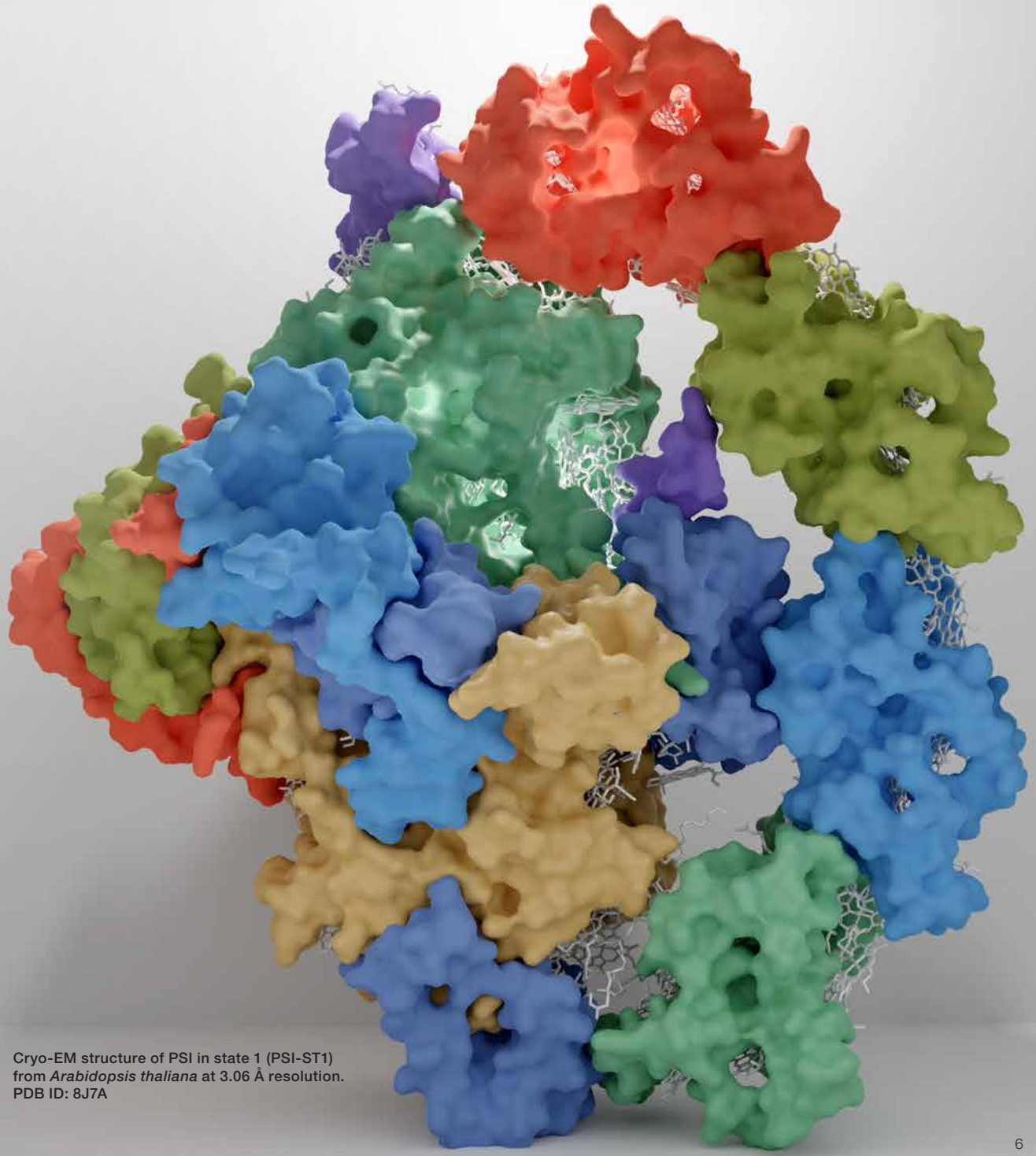
Electron microscopy has served as a valuable tool for identifying and understanding the roles of key proteins and complexes in photosynthetic pathways.

Exploring the impact of changing light conditions on plant photosystems

Plants are known to react to changing environmental light conditions, but the mechanisms behind this response were not fully understood. Wu *et al.* obtained novel structural and functional insights into the mechanisms of light acclimation during state transitions in *Arabidopsis thaliana* using single particle cryo-EM. Previously, these regulatory pathways underlying state transitions were not known for higher plants.

Wu, J, *et al.* **Regulatory dynamics of higher plant PSI-LHCI supercomplex during state transition.** *Molecular Plant* 6:12 p1937–1950, 2023. doi: 10.1016/j.molp.2023.11.002

Workflow: SPA



Cryo-EM structure of PSI in state 1 (PSI-ST1) from *Arabidopsis thaliana* at 3.06 Å resolution. PDB ID: 8J7A

Relying on translocon complexes to import nuclear-encoded proteins

Translocation of critical proteins is a vital but highly intricate process, requiring the interaction of various membrane proteins in complexes or even supercomplexes. Liu *et al.* show how the machinery of the *C. reinhardtii* translocon complexes (the TOC and TIC complexes found in the outer and inner envelope membrane respectively) have evolved to specifically import either photosynthetic or housekeeping proteins from the cytosol into chloroplasts.

Liu, H, *et al.* **Architecture of chloroplast TOC–TIC translocon supercomplex.** *Nature* 615 p349–357, 2023. doi: 10.1038/s41586-023-05744-y

Workflow: SPA

Understanding the role of hydrogen bonds in spectral tuning

Antenna complexes in phototrophic bacteria are responsible for capturing the solar energy needed for photosynthesis. Qian *et al.* compared the structures of these light-harvesting (LH) complexes, revealing altered patterns of hydrogen bonds between LH2 αβ-sidechains and the bacteriochlorin rings. This demonstrates the major role that hydrogen bonds play in the spectral tuning of these bacterial antenna complexes.

Qian, P, *et al.* **Cryo-EM structures of light-harvesting 2 complexes from Rhodopseudomonas palustris reveal the molecular origin of absorption tuning.** *PNAS* 119:43 e2210109119, 2022. doi: 10.1073/pnas.2210109119

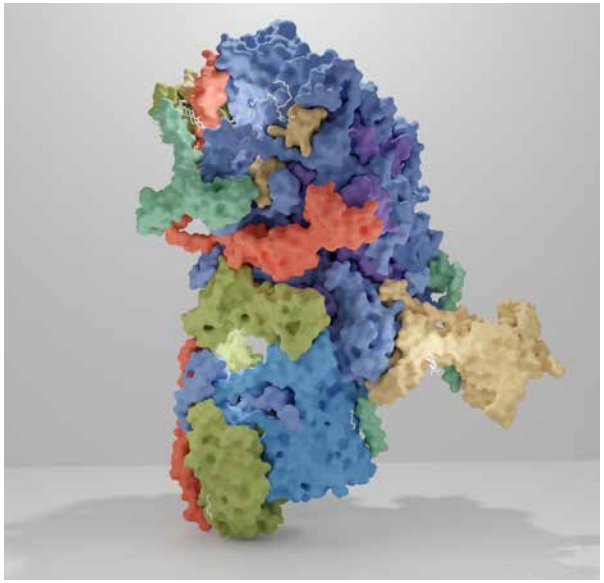
Workflow: SPA

Illuminating light-harvesting mechanisms in cyanobacterium

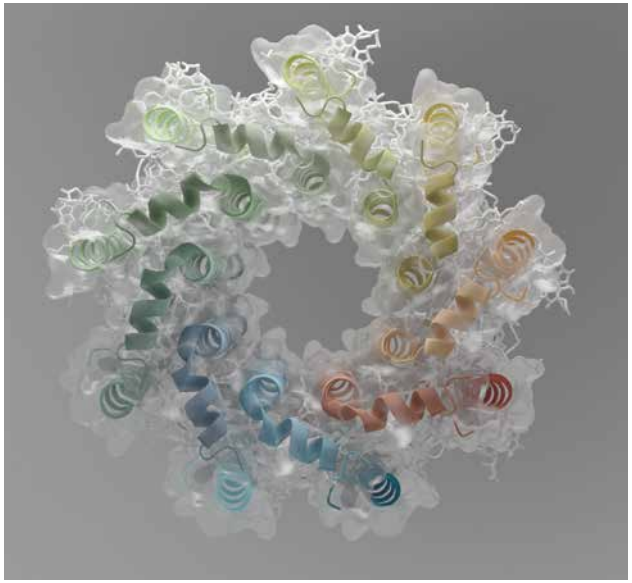
Domínguez-Martín et al. provide detailed insights into the biophysical underpinnings of cyanobacterial light harvesting. The results can help drive further bioengineering of phycobilisome (PBS) protein complexes in natural and artificial light-harvesting systems.

Domínguez-Martín, MA, *et al.* **Structures of a phycobilisome in light-harvesting and photoprotected states.** *Nature* 609 p835–84, 2022. doi.org/10.1038/s41586-022-05156-4

Workflow: SPA



Cryo-EM structure of the TOC-TIC supercomplex from *Chlamydomonas reinhardtii* at 2.77 Å resolution. PDB ID: 7XZI

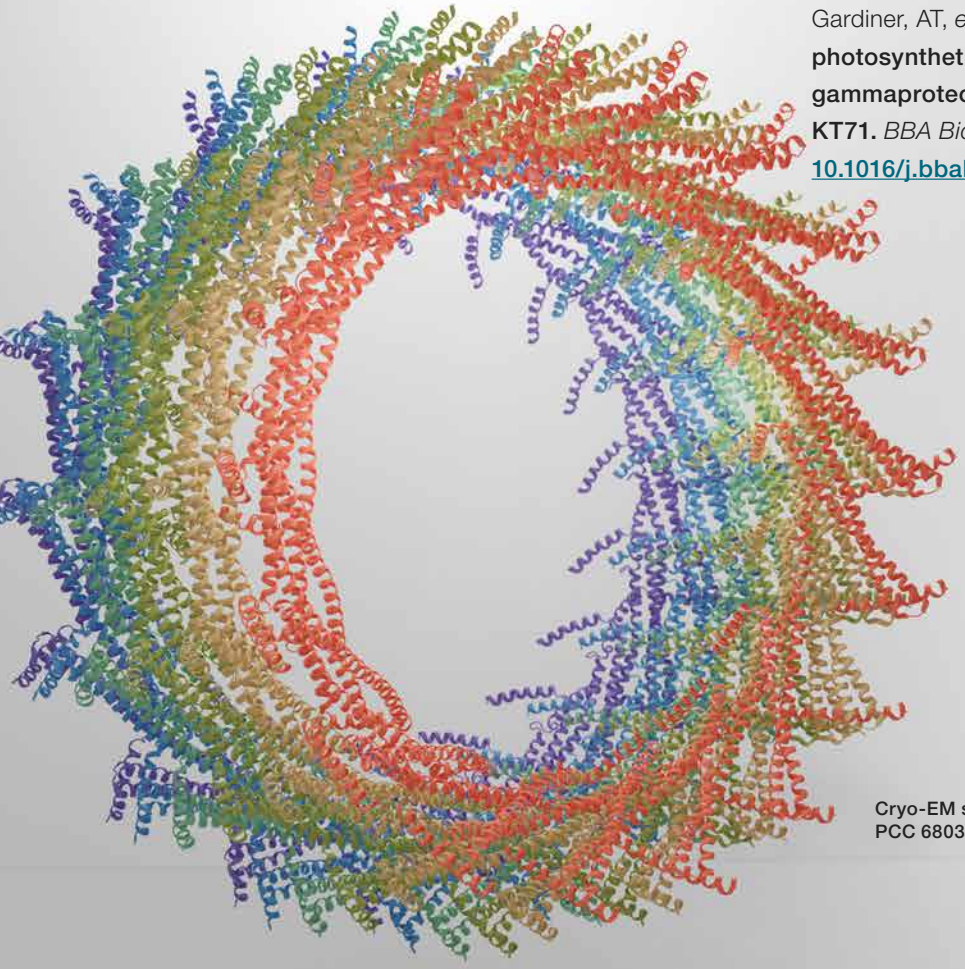


Cryo-EM structure of PucA-LH2 complex from *Rps. palustris* at 2.70 Å resolution. PDB ID: 7ZCU

Cryo-EM structure of phycobilisome (PBS) protein complex from *Synechocystis* PCC 6803 at 2.10 Å resolution. PDB ID: 7SC8.

