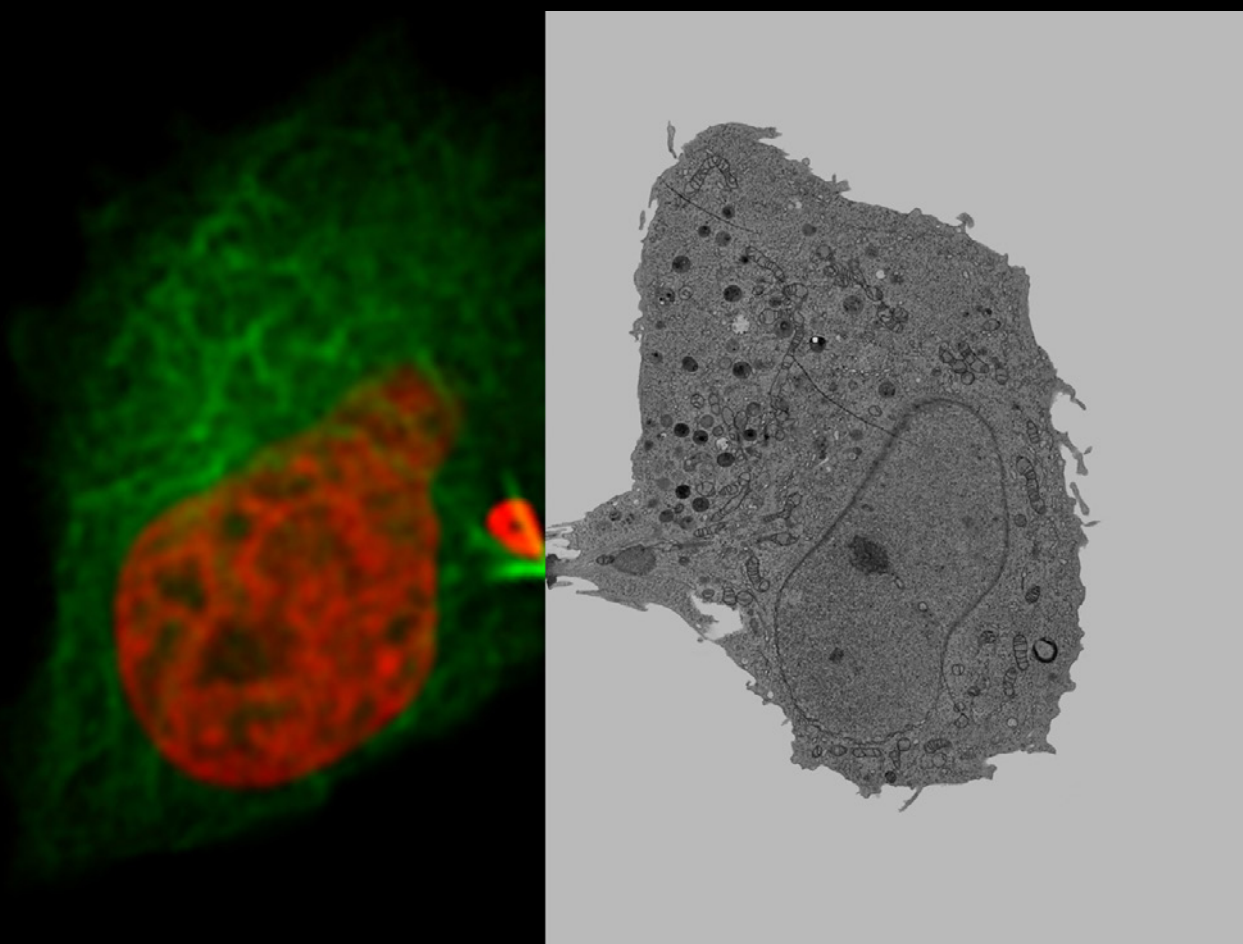


From Eye to Insight

Leica
MICROSYSTEMS

LIVE CELL CLEM WORKFLOW

Coral Life





HeLa cells expressing H2B-mCherry and alphaTubulin-mEGFP to visualize microtubules and DNA were cryofixed by high-pressure freezing 60 min after cytokinesis onset. The intercellular bridge was imaged in TEM after freeze substitution and resin infiltration.

