Trends in Biochemical Sciences | Technology of the Month

Correlative light-electron microscopy: integrating dynamics to structure

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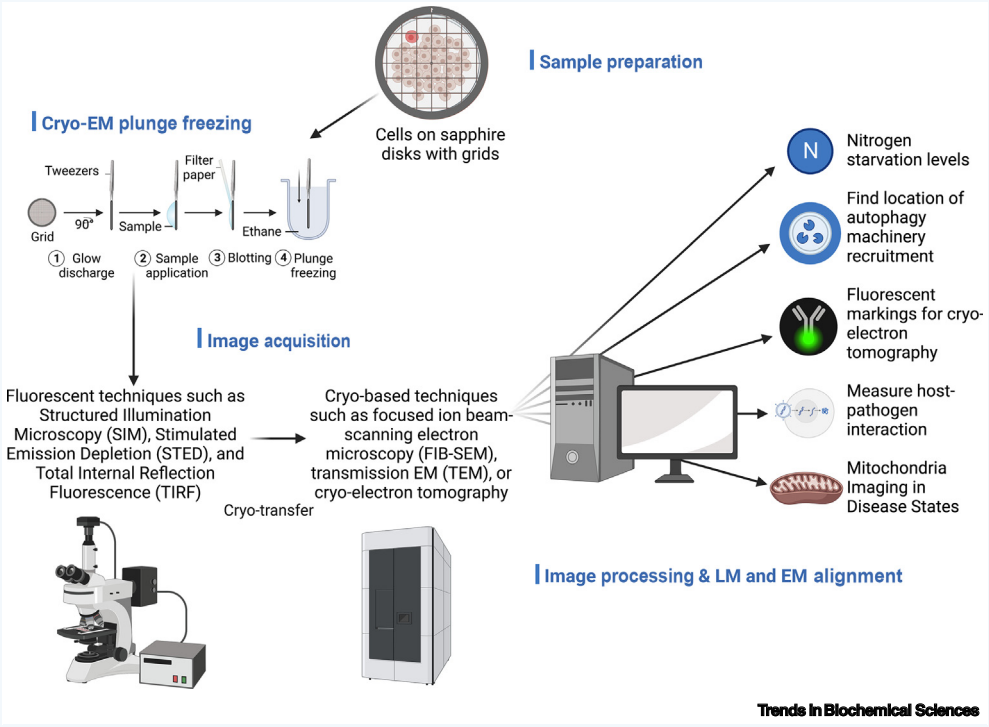
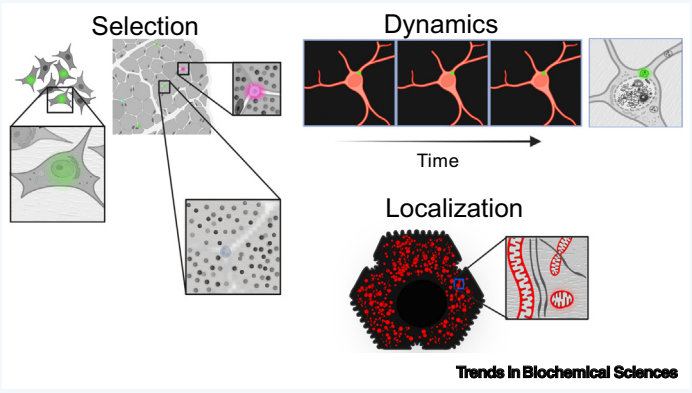


A challenge of classical electron microscopy (EM) modalities is the static and limited view they present of dynamic biological processes. Correlative light and EM (CLEM) pairs the precise imaging of the cellular location in light microscopy (LM) and EM, taking advantage of both methods. Recent advances in protein labeling, fluorescent markers, super-resolution microscopy, and EM technologies have allowed widespread application in modern biological research. Because of the wide versatility of CLEM, it is difficult to describe a single workflow. In general, samples are prepared and imaged by LM before EM preparation and imaging, with software utilized for the correlation of the two images.



Fluorescent tags can help to select and localize structures for EM or add dynamics through live-cell imaging. Future developments in multimodal probes showing electron density and fluorescence activity will allow greater integration of the modalities and high-throughput workflows.

ADVANTAGES:
Can be paired with focused ion beam-scanning EM (FIB-SEM), cryo-EM, transmission EM (TEM), and serial block-face SEM (SBF-SEM), as well as structured illumination microscopy (SIM), stimulated emission depletion (STED), and total internal reflection fluorescence (TIRF).

Amenable to live-cell imaging, in part due to advances that can quickly freeze a sample for cryo-EM.

Systems to pair with microscopic X-ray computed tomography to provide non-destructive forms of 3D imaging alongside high-resolution micrographs obtained by TEM and fluorescence.

Sensitive enough for applications such as virus–host cell membrane interactions.

Alongside other techniques, can localize the distribution of specific proteins or structures within a sample and correlate this information with the ultrastructural details provided by EM.

CHALLENGES:
Finding the appropriate FM and EM microscopes may be difficult and expensive.

Preparing samples for both modalities requires careful planning and optimization so they are properly preserved, labeled, and processed for the chosen application.

The growth of cells on the grid during the LM phase can make correlational positioning difficult.

Aligning LM and EM micrographs can be challenging and time-consuming due to differences in resolution and sample shrinkage.

Lower throughput and smaller region-of-interest size than traditional FM techniques due to being reliant on EM acquisition and additional sample preparation times.

https://doi.org/10.1016/j.tibs.2023.05.003

Acknowledgments

We thank Kit Neikirk for his thoughtful suggestions and edits to the manuscript. Funding by the UNCF/Bristol-Myers Squibb E.E. Just Faculty Fund, BWF Career Awards at the Scientific Interface Award, BWF Ad-hoc Award, National Institutes of Health (NIH) Small Research Pilot Subaward to 5R25HL106365-12 from the NIH PRIDE Program, DK020593, Vanderbilt Diabetes and Research Training Center for DRTC Alzheimer's Disease Pilot & Feasibility Program. CZI Science Diversity Leadership grant number 2022- 253529 from the Chan Zuckerberg Initiative DAF, an advised fund of Silicon Valley Community Foundation (to A.H.). The images were arranged using BioRender and all representative images are original.

Declaration of interests

The authors have no interests to declare.

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https://doi.org/10.1016/j.tibs.2023.05.003