Protein encapsulation protects thylakoid membrane

Thylakoids are the essential structures in chloroplasts where photosynthesis occurs. Gupta *et al.* studied the vesicle-inducing protein in plastids 1 (VIPP1), which is responsible for thylakoid membrane maintenance, in order to understand how VIPP1 performs this vital function. Using cryogenic correlative light and electron microscopy (cryo-CLEM), they found that VIPP1 forms an oligomeric structure that encapsulates the thylakoid membrane, potentially providing both structure and protection from environmental stresses.



Cryo-EM structure of VIPP1 from *Synechocystis sp.* PCC 6803 at 4.90 Å resolution.

Gupta, TK, *et al.* **Structural basis for VIPP1 oligomerization and maintenance of thylakoid membrane integrity** *Cell* 184:14 p3643–365, 2021. doi: 10.1016/j.cell.2021.05.011

Workflow: SPA

Further reading

Swainsbury, D, *et al*. **The structure and assembly of reaction centre-light-harvesting 1 complexes in photosynthetic bacteria.** *Bioscience Reports* 43:5, 2023. doi: 10.1042/BSR20220089

Gardiner, AT, *et al.* **Characterisation of the photosynthetic complexes from the marine gammaproteobacterium Congregibacter litoralis KT71.** *BBA Bioenergetics* 1864:2 148946, 2023. doi: 10.1016/j.bbabio.2022.148946

Cupellini, L, *et al.* **Quantum chemical elucidation of a sevenfold symmetric bacterial antenna complex.** *Photosynth Res* 156 p75–87, 2023. doi: 10.1007/s11120-022-00925-8

Naschberger, A, *et al.* **Algal photosystem I dimer and high-resolution model of PSI-plastocyanin complex.** *Nature Plants* 8 p1191–1201, 2022. doi: 10.1038/s41477-022-01253-4

Seki, S, *et al.* **Structural insights into blue-green light utilization by marine green algal light harvesting complex II at 2.78 Å.** *BBA Advances* 2 100064, 2022. doi: 10.1016/j.bbadva.2022.100064

Sutherland, GA, *et al.* **Engineering purple bacterial carotenoid biosynthesis to study the roles of carotenoids in light-harvesting complexes.** *Methods in Enzymology* 674 p137–184, 2022. doi: 10.1016/bs.mie.2022.04.001

Qian, P, *et al.* **2.4-Å structure of the double-ring Gemmatimonas phototrophica photosystem.** *Science Advances* 8:7, 2022. doi: 10.1126/sciadv.abk3139

Wahlgren, WY, *et al.* **Cryo-Electron Microscopy of Arabidopsis thaliana Phytochrome A in Its Pr State Reveals Head-to-Head Homodimeric Architecture.** *Front Plant Sci* 12, 2021. doi: 10.3389/fpls.2021.663751

Deconvoluting fundamental processes in plant biology

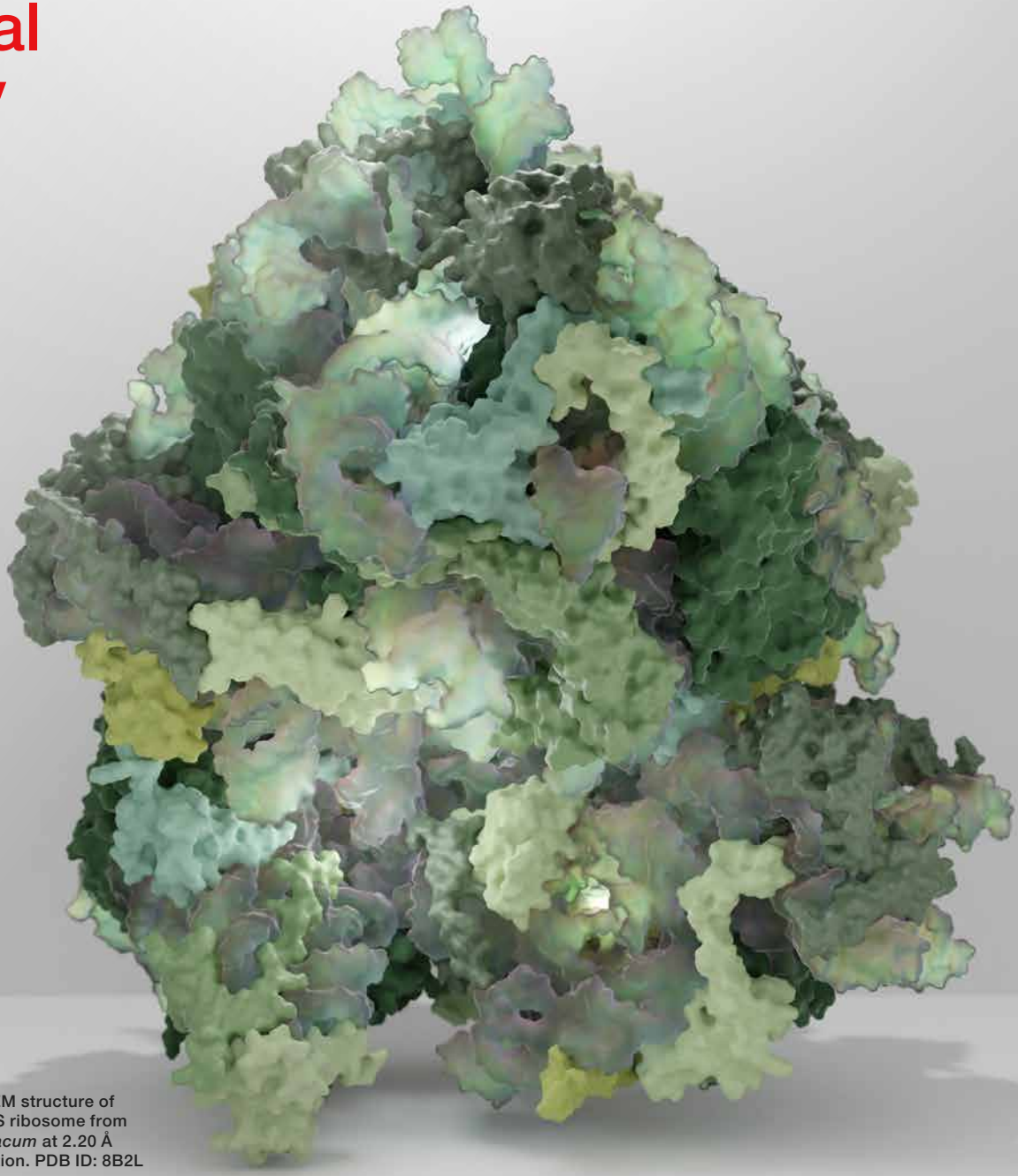
Compared to other eukaryotic organisms, plants utilize a number of unique processes and mechanisms, including those for transcription, translation, and respiration. Until recently, the structural details for some of these processes, including the structure of RNA polymerase as well as ribosomal and respiratory complexes, have remained unknown. By obtaining high-resolution cryo-EM structures of these complexes, scientists have expanded our understanding of these processes and generated new mechanistic hypotheses.

Characterizing structures of the 80S ribosomes in tobacco

Ribosome structures can elucidate how translation occurs in various areas of the plant cell. Smirnova *et al.* obtained high-resolution structures of the *Nicotiana tabacum* 80S ribosome in the cytosol during translation. This structure, obtained at 2.2 Å resolution, offered insights into the fundamental mechanisms of cytosolic translation in plants.

Smirnova, J, *et al.* **Structure of the actively translating plant 80S ribosome at 2.2 Å resolution.** *Nature Plant*s 9 p987–1000, 2023. doi: 10.1038/s41477-023-01407-y

Workflow: SPA



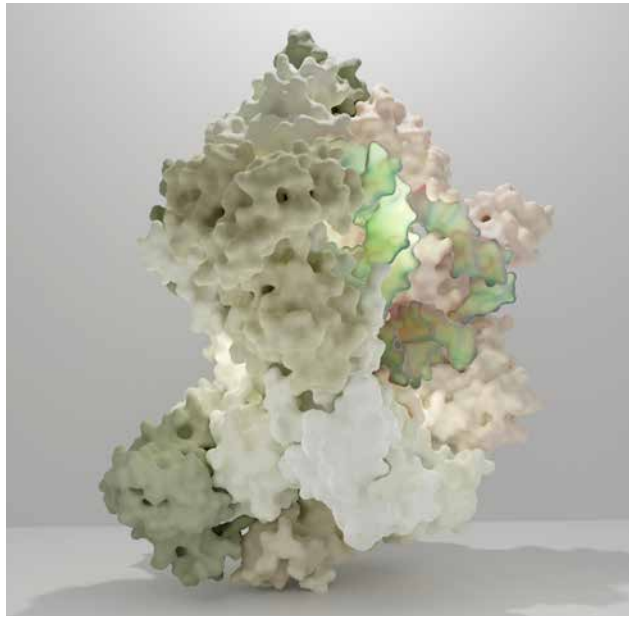
Cryo-EM structure of the 80S ribosome from *N. tabacum* at 2.20 Å resolution. PDB ID: 8B2L

Identifying a unique RNA polymerase in cauliflower

Plants have a unique RNA-directed DNA methylation pathway that was not fully understood. This process is mediated by DNA-dependent RNA polymerases, which are part of a family that transmits genetic information from DNA to RNA. Xie *et al.* visualized a plant-specific polymerase, Pol V, in cauliflower (*Brassica oleracea*) that is involved in this pathway. This structural information provides additional insights into the transcription processes of plants.

Xie, G, *et al.* **Structure and mechanism of the plant RNA polymerase V.** *Science* 379:6638, 2023. doi: 10.1126/science.adf8231

Workflow: SPA



Cryo-EM structure of RNA polymerase V elongation complex from *B. oleracea* at 2.73 Å resolution. PDB ID: 8HIM

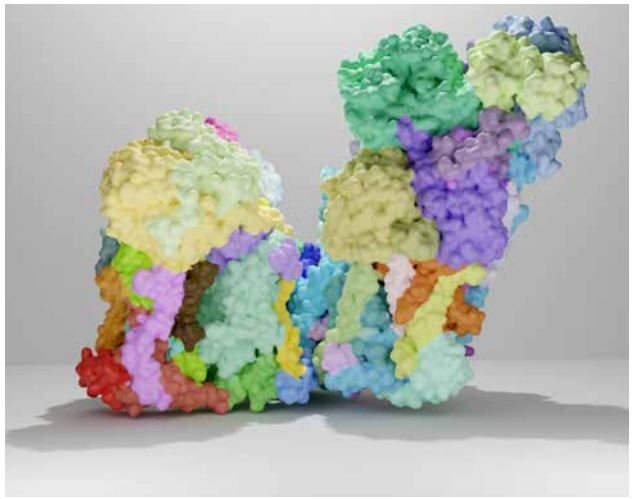
Exploring cellular respiration in different plant species

Oxidative phosphorylation (OXPHOS) is the final step in the cellular respiration of plants and is carried out by protein complexes in the inner mitochondrial membrane. With cryo-EM structures, the physiological function of supercomplexes involved in the OXPHOS electron transport chain can be deconvoluted. This structural analysis provides critical details toward our understanding of cellular respiration in plants.

Maldonado *et al.* identified the respiratory supercomplex I + III2 in mung beans (*Vigna radiata*) at 3.2 Å resolution, revealing that the supercomplex interfaces are plant specific.