200 μm

PUTTING DYNAMIC LIVE CELL DATA INTO THE ULTRASTRUCTURAL CONTEXT

Take advantage of correlative light and electron microscopy

Identify directly the right cell at the right time and put dynamic live cell data into their ultrastructural context with our new live cell CLEM solution Coral Life.

Coral Life is an integrated solution, combining live-cell correlative light and electron microscopy (CLEM) with time-specific fixation by high-pressure freezing, which enables the investigation of dynamic events with nanometer resolution. A massive surplus of context data can be obtained by the combination of dynamic data, specifically targeted entities by fluorescent live-cell imaging, and precisely timed EM ultrastructural analysis.

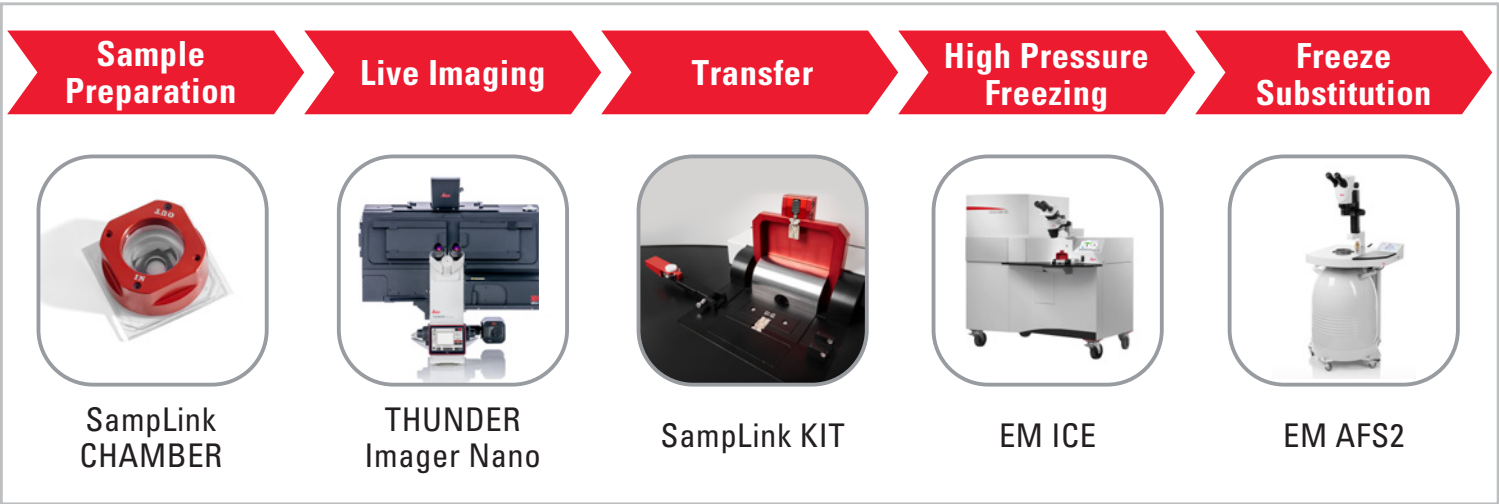
Now there is no need to guess about the underlying mechanisms when analyzing a wide range of biological events, like intracellular trafficking, nuclear envelope formation, or mitotic spindle assembly. The Coral Life workflow gives you an integrated solution, from targeting to insights, that will boost your research.

The right balance of patience, timing, and speed

Many electron microscope workflows start with the fixation of cells, followed by sample preparation and EM imaging. However, cells displaying interesting behavior tend to be rare. Making things even more complicated is the fact that the cellular behavior or processes of interest might be very transient. Finding the “right cell” is often very time-consuming and tedious work which seems like “looking for a needle in a haystack”.

The Live-cell CLEM workflow allows you to capture dynamic information of a relevant biological process when it happens and enables you to put these observations into their ultrastructural context. With Coral Life, you can rest assured that you obtain the results you are looking for. Find the right cell in the right state at the right time with the light microscope, observe it, and retrieve it rapidly and reliably once the sample is loaded in the electron microscope. Enjoy peace of mind while looking for the cell of interest with the light microscope and quickly “snap-freeze” it precisely at the correct moment.

#Capturelife the moment it happens



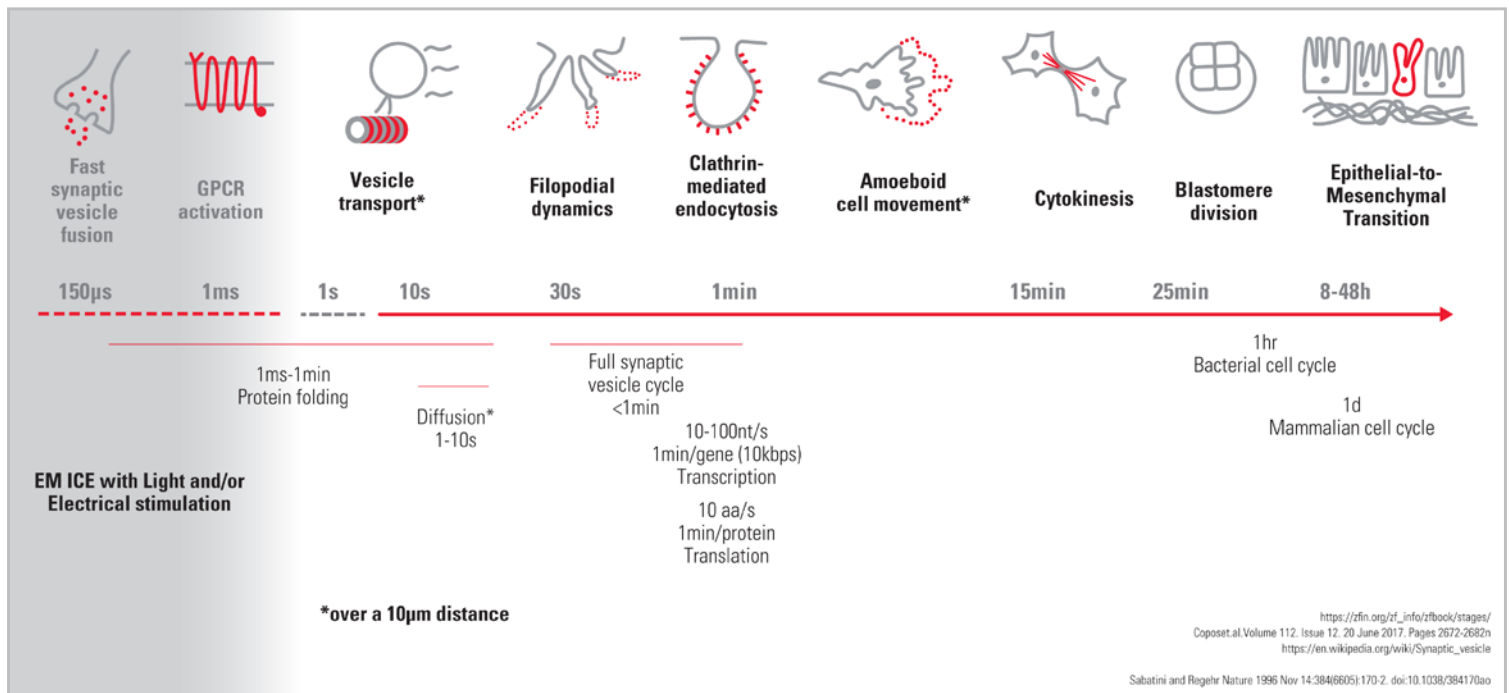
Coral Life

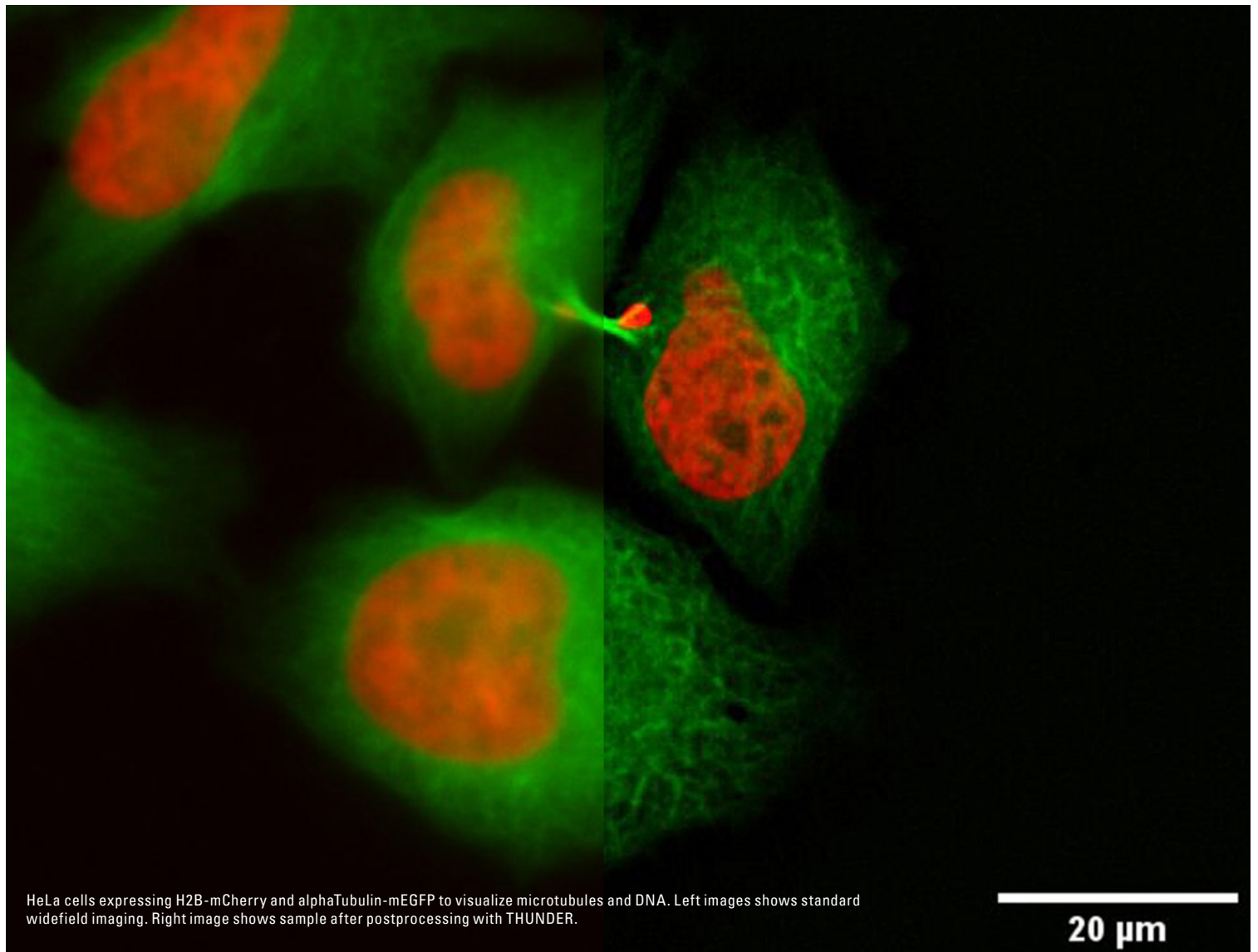
A streamlined workflow for reproducible results

Increase your success rate with the first streamlined live-cell-to-vitrification workflow without compromising the health and integrity of the specimen. The different workflow components work seamlessly together, enabling the culturing, transport, imaging, and cryo-fixation of cells on sapphire without any physiological interruption.

- > first streamlined live-cell-to-vitrification workflow
- > specimens stay healthy and intact
- > different components work seamlessly together
- > no physiological interruption during culturing, transport, imaging, and cryo-fixation of cells on sapphire

*[in graphic] For processes in the sub-second range, the EM ICE high pressure freezer, which is part of the Coral Life workflow, offers you electrical or optical stimulation with millisecond precision.





An imaging window for dynamic processes

A transfer time of just 5 seconds opens a window to a wide range of dynamic processes which occur in living cells. With some experience, you can even refine the exact point in time of the processes which you want to capture in the prepared specimens. Coral Life gives you complementary dynamic and ultrastructural information whether you are interested in

- > vesicle transport and sorting,
- > filopodial dynamics, endocytosis,
- > cell migration,
- > cytokinesis,
- > or a plethora of other biological processes.

The generous time window before the transfer ensures that you to have enough room for sample identification and imaging. Without the pressure to make a fast decision, typical of other live-cell CLEM systems where often there are only a few minutes to choose a region, you can make the selection more calmly, the success rate increases, and the results are of better quality and more reproducible.