Maldonado, M, *et al*. **Plant-specific features of respiratory supercomplex I + III2 from Vigna radiata.** *Nature Plants* 9 p157–168, 2023. doi: 10.1038/s41477-022-01306-8

Workflow: SPA



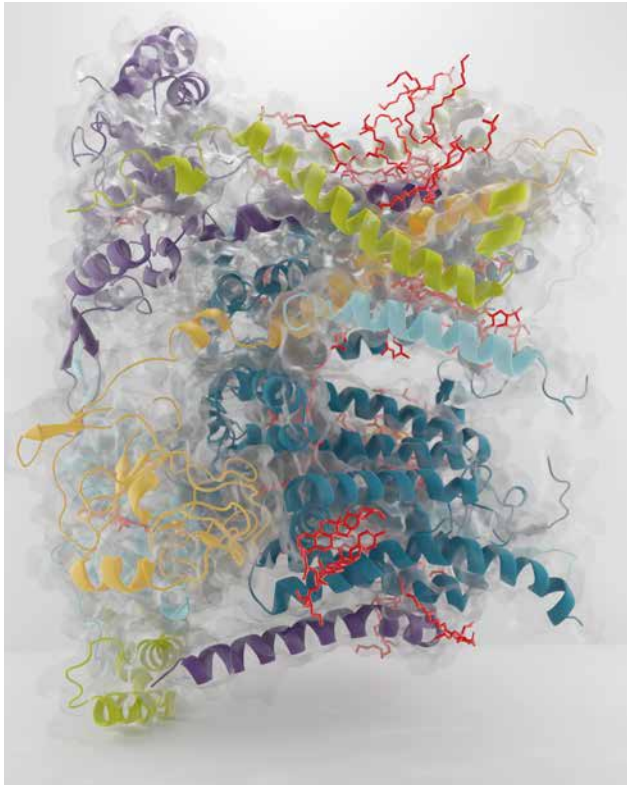
Cryo-EM structure of respiratory supercomplex I+III2 (full bridge) from *V. radiata* at 3.20 Å. PDB ID: 8E73

Arabidopsis supercomplex assembly

Structural analysis with cryo-EM enabled Klusch *et al.* to determine the structure of the Arabidopsis I + III2 supercomplex. They gained new insights, not only into the function of the individual supercomplex components at near-atomic detail, but also into supercomplex assembly.

Klusch, N *et al.* **Cryo-EM structure of the respiratory I + III2 supercomplex from Arabidopsis thaliana at 2 Å resolution.** *Nature Plants* 9, p142–156, 2023. doi: 10.1038/s41477-022-01308-6

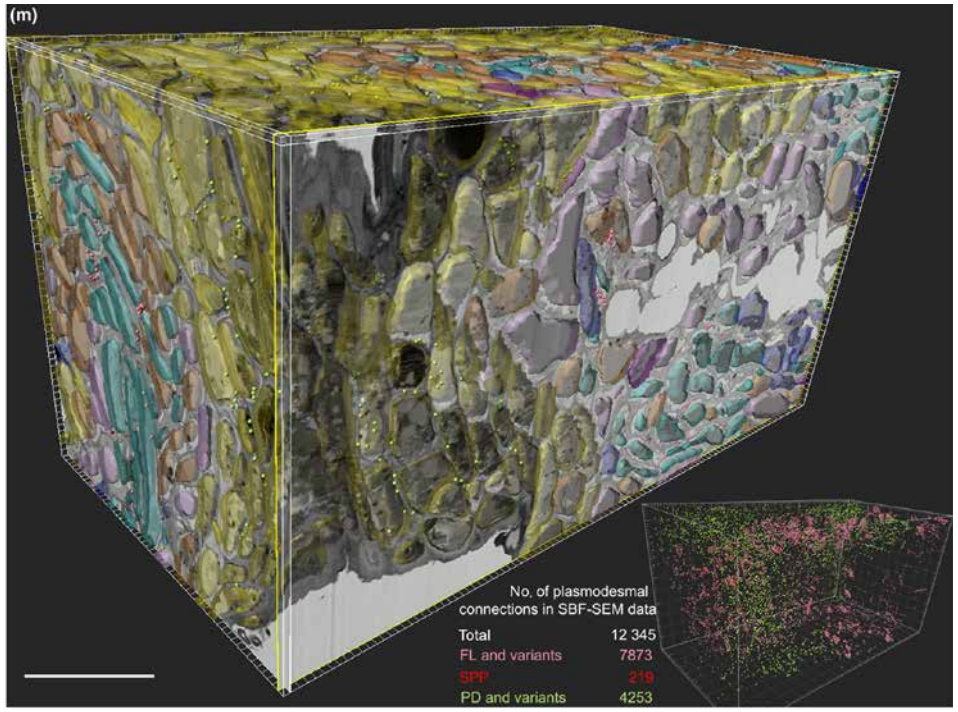
Workflow: SPA



Cryo-EM structure of the respiratory I + III2 supercomplex from *A. thaliana* at 2.25 Å resolution. PDB ID: 8BEL

Host-pathogen interactions

Infectious plant diseases are caused primarily by pathogenic organisms such as fungi, bacteria, and viruses. Driven by climate change, these pathogens are on the rise and are a growing threat to global agriculture and food security. Alongside genomic, proteomic, biochemical, and other analyses of plant-pathogen interactions, structural studies of virulent pathogenic proteins, as well as plant immune receptors, have uncovered key insights into the functions of these molecules.[1, 2]



Overview of all plasmodesmal connections in the phloem region of a plant tumor. Figure reproduced under CC BY-ND 4.0.

Limiting virus movement within the phloem

A key component of viral infection is virion movement between cells. Lv *et al.* used high-throughput volume electron microscopy to show that sieve plate pores and flexible gateways of the phloem had a sufficiently large size exclusion limit (SEL) to accommodate virions and potentially serve as pathways for virion movement. A working model was proposed to demonstrate the mechanism underlying the limitations of virus movement within the phloem.

Lv, M, *et al.* **Volume electron microscopy reconstruction uncovers a physical barrier that limits virus to phloem.** *New Phytol.* 241 p343–362, 2024. **doi: 10.1111/nph.19319**

Workflow: SBF-SEM, FIB-SEM

Gaining direct structural visualization of the viral life cycle in tobacco

FIB-SEM tomography offers unprecedented views into the interior of cells, including the behavior and life cycles of the viruses within them. Guo *et al.* visualized the ultrastructural details of tomato spotted wilt virus (TSWV) in *Nicotiana rustica* cells using FIB-SEM. They were able to see novel connections, including those in the cell wall, or wrapped by the cell membranes of neighboring cells. These could not be visualized using other EM methods.

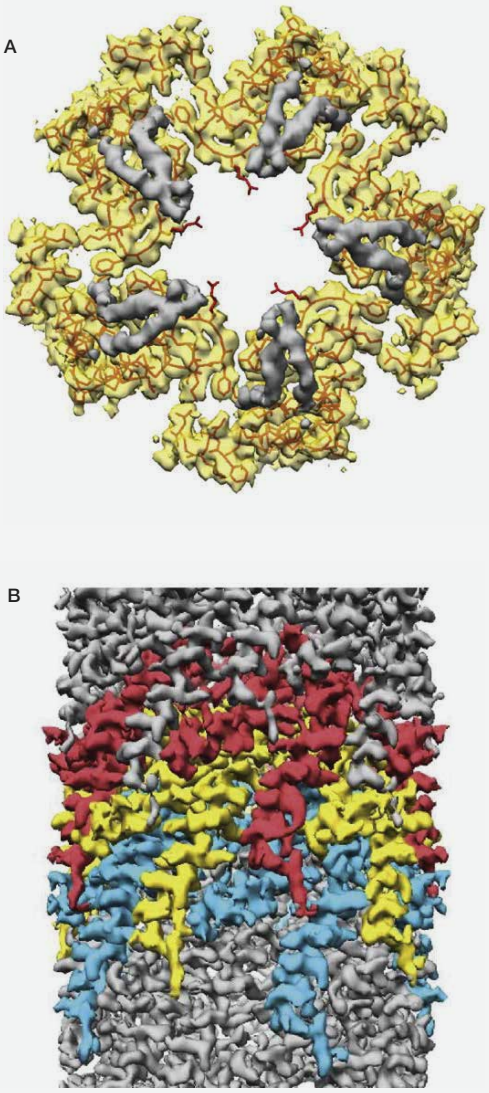
Guo, J, *et al.* **Three-dimensional analysis of membrane structures associated with tomato spotted wilt virus infection.** *Plant, Cell & Environment* 46 p650–664, 2023. **doi: 10.1111/pce.14511**

Workflow: FIB-SEM, 2D TEM

Learn more:

1. Wirthmueller, L, *et al.* **On the front line: structural insights into plant–pathogen interactions.** *Nature Reviews Microbiology* 11 p761–776, 2013. **doi: 10.1038/nrmicro3118**

2. Outram, MA, *et al.* **Seeing is believing: Exploiting advances in structural biology to understand and engineer plant immunity.** *Current Opinion in Plant Biology* 67:102210, 2022. **doi: 10.1016/j.pbi.2022.102210**



The T-pilus composed of VirB2 and PG-phospholipid. A) Map density of a helical layer as seen from the top. B) The filament seen from the side with three layers of VirB2. Figure reproduced under CC BY-ND 4.0.

Understanding how membrane-less organelles control cellular activities

Membrane-less organelles have been an area of growing interest due to their ability to phase-separate and compartmentalize cellular activities. Huang *et al.* used liquid–liquid phase separation (LLPS) to identify a family of nuclear guanylate-binding protein (GBP)-like GTPases (GBPLs) that protect against infection and autoimmunity. Uncovering these new LLPS-dependent resistance pathways and GBPL defense-activated condensates (GDACs) has implications for crop management and food production in the face of climate change.

Huang, S, *et al.* **A phase-separated nuclear GBPL circuit controls immunity in plants.** *Nature* 594 p424–429, 2021. doi: 10.1038/s41586-021-03572-6

Workflow: SPA

Uncovering the structure of the filament behind gall disease

While gall disease is widely known to produce abnormal tissue growth on plants, the molecular behavior of these growths is poorly understood. Kreida *et al.* identified the structure of T-pilus in *A. tumefaciens*, an extracellular filament involved in the mating pair formation between the bacterium and the recipient plant cell. The assembled T-pilus, composed of VirB2 proteins and PG, is proposed to be a conduit for the ssDNA transfer that leads to the disease.

Kreida, S, *et al.* **Cryo-EM structure of the Agrobacterium tumefaciens type IV secretion system-associated T-pilus reveals stoichiometric protein-phospholipid assembly.** *Structure* 31 p385–394, 2023. doi: 10.1016/j.str.2023.02.005

Workflow: SPA

Further reading

Wang, W, *et al.* **WeiTsing, a pericycle-expressed ion channel, safeguards the stele to confer clubroot resistance.** *Cell* 186:12 p2656–2671.e18. doi: 10.1016/j.cell.2023.05.023

Sun, Y, *et al.* **Plant receptor-like protein activation by a microbial glycoside hydrolase.** *Nature* 610 p335–342, 2022. doi: 10.1038/s41586-022-05214-x

Lecorre, F, *et al.* **The cryo-electron microscopy structure of Broad Bean Stain Virus suggests a common capsid assembly mechanism among comoviruses.** *Virology* 530 p75–84, 2019. doi: 10.1016/j.virol.2019.02.009

Hesketh, EL, *et al.* **The 3.3 Å structure of a plant geminivirus using cryo-EM.** *Nature Communications* 9:2369, 2018. doi: 10.1038/s41467-018-04793-6

Agirrezabala, X, *et al.* **The near-atomic cryoEM structure of a flexible filamentous plant virus shows homology of its coat protein with nucleoproteins of animal viruses.** *eLife* 4:e11795, 2015. doi: 10.7554/eLife.11795

Crop sciences

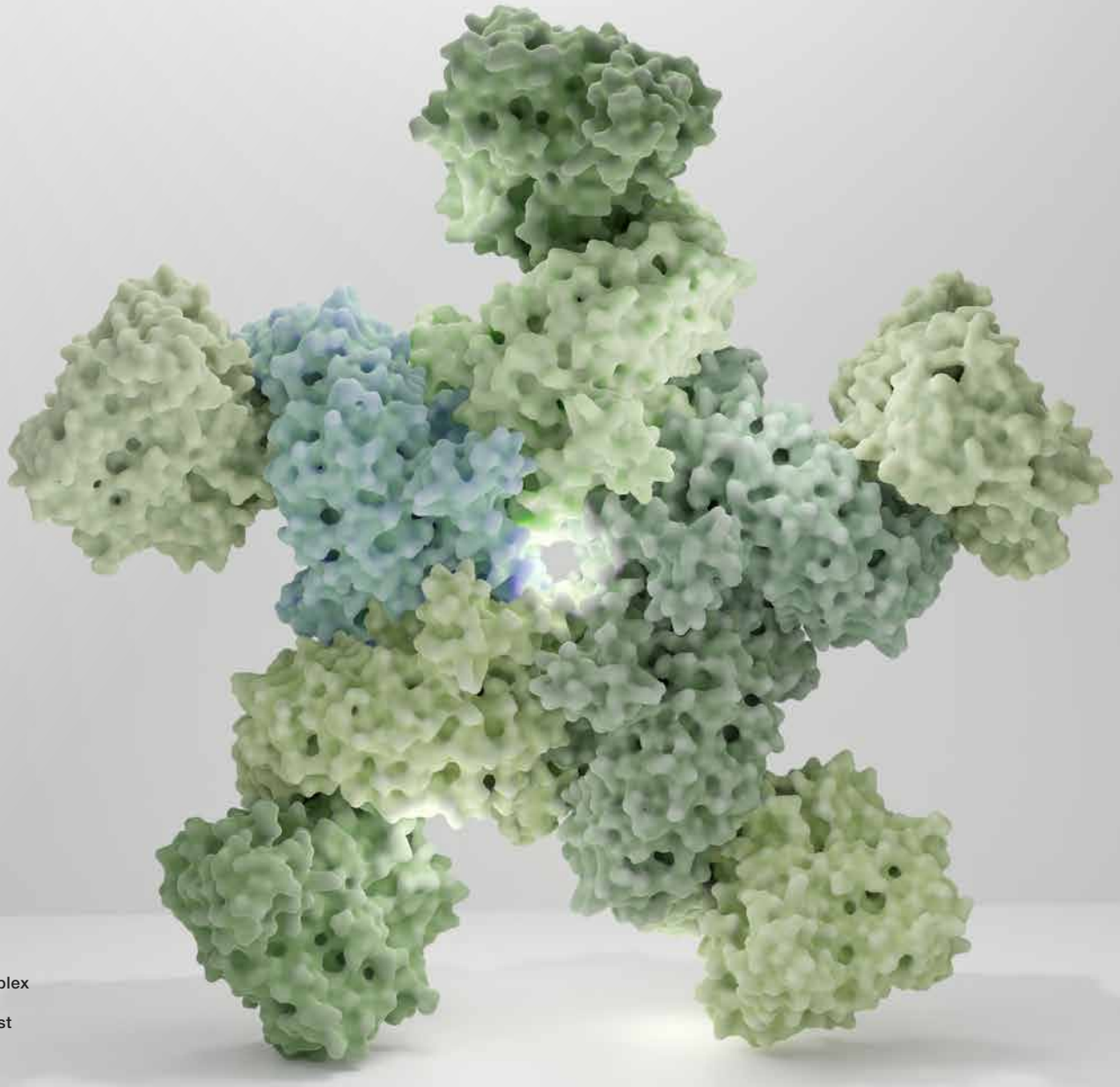
Crop sciences encompass a range of disciplines, including agronomy, physiology, and genetics, in the pursuit of increasing food security as well as environmental protection. This research impacts a number of agricultural areas such as plant breeding and crop production, with a recent focus on climate change and the mitigation of threats to environmental and nutritional security.

Harnessing resistosomes for crop improvement

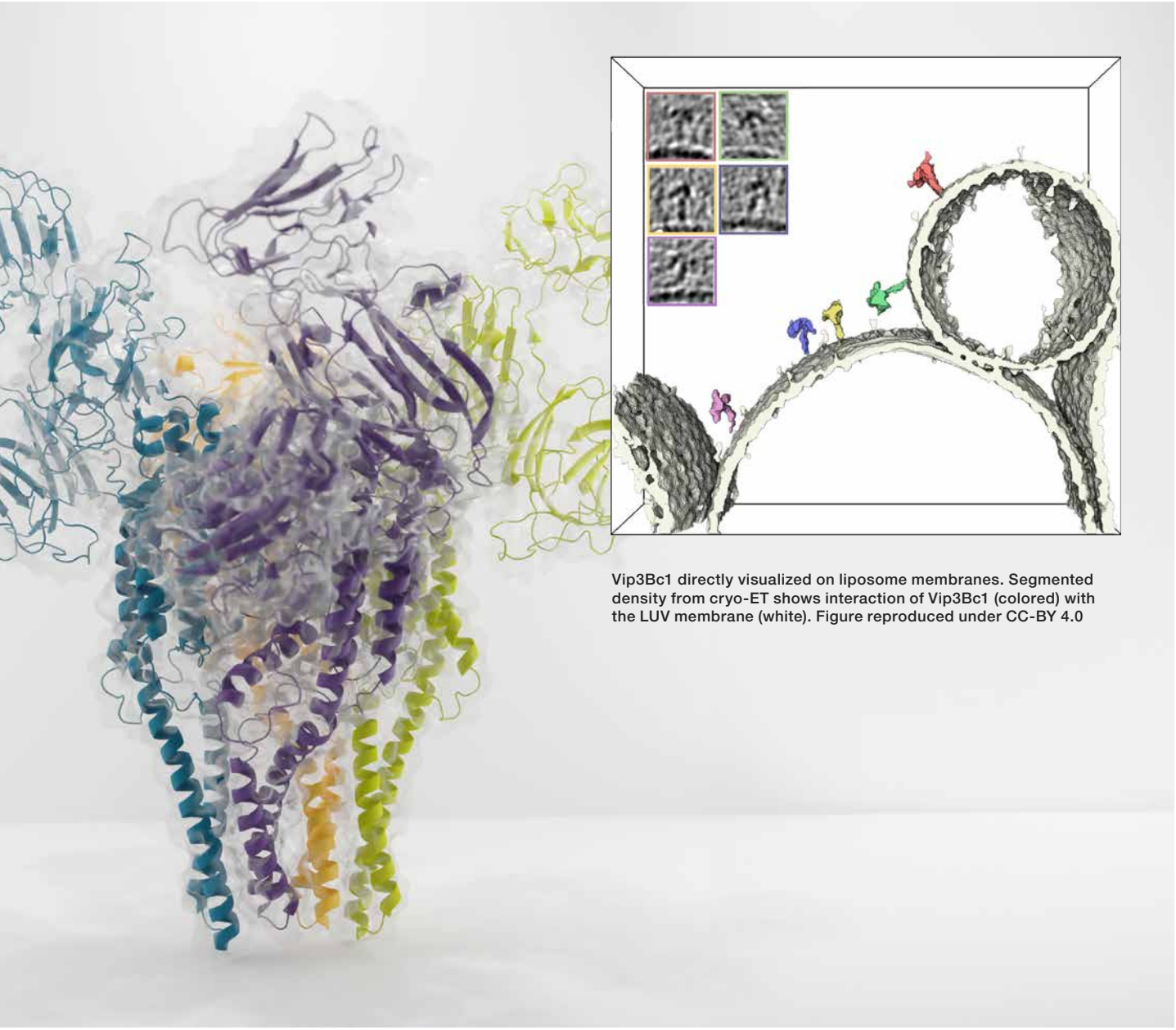
Resistosomes are the active, oligomeric state of a family of plant receptors, formed when the proteins bind a pathogen's effector and trigger an immune response. Harnessing the power of these nucleotide-binding leucine-rich repeat receptors (NLRs) through structural engineering could provide a novel pathway to better pathogen-resistant crops. NLRs bound to fungal wheat stem rust were captured with cryo-EM by Förderer *et al.*, providing a proof of principle for the structural characterization and engineering of NLRs.

Förderer, A, *et al.* **A wheat resistosome defines common principles of immune receptor channels.** *Nature* 610 p532–539, 2022. doi: 10.1038/s41586-022-05231-w

Workflow: SPA



Cryo-EM structure of oligomeric complex formed by wheat CNL Sr35 and the effector AvrSr35 of the wheat stem rust pathogen at 3.00 Å.PDB ID: 7XC2



Cryo-EM structure of the Vip3Bc1 tetramer *B. thuringiensis* at 3.9 Å resolution.
PDB ID: 6YRF

Vip3Bc1 directly visualized on liposome membranes. Segmented density from cryo-ET shows interaction of Vip3Bc1 (colored) with the LUV membrane (white). Figure reproduced under CC-BY 4.0

Revealing the insecticidal activity of Vip3

Insects have been a major threat to crop production since the dawn of agriculture. Cutting-edge methods of pest control include the production of transgenic plants that create their own insecticidal proteins, such as vegetative insecticidal protein 3 (Vip3). Byrne *et al.* used cryo-EM and cryo-ET methods to gain insight into the architecture and mechanism of Vip3 activation and toxicity.

Byrne, MJ, *et al.* **Cryo-EM structures of an insecticidal Bt toxin reveal its mechanism of action on the membrane.** *Nat Commun* 12:2791, 2021. doi: 10.1038/s41467-021-23146-4

Workflow: Cryo-ET, SPA

Adapting to abiotic stresses such as drought

Li *et al.* identified a new mechanism underlying a distinct class of ER-derived giant coat protein complex II (COPII). They found that COPII vesicles formed to modulate endomembrane trafficking of stress-regulated channels or transporters to adapt to abiotic stresses, shedding light on the evolutionary importance of gene duplication in plant development.

Li, B, *et al.* **A distinct giant coat protein complex II vesicle population in Arabidopsis thaliana.** *Nature Plants* 7, p1335–1346, 2021. doi: 10.1038/s41477-021-00997-9

Workflow: Cryo-ET, SPA

Method development

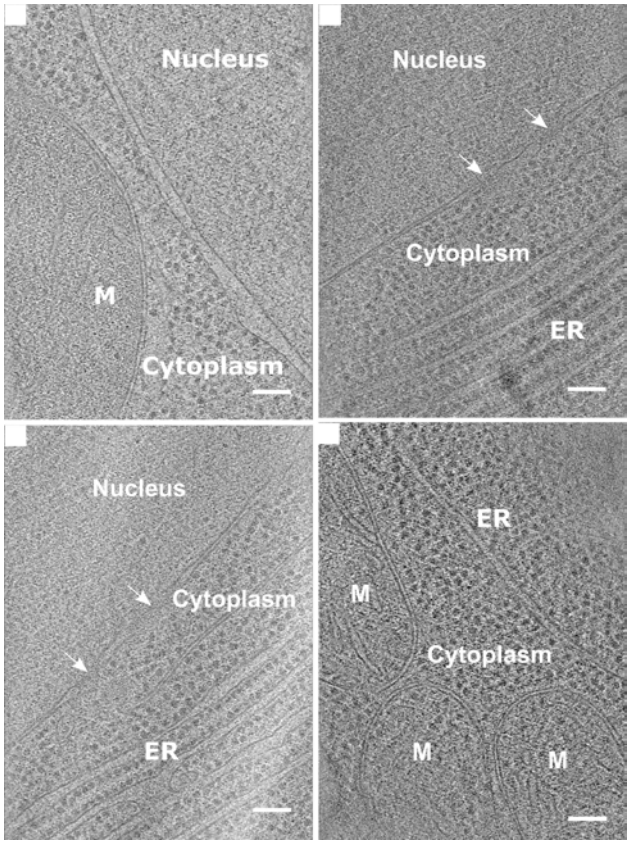
Both transmission (TEM) and scanning (SEM) electron microscopy are essential tools for visualizing plant proteins and structures, enabling critical insights into how plants function. Room temperature methods have provided valuable details related to plant ultrastructure, while cryo-electron microscopy methods like single particle analysis and cryo-electron tomography (cryo-ET) have helped to identify the roles of single proteins, complexes, and biological pathways.