INDIVIDUAL WORKFLOW STEPS - SEAMLESSLY CONNECTED



SampLink chambers and OxyGenie's incubator system

Keep your samples under optimal conditions

The SampLink chambers keep live cells healthy, giving you enough time to select and image a region of interest until you find cells in the exact state you are looking for.

- > Permits long term imaging under physiological conditions thanks to the open sample container with continuous gas exchange
- > Gas and temperature control during cell cultivation and transport with OxyGenie's incubator system



THUNDER Imager Nano

Optimized image resolution

The THUNDER Imager Nano enables high resolution and simplifies live-cell imaging, as one does not need to use dip-in objectives which increase the risk of contaminating the cells. SampLink chambers can be loaded directly into the THUNDER Imager Nano.

- > Fast 3D live-cell imaging for a more accurate study of physiology and blur removal for better target identification with THUNDER
- > Stable physiological conditions of the sample due to the connection to the OxyGenie system and temperature control via the microscope
- > Optimized image resolution with the equipped 40x / 1.1 NA sapphire-corrected water-immersion objective



Easy adaptation to the EM ICE

Time is key

To minimize the time between live-cell imaging and freezing, Coral Life comes with an intuitive transfer system that allows users to start the freezing directly after imaging, while keeping the sample under physiological control.

- > Capture transient events with a transfer that takes less than 5 seconds
- > Benefit from an all-in-one package with complementary consumables
- > Adapt easily and quickly to the EM ICE with the slide-2-release mechanism for the middle plate



Cryo-immobilisation with the EM ICE High Pressure Freezer

Optimal sample fixation

With the EM ICE, the imaged samples are ultimately cryo-immobilized. It is the optimal system for combining classic high-pressure freezing with new workflow solutions.

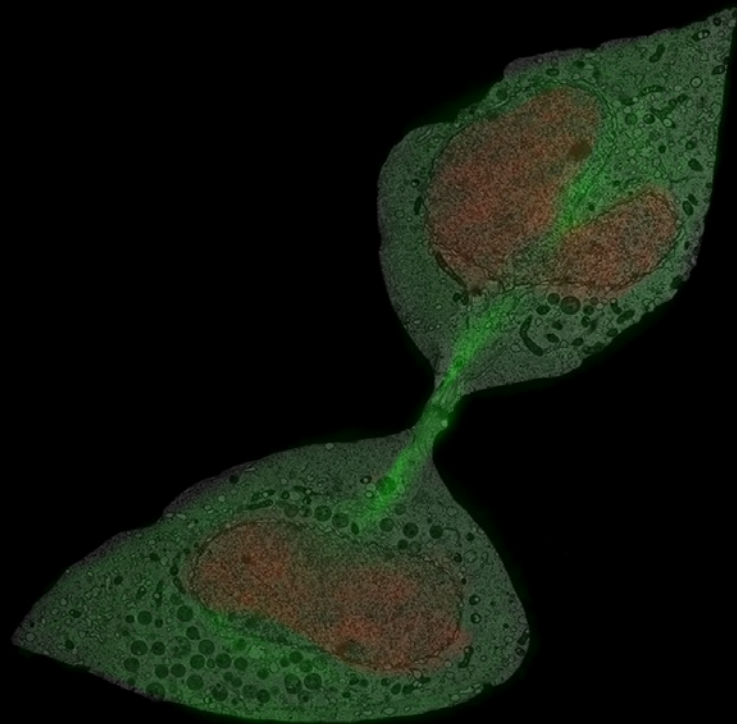
- > Retrofittable to Coral Life
- > Full vitrification of adherent cells
- > Live-cell CLEM can be combined with optical stimulation experiments thanks to the optional upgrade for light and electrical stimulation



Freeze Substitution System EM AFS2

Downstream workflows

Benefit from our established freeze substitution downstream workflow using the EM AFS2, EM TRIM2, EM UC7, or EM AC20 to prepare your samples for subsequent TEM imaging. Coral Life is also suitable for SEM tomography analysis. Use the ARTOS 3D to prepare samples on wafers.



Correlation of the light- and electron microscopic image with 40% transparency. HeLa cells were labelled with H2B-mcherry and alphaTubulin-EGFP.

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