Preparing plant protoplasts for cryo-ET

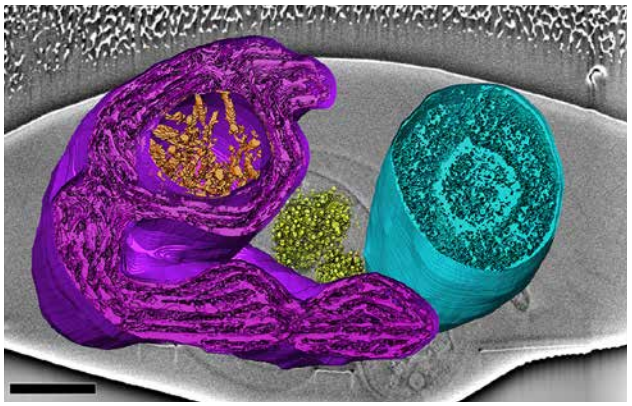
Sanchez Carrillo *et al.* describe a workflow to prepare root protoplasts from *A. thaliana* for cryo-ET. It includes protoplast isolation and vitrification on EM grids followed by cryo-focused ion beam (cryo-FIB) milling, with the aim of tilt series acquisition. Their protocol provides a novel use of plant protoplasts as a tool for cryo-ET of transient gene expression.

Sanchez Carrillo, IB, *et al.* **Preparing Arabidopsis thaliana root protoplasts for cryo electron tomography.** *Front. Plant Sci* 14, 2023. doi: 10.3389/fpls.2023.1261180

Workflow: Cryo-ET, cryo-FIB-SEM



Representative tomograms from vitrified and milled root protoplasts of *Arabidopsis thaliana*. Lamella thickness: ~200 nm or less. Scale bars: 100 nm. Figure reproduced under CC-BY 4.0



3D rendering of *chlamydomonas* organelles, showing the chloroplast and its thylakoid membranes (purple), pyrenoid tubules (orange), Golgi (yellow), and nucleus (cyan). Scale bars = 1 µm. Dataset acquired on a Thermo Scientific™ Helios Hydra™ Plasma FIB-SEM. Figure reproduced under CC BY 4.0.

Investigating plant-microbe interactions with different EM methods

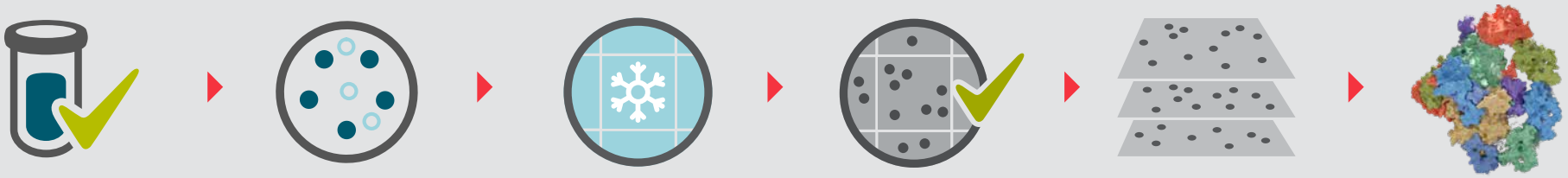
Czymmek *et al.* describe a range of routinely performed microscopy techniques for the exploration of cell biology in plants and other organisms. They highlight emerging room-temperature and cryo-EM technologies that have a particular potential to expand our understanding of plant structure and function as well as plant-pathogen interactions.

Czymmek, KJ, *et al.* **Realizing the Full Potential of Advanced Microscopy Approaches for Interrogating Plant-Microbe Interactions.** *MPMI* 36:4 p245-255, 2023. doi: 10.1094/MPMI-10-22-0208-FI

Workflow: Cryo-ET, cryo-FIB-SEM

Single particle analysis

Single particle analysis (SPA) is a well-established cryo-EM technique that has enabled the near-atomic structural determination of challenging proteins and protein complexes, without the need for crystallization. Samples can be studied directly in solution. High-quality data collection from cryo-EM has been facilitated by recent advances in sample preparation and data processing.



Sample preparation

The quality of structural analysis is directly related to sample preparation: purified, homogeneous, and biochemically active proteins/macromolecules in a stable buffer typically provide the best results.

Negative-stain screening

Negative-stain electron microscopy is an easy and cost-effective method for the quality assessment of purified biological specimens at room temperature. This screening allows you to qualitatively assess particle composition and conformational homogeneity, which can only be done at the microscopic scale.

Vitrification

Once sample purity has been verified, the sample is vitrified (i.e., rapidly frozen to suspend the specimens in a layer of amorphous (vitreous) ice). By avoiding ice crystallization, the samples are preserved in a near-native state, essentially taking a snapshot of their structures in solution. Ice consistency as well as sample distribution and orientation are critical for data collection, and automated plunge freezing is the general method of choice for consistent sample vitrification.

Cryo-EM grid screening

Even the best vitrification system is not 100% consistent, and therefore the sample (frozen atop an EM grid) must be screened in order to find optimal areas for data collection. Ideally, the ice would uniformly cover the grid holes, and a large amount of specimen is distributed evenly throughout the visible ice. Only a moderate-resolution TEM scan is required at this stage, as this is a largely qualitative assessment.

Data acquisition

Data collection consists of high-resolution imaging with a specifically designed cryo-TEM. With advances in data collection software, individual particles can be automatically identified in the TEM image and grouped according to particle orientation. For every sample, imaging and identification can be simplified by robust, reliable automation.

Structure visualization

Once sufficient particle data is collected (ideally representing the sample from as many different orientations as possible), it can be recombined into a 3D representation of the protein/macromolecule. This uses 2D data from tens of thousands of particles and typically involves multiple data processing steps, requiring high data storage capacity and computational power. A number of professionally developed and open-source data processing solutions exist to simplify and expedite this process.

Learn more at **thermofisher.com/spa**

Sample preparation and vitrification for single particle analysis

Explore our integrated solutions and support for the entire single particle analysis workflow, from sample preparation to data analysis.

Optimizing your sample preparation

Thermo Scientific™ VitroEase™ products are specifically designed to facilitate cryo-EM sample preparation, helping you optimize your samples and prepare grids that will yield high-resolution cryo-EM structures. You can optimize samples with the **VitroEase Buffer Screening Kit** and use the **VitroEase Apoferritin Standard** to validate workflows. The **VitroEase Cryo-EM Training Kit** facilitates training for the next generation of cryo-EM users.

The **VitroEase Methylamine Vanadate** and **Methylamine Tungstate Negative Stains** are ready-to-use solutions that are designed to help you prepare samples for negative stain assessment. Often, this is done on a simple side-entry microscope (e.g., **Thermo Scientific™ Talos™ L120C (S)TEM**), since screening is usually done one grid at a time, and the actual time spent on the microscope is short.

Simplifying the vitrification process

The entire vitrification procedure can be simplified using semi-automated plungers such as the **Thermo Scientific™ Vitrobot™ System**, which can be combined with the **VitroEase Cryo-EM Training Kit** to help anyone learn the intricacies of the vitrification process.

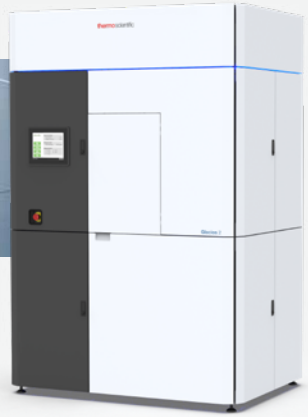
Cryo-transmission electron microscopes

Thermo Fisher Scientific offers a range of cryo-electron microscopy instruments suited to a variety of analytical needs. With the Thermo Scientific™ Tundra™ Cryo-TEM, you can expand the possibilities of your biochemical research without prior microscopy experience and at a more affordable price point. This offers your laboratory a cost-effective, easier-to-use cryo-EM solution optimized for single particle analysis. The Thermo Scientific™ Glacios™ 2 and Krios™ G4 Cryo-TEMs are capable of producing higher resolution results and have the ability to perform additional cryo-EM methods such as MicroED and cryo-electron tomography.

In addition, our solution for 60°C heat decontamination allows the Krios Cryo-TEM to be installed in higher biosafety-level containment facilities (e.g. BSL-3).

See a full demo of the Tundra Cryo-TEM in action

Watch to learn more about the Glacios 2 Cryo-TEM



Watch to learn more about the Krios G4 Cryo-TEM

Tundra Cryo-TEM: accessible & smart

- Fully automated and requires minimal expertise to use
- Cost-effective platform for labs that are new to cryo-EM
- Ideal for sample optimization for analysis on higher resolution platforms

Intermediate-resolution SPA	100 kV, <3.5 Å*
Medium throughput	dataset in 24 hours
Sample type	proteins, macromolecules
Applications	SPA

Glacios 2 Cryo-TEM: powerful & versatile

- Automated sample assessment and acquisition of large data sets for higher throughput
- Improved detector and AI-enabled software work together to provide rapid, high-quality results

High-resolution SPA	200 kV, <2.5 Å*
High throughput	dataset in 30 minutes
Sample type	proteins, crystals, cells, macromolecules
Applications	SPA, MicroED, tomography

Krios G4 Cryo-TEM: Unparallel performance

- Designed for true atomic-resolution cryo-EM and speed
- Highest level of automation from sample vitrification to data analysis

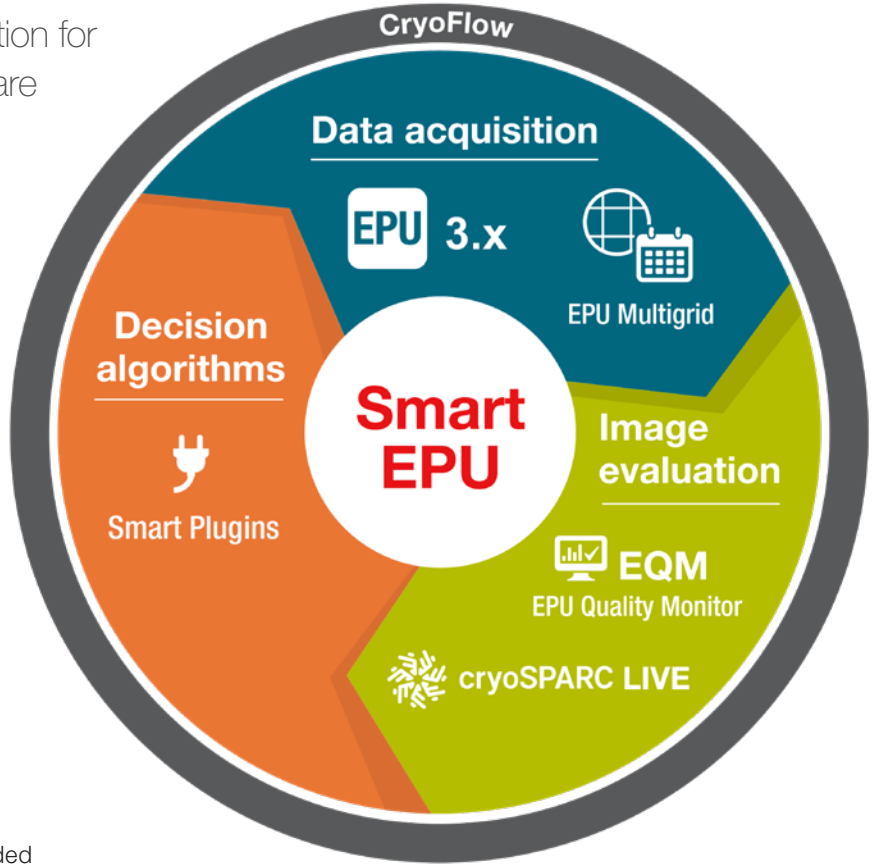
Ultra-high-resolution SPA	300 kV, <1.5 Å*
Highest throughput	dataset in minutes
Sample type	proteins, crystals, cells, macromolecules
Applications	SPA, MicroED, tomography

* Based on best published performance, actual results will depend on non-microscope factors such as sample and user experience. Not a promise of biological resolution performance.

Learn more at **thermofisher.com/cryoem**

Software for single particle analysis

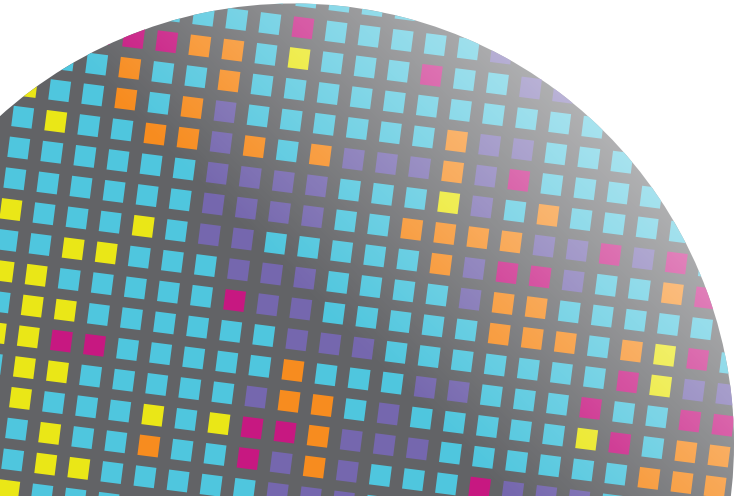
Thermo Scientific Smart EPU Software is an innovative and easy-to-use solution for streamlined and accurate single particle analysis data acquisition. This software suite combines automation and user guidance to increase throughput and deliver reproducible results. With Smart EPU Software, you can make efficient use of the microscope and focus more on your research.



Smart EPU Software is comprised of interactive components that automate multiple image acquisition steps, as well as AI-driven technology that helps you make decisions, including:

- The user-friendly **EPU 3 Interface**, which helps you set up experiments quickly and easily, regardless of experience level
- **EPU Multigrid**, which sets up a queue of automated acquisitions across multiple grids to maximize efficiency
- **EPU Quality Monitor** analyzes images in real-time to determine the quality of your data as it is generated
- **Embedded CryoSPARC Live™** processes images on the fly to assess sample quality and accelerate structural determination
- **Smart Plugins** automatically adjust data collection settings based on output from real-time image analysis
- **Thermo Scientific™ CryoFlow™ Software** provides easy access to your results through a web portal, allowing you to manage your data and reporting anywhere

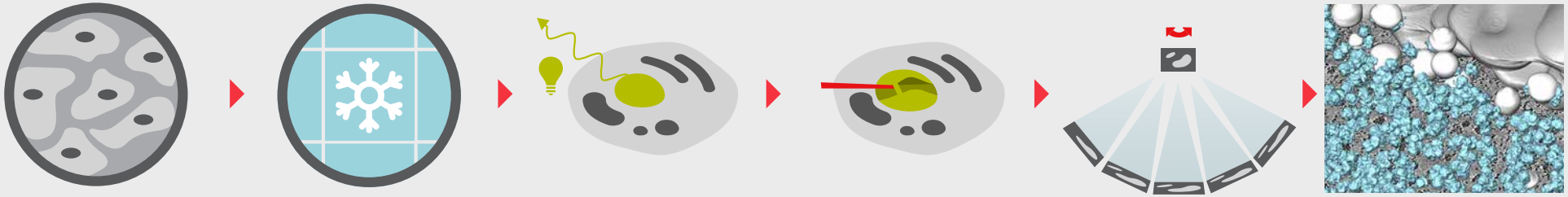
Some of these components are already included with the Tundra, Glacios, and Krios Cryo-TEMs, while others can be added to further enhance your productivity and improve results.



Learn more at **thermofisher.com/SmartEPU**

Cryo-electron tomography

Cryo-electron tomography (cryo-ET) provides label-free, fixation-free, nanometer-scale imaging of a cell's interior in 3D and visualizes protein complexes within their physiological environments. Using a correlative light and electron microscopy approach allows targeting of tagged proteins by fluorescence microscopy before subsequent higher-resolution cryo-EM imaging. Many cells are too thick for electrons, so the vitrified cells must be thinned with a cryo-focused ion beam microscope (cryo-FIB) prior to imaging in a transmission electron microscope.



Cell culture
Cells prepared by routine culture methods are grown on carbon-coated gold electron microscopy grids.

Sample preparation by vitrification
Cells are either vitrified through plunge-freezing (like SPA specimens) or high-pressure freezing (HPF). The water in the sample freezes rapidly and does not crystallize, thus avoiding the molecular-scale disruption that would occur with ice crystal formation during normal slow freezing.

Localization by fluorescence
Using cryo-correlative microscopy, the sample is transferred to a cryo-fluorescence light microscope (cryo-FLM), with which structures of interest are identified. A dedicated cryo-FLM stage keeps the sample in its vitrified state during cryo-fluorescence imaging.

Thinning by milling
A dedicated cryo-FIB prepares a thin, uniform lamella at the vitreous temperature (approximately -170°C).

Imaging by TEM
During cryo-ET, the sample is tilted in known increments about an axis. The individual projection images from the tomographic tilt series are then combined computationally in a procedure known as back-projection, which creates the 3D tomographic volume.

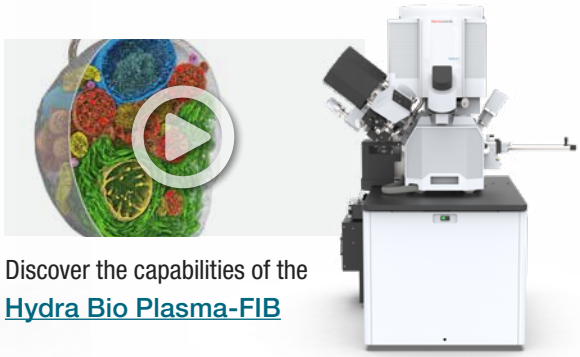
Reconstruction and visualization
The 3D tomogram featuring cellular structures can be segmented and colored in a variety of ways to enhance its display and presentation. From the tomogram, small subsets of data containing the structures of interest can be computationally extracted and subjected to image processing methods.

Learn more at **thermofisher.com/cryo-tomography**

Sample preparation for cryo-tomography

Our end-to-end cryo-electron tomography workflow takes you from flash freezing of cells to final 3D visualization. A cryo-TEM captures the interior cellular regions at nanometer-scale resolution from cryo-lamellae that were precisely prepared with cryo-FIB or cryo-PFIB instruments. Thermo Fisher Scientific offers a range of cryo-FIB tools to match your needs:

Watch to learn more about the
Aquilos 2 Cryo-FIB



Discover the capabilities of the
Hydra Bio Plasma-FIB

Explore the
Arctis Cryo-Plasma-FIB

Aquilos 2 Cryo-FIB

Dedicated cryo-FIB for cellular cryo-ET sample preparation. Cost-effective platform for labs that are new to cryo-EM

The Thermo Scientific™ Aquilos™ 2 Cryo-FIB allows you to prepare high-quality cryo-lamellae for cryo-ET. Key steps can be automated through user-friendly milling recipes.

- View the 3D native subcellular structure using charging contrast and identify features for high-resolution tomography
- Target and extract your structure of interest with our high-precision cryo-lift-out solution
- Extended run times ensure samples remain contamination-free and vitrified throughout the experiment

Learn more

Hydra Bio Plasma-FIB

Versatile plasma-FIB for multi-application labs

The Thermo Scientific™ Hydra Bio™ Plasma-FIB enables high-quality results for large volume microscopy along with TEM lamella preparation at both cryogenic and room temperature.

- Provides a versatile solution for multiple sample types (tissues to proteins)
- Offers a flexible workflow for both volume EM and cryo-ET lamella preparation
- Choose from four ion species (argon, nitrogen, oxygen, and xenon) to optimize your results

Learn more

Arctis Cryo-Plasma-FIB

Automated cryo-plasma-FIB for throughput and connectivity

Automate high-throughput TEM lamellae production with the Thermo Scientific™ Arctis Cryo-PFIB.

- Autoloader connectivity provides a unique, direct connection between cryo-FIB-SEM sample preparation and cryo-TEM
- Ensure correct cryo-lamellae alignment with TomoGrids
- Contains an integrated wide-field fluorescence microscope (iFLM) which allows the same sample area to be observed with light, ion, and electron beams

Learn more

Learn more at **thermofisher.com/cryofib**

Cryo-electron tomography imaging

Our cryo-TEMs are equipped with advanced technology that enables high-resolution imaging of cellular regions at the nanometer scale from cryo-lamellae. With the Autoloader equipped instruments, there is no need for manual grid handling and transfer steps between FIB-SEM and TEM, as the connection is direct. Combined with Thermo Scientific Amira Software, you can visualize, analyze and obtain quantitative information from your images.

Watch to learn more about the Glacios 2 Cryo-TEM

Watch to learn more about the Krios G4 Cryo-TEM

Glacios 2 Cryo-TEM: powerful & versatile

The Glacios 2 Cryo-TEM includes optional Tomography 5 Software, which automates acquisition of tilt-tomograms. This can be paired with optional Thermo Scientific Tomo Live Software, which reconstructs tomograms into 3D volumes, to help you easily assess sample quality and obtain 3D data stacks for cell biology and structural biology workflows. Key features include:

- 200kV accelerating voltage and optional cold field emission gun E-CFEG
- Autoloader with 12 grid capacity
- Falcon 4i Direct Electron Detector
- Selectris X Imaging Filter

Learn more

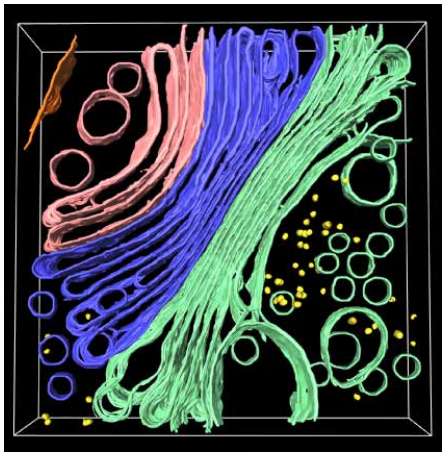
Krios G4 Cryo-TEM: unparallel performance

The Krios G4 Cryo-TEM is designed for the highest resolution, speed and automation for cryo tomography, from sample preparation to data analysis. Key features include:

- 300kV accelerating voltage and optional cold field emission gun E-CFEG
- Autoloader with 12 grid capacity
- Falcon 4i Direct Electron Detector
- Selectris X Imaging Filter
- Biosafety-level containment facilities (e.g., BSL3)

Learn more

A



B

3D visualization of a Golgi apparatus from the green algae Chlamydomonas reinhardtil using the A) Glacios 2 Cryo-TEM and B) Krios G4 Cryo-TEM from a sample prepared with Thermo Scientific Aquilos Cryo-FIB. Data segmentation and visualization by Thermo Scientific Amira Software. Data courtesy of Dr. Benjamin Engel, Department of Molecular Structural Biology, Max Planck Institute for Biochemistry, Martinsried, Germany.

Learn more at **thermofisher.com/cryo-tomography**

Software for cryo-tomography

Our Thermo Scientific™ Tomography 5 and Tomo Live™ Software suite offers: automated 3D data acquisition for multi-site batch tomography, high throughput with multi-shot acquisition, automatic cassette mapping of up to 12 grids for grid-quality assessment and lamella identification, as well as an intuitive graphical setup of tomography areas.

Tomography 5 Software

Robust, automated solution for EM tomography

A user-friendly interface enables both experienced and new users to jump-start their tomography data acquisition. Tomography 5 Software follows the same intuitive logic as Smart EPU Software for single particle analysis. This eases the learning curve and ensures a seamless transition between the two software packages. Tomography 5 Software also connects with Tomo Live Software for automatic on-the-fly data reconstruction and review in any browser.

Tomo Live Software

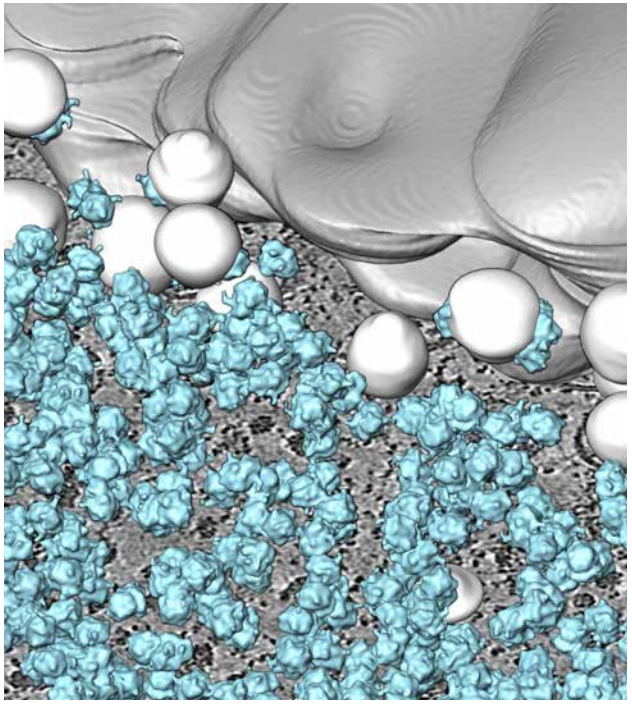
Automatic, on-the-fly reconstruction with data curation

This optional add-on software monitors the quality of data generated with Tomography 5 Software by performing real-time reconstruction of tilt series into 3D volumes. It performs motion correction of fraction movies, fiducial-free tilt series alignment, and 3D reconstruction without user intervention during tilt series recording. These reconstructed tomograms allow for impromptu data quality evaluation, identification of high- and low-quality tilt series, and exporting of selected tilt series and reconstructed tomograms for further processing, archiving, and sharing.

Comet, M, *et al.* **Tomo Live: an on-the-fly reconstruction pipeline to judge data quality for cryo-electron tomography workflows.** *Acta Cryst.* D80, 2024. doi.org: 10.1107/S2059798324001840

The combination of Tomography 5 and Tomo Live Software:

- Provides step-by-step set-up of tomographic data acquisition
- Automates 3D data acquisition for multi-site tomography
- Helps set up regions for analysis and offers a range of imaging presets, such as feature-of-interest tracking



***In-situ* tomogram of a *Chlamydomonas* cell, with 3D visualization of the Golgi complex and ribosomes (light blue). Data courtesy of S. Khavnekar, W. Wietrzynski, and P.S. Erdmann, Max Planck Institute of Biochemistry, Martinsried, Germany.**

Learn more at **thermofisher.com/tomolive**

Volume electron microscopy

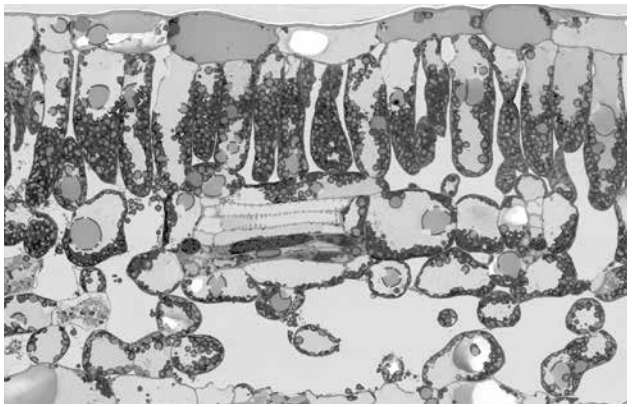
Volume electron microscopy (EM) refers to a variety of imaging approaches that explores below the surface of cellular ultrastructure, tissue, and small model organisms in 3D, at micron to millimeter volume scales, at nanometer-level resolutions, and even at native state under cryogenic conditions.

Serial block-face imaging

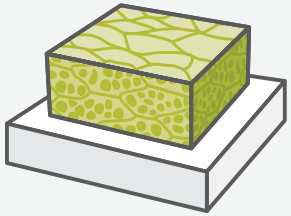
Serial block-face SEM (SBF-SEM) combines *in-situ* sectioning and SEM imaging of plastic-embedded tissue blocks within an SEM vacuum chamber. This fully automated process allows for the reconstruction of large tissue volumes.

Serial FIB/PFIB imaging

This technique uses the FIB to remove sample material layer by layer, revealing a new, deeper surface for imaging at each step. Plasma-FIB (PFIB) SEM tomography utilizes cross-sectioning, where the sample is perpendicular to the ion beam and remains tilted during the imaging process. This approach utilizes the oxygen PFIB of the Hydra Bio Plasma-FIB to access and investigate large areas up to 1 mm in size.

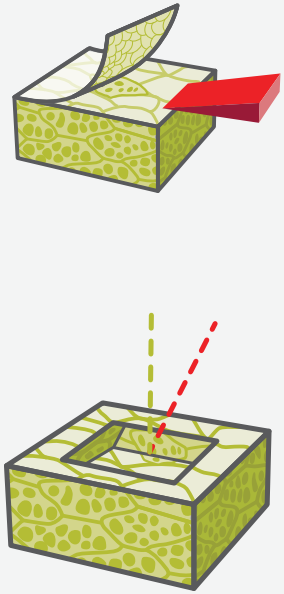


Cross section of a tobacco leaf, stained and embedded in Quetol resin imaged on the Hydra Bio Plasma-FIB. Sample courtesy of Kirk Czymmek, Donald Danforth Plant Science Center, USA



Sample preparation

The resin-embedded sample is prepared using conventional protocols.



Serial block-face imaging

Data acquisition

The electron beam is first used to scan the surface of the resin-embedded tissue sample, capturing a 2D image of the specimen. This top surface is subsequently removed with an *in-situ* microtome. The thickness of each section is user-defined, but is typically less than 15–20 µm.

An image of the fresh surface is then collected with the SEM. This process is repeated until the whole sample has been imaged.

Serial FIB/PFIB imaging

Data acquisition

The resin-embedded sample is imaged with the SEM and the FIB mills away as little as 3–10 nm before it is sequentially imaged again.

This milling and imaging process is repeated until the structure is completely imaged.

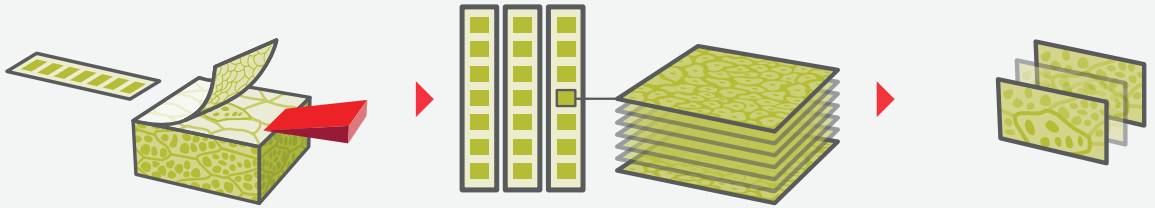
Image processing

The serial stack of images is digitally aligned and processed using 3D rendering software. Cellular compartments can be identified and segmented with analysis software, such as Thermo Scientific™ Amira™ Software.

Learn more at **thermofisher.com/volumeEM**

Array tomography

In array tomography, samples are first cut into a series (array) of sections that are then imaged in sequence with SEM. Advanced software is used to align and recombine these images into a 3D reconstruction.



Sample preparation

Samples are prepared in resin and then cut into consecutive sections with an ultramicrotome.

Data acquisition

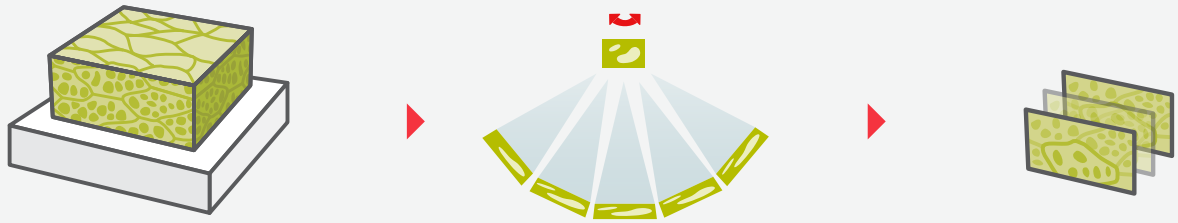
Each serial section is imaged with SEM.

Image processing

The acquired serial images are digitally aligned and processed using 3D rendering software. Cellular compartments can be identified and segmented with analysis software, such as Amira Software.

3D TEM tomography workflow

TEM tomography collects 2D images of electron-thin samples at a range of different angles (i.e., a tilt series). The results are recombined into a 3D reconstruction of the sample.



Sample preparation

The sample is prepared in resin and then cut into consecutive sections with an ultramicrotome.

Data acquisition

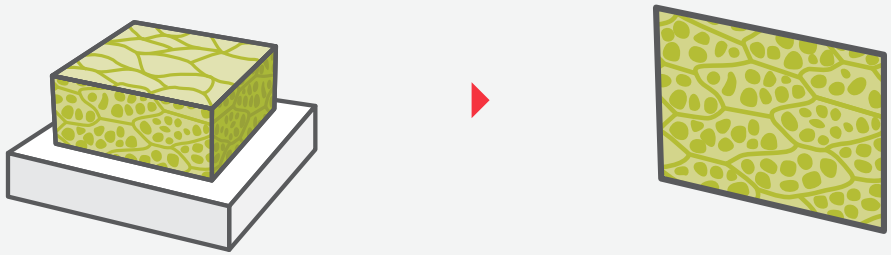
2D images of the electron-thin samples are collected at a range of different angles, producing a tilt series.

Image processing

The volumetric structure of the biological sample is reconstructed from the high-resolution projection images.

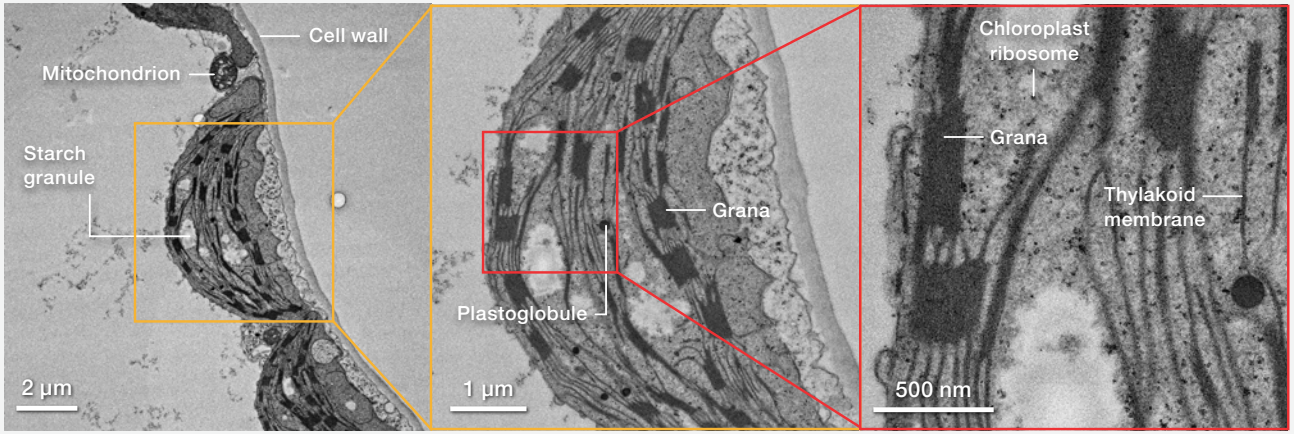
2D transmission electron microscopy

In 2D TEM, electrons are transmitted through a thin, plastic-embedded specimen, forming an image. This enables increased resolution and visualization compared to light microscopy techniques. 2D TEM can be used to study cell morphology and organelles, as well as to identify and characterize viruses, bacteria, fungi, and plant tissue.



Sample preparation
Cells or tissues are perfused and fixed, followed by decalcification. The samples are then stained, dehydrated, and infiltrated with resin.

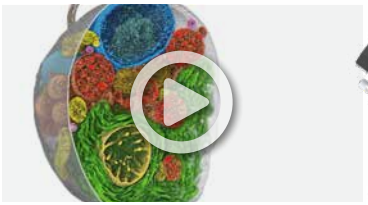
Data acquisition
Regions of interest can be quickly identified and 2D images recorded at high resolution.



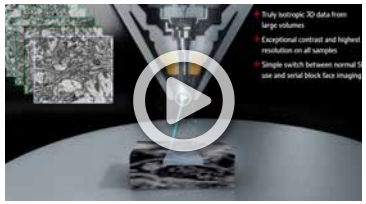
TEM Image of resin-embedded tobacco leaf tissue, imaged with Tundra Cryo-TEM using the Ceta-F CMOS Camera. Sample courtesy of Sarah Powers, Doug Allen, Janithri Wickramanayake, and Kirk Czymmek, Donald Danforth Plant Science Center.

Sample preparation and imaging for volume EM

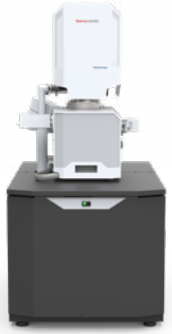
Our volume EM technologies offer integrated solutions for correlative light and electron microscopy, multiple plasma ion-sources, large-area automated serial milling, as well as operation at room temperature or cryogenic conditions.



See product highlights for the
Hydra Bio Plasma-FIB



Take a tour of the
Volumescope 2 SEM



Learn more about the
Talos L120C (S)TEM

Hydra Bio Plasma-FIB

Flexible PFIB imaging at cryogenic or room temperature

The Hydra Bio Plasma-FIB is designed for the volumetric imaging of frozen-hydrated and resin-embedded biological samples. It features proven automation and cryo-technologies for versatile cryo-electron tomography lamella preparation.

- The choice of four different plasma source ions for optimized milling and imaging
- Identify, target, and confirm regions of interest on the same instrument with integrated, correlative light microscopy
- Uncover large sample areas and visualize regions of interest with the Spin Mill Bio Method

Learn more at thermofisher.com/hydrabio

Volumescope 2 SEM

3D SEM for large volume samples

The Thermo Scientific™ Volumescope™ 2 SEM is a dedicated serial block-face imaging system with multi-energy deconvolution, enabling large-volume imaging with isotropic 3D resolution. Keep control of your experiments with easy to use technology that also protects your valuable samples.

- Automated, 3D reconstruction of large tissue volumes with *in situ* sectioning and imaging of plastic-embedded tissue blocks within the SEM vacuum chamber
- Optimize your 3D volume acquisition and reconstruction with live visualization and navigation in Amira Live Tracker Software
- Easily swap between normal SEM operation and automated array tomography with fast microtome mounting/exchange

Learn more at thermofisher.com/volumescope

Talos L120C (S)TEM

Versatile (S)TEM for 2D/3D visualization

Explore a range of biological samples down to sub-nanometer resolution with the Thermo Scientific™ Talos L120C (Scanning) Transmission Electron Microscope.

- Ideal for sections of resin-embedded cells and tissue, isolated particles of protein complexes, and viral assemblies
- Quickly identify regions of interest in pathology samples with high-resolution 2D imaging
- Visualize the organization of intracellular organelles and other structures in 3D
- Characterize nanoparticle quality during process development
- Assess sample quality and optimize sample preparation for high-resolution cryo-EM imaging

Learn more at thermofisher.com/talos-l120c-tem

Software for Volume EM

Explore our reliable software solutions to visualize, segment, and extract quantitative information from your volume EM images.

Maps Software

Array tomography and correlative electron microscopy

Thermo Scientific Maps Software reduces the time and effort needed to record nanometer-resolution data from plastic-embedded cell and tissue sections.

- Easily record as many imaging regions as necessary in any number of sections
- Quickly setup and image with advanced image recognition features
- Precisely place imaging regions for the imaging of smaller fields
- Fully automate your workflow with no manual actions per section

Learn more

Auto Slice & View Software

Automated FIB serial sectioning software

Thermo Scientific™ Auto Slice & View™ 5 Software is designed for automated FIB-SEM serial sectioning.

- Acquire multiple imaging and analysis modalities for every slice, including information such as material and channeling contrast
- Collect elemental information with EDS and grain orientation/strain-texture analysis with EBSD mapping
- Optional Spin Mill Bio Method Software is available on the Hydra Bio PFIB for automated sequential milling and imaging of areas up to 1 mm in size

Learn more

Amira Software

2D–5D visualization, processing, and analysis of EM images

Thermo Scientific™ Amira™ Software is a powerful, comprehensive, and versatile software solution supporting multiple imaging modalities, including optical and electron microscopy, CT, MRI, and more.

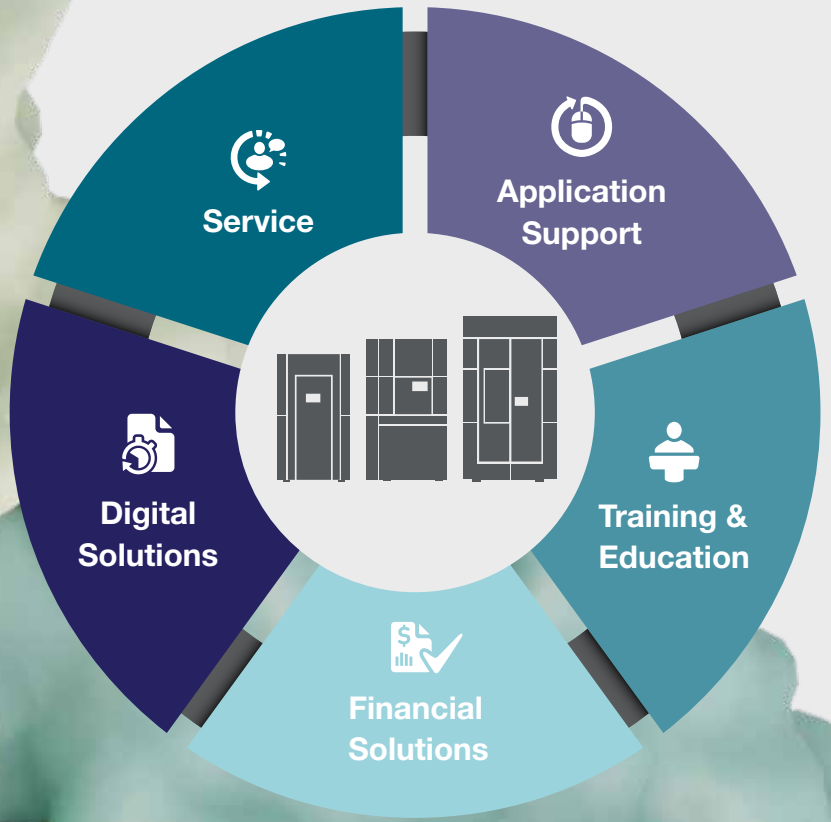
- Easily process 3D image data with AI-powered automation, including time series and multi-channel datasets
- Extract valuable insights from your data, regardless of the modality or scale

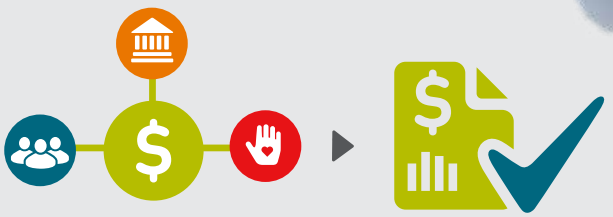
Learn more

Service, support, and financial options

Our **dedicated support network** is committed to your success when you purchase our cutting-edge cryo-electron microscopes with complete end-to-end workflow solutions

- Remote fleet monitoring for maximum uptime
- Local technical experts just a call away
- Financing to meet changing needs, including construction and innovation
- Dedicated program manager to ensure success
- Full remote operation for any site





The **Electron Microscopy Funding Support Center** offers technical content, resources, and proposal writing tools to help you acquire the research instrument you need.

Resources

Getting started with Cryo-EM eBook

Explore how cryo-EM can overcome the current limitations of traditional techniques such as X-ray crystallography (XRD). Learn about key methods, including single particle analysis, microcrystal electron diffraction (MicroED), and cryo-tomography, and how these techniques are used to answer important scientific questions. Discover how cryo-EM has become easier to adopt and more affordable than ever before.

Single particle cryo-EM eBook

Learn more about cryo-EM single particle analysis, the mainstream structural biology technique for the 3D characterization of macromolecules, proteins, and protein complexes at atomic resolution. Read high impact publications and educational resources to learn how this growing technique is providing valuable insights into protein structure and function across; membrane proteins, cell signaling, neuroscience, virology, cancer, and plant sciences.

Cryo-tomography eBook

Access the inner workings of cells through 3D sample reconstruction at unprecedented nanoscale resolution. Results from this technique are having profound effects on our understanding of cell biology, revealing native cellular architecture with molecular clarity. Explore a curated collection of publications highlighting the use of cryo-ET.

Integrative structural biology eBook

Learn more about integrative structural biology, which combines multiple structural determination techniques to enable the full characterization of macromolecular systems. Advances in mass spectrometry, in combination with cryo-EM and other structural tools, are revolutionizing our understanding of protein structure, function, and dynamics. Learn more about tools for integrative structural biology, the benefits of combining cryo-EM, cryo-ET, and mass spectrometry, and the advantages that integrated modeling provides.

Tundra Cryo-TEM brochure

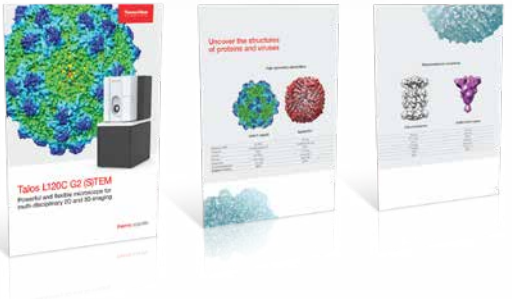
Discover how the Tundra Cryo-TEM allows you to perform structural analyses on challenging proteins and macromolecules with unprecedented ease of use, making cryo-EM more accessible and enabling discoveries in infectious and neurodegenerative diseases, cancer, and more. Validate ligand-protein binding, perform particle characterization, prepare cryo-EM grids, and reveal high-resolution structures.

Glacios 2 Cryo-TEM brochure

Explore how the Glacios 2 Cryo-TEM can provide deeper insights into protein structures, transform drug discovery and development, uncover the interactions of proteins/complexes, and overcome the challenges of small molecule visualization. Discover cryo-EM that is accessible and versatile with improved ease-of-use and productivity for both new and experienced users.

Krios G4 Cryo-TEM brochure

Discover how the award-winning 300 kV Krios G4 Cryo-TEM offers unmatched biological insights through the 3D visualization of proteins and molecular machines, their localization, and dynamics within the architecture of the biological cell.



Talos L120C (S)TEM brochure

Learn how the versatile Talos L120C (S)TEM can be used to visualize sections of resin-embedded cells and tissues, isolate particles of protein complexes and viral assemblies, as well as other materials down to sub-nanometer resolutions. With a flexible base configuration, the system can be adapted to multiple applications and use different imaging and detection techniques, including cryo-EM single particle analysis.

Arctis Cryo-Plasma-FIB brochure

The Thermo Scientific Arctis Cryo-Plasma Focused Ion Beam (Cryo-PFIB) is specifically designed for automated, high-throughput production of cryo-lamellae from vitrified cells. The microscope features a state-of-the-art plasma ion source, an automatic sample loading system (Autoloader), integrated fluorescence microscopy for correlative imaging, and direct connectivity to the cryo-transmission electron microscope.

Volume Electron Microscopy brochure

Our volume EM technology offers integrated solutions for correlative light and electron microscopy, multiple plasma ion-sources, large area automated serial milling, as well as operation at room temperature or cryogenic conditions.

Learn more at **thermofisher.com/plantbiology-EM**