



Leica EM HPM100

High Pressure Freezer

Superior cryofixation of biological and industrial samples

Living up to Life

Leica
MICROSYSTEMS

High Pressure Freezing

High pressure freezing is by far the most significant sample preparation method for morphological and immunocytochemical high resolution studies for electron microscopy.

High pressure freezing has made it possible to observe aqueous biological and industrial samples near to native state.

The 2100 bar of high pressure applied to the sample during high pressure freezing using the Leica EM HPM100 suppresses ice crystal formation and growth, while cryo immobilization immediately after pressurization prevents structural damage to the sample.

High pressure frozen samples can be completely vitrified up to a thickness of 200 μm , a 10 to 40-fold increase in the depth of amorphous ice. No conventional freezing method can generate such large, well frozen samples.

The unique 6 mm diameter carrier system of the Leica EM HPM100 allows even more sample area to be frozen, like no other high pressure freezing instrument.

The state-of-the-art design of the Leica EM HPM100 enables express sample handling and easy use with perfect freezing results.



Inspired to Make the Difference

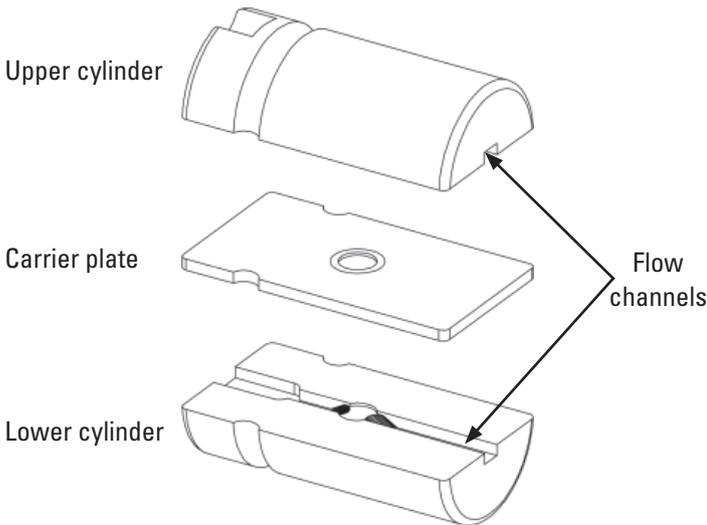


The Unique Leica EM HPM100 Features

- Automatic freezing of samples with one button operation
- Extremely high cooling rates
- Perfect freezing of samples up to 6 mm diameter
- No cryoprotectants required
- Rapid transfer loading device for CLEM work
- Fast, ergonomic sample loading
- Express sample handling for fast freezing
- Graphical data display of actual temperature, time and pressure for each run
- Integrated work station
- Integrated stereomicroscope and adjustable LED illumination
- Universal and application specific sample carriers
- Integrated Dewar with drain outlet
- Automatic sample transfer to LN₂ – no manipulation required
- Automatic bake-out cycle
- Thermally insulated process chamber
- Integrated touch control screen with prompt window
- Freezing process cover for ultimate safety
- USB interface for data storage and transfer
- Integrated air compressor
- Compact and mobile
- Uses standard 100–250 V power supply

Simplicity with Technology

The Leica EM HPM100 is an automatic and easy to handle high pressure freezing instrument. With a one button operation, a cartridge holding the sample is inserted into the high pressure chamber.



Sample cartridge

The sample cartridge is made of high performance insulating polymer and consists of three components: two cylinders with a flow channel, and a carrier plate with an opening to hold the sample carrier assembly.

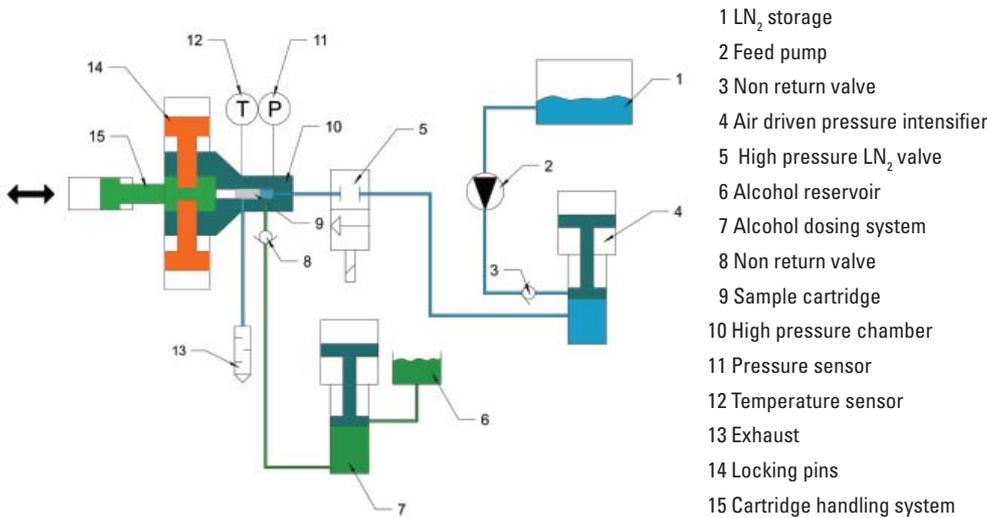
The sample cartridge functions as process chamber and sample holder at the same time.

The integrated insulating flow channels direct the freezing onto the sample.

This results in well frozen samples and low liquid nitrogen consumption.

An air-driven intensifier pressurizes the liquid nitrogen to 2100 bar. The process chamber is filled with a small volume of alcohol prior to freezing. The high pressure liquid nitrogen valve opens after the freezing pressure is reached. The pressurized liquid nitrogen is injected into the process chamber. The flow channels direct the rapid flow of liquid nitrogen along the sample carrier surfaces. This induces extremely high cooling rates.

High cooling rates with the high-pressurized liquid nitrogen system



- 1 LN₂ storage
- 2 Feed pump
- 3 Non return valve
- 4 Air driven pressure intensifier
- 5 High pressure LN₂ valve
- 6 Alcohol reservoir
- 7 Alcohol dosing system
- 8 Non return valve
- 9 Sample cartridge
- 10 High pressure chamber
- 11 Pressure sensor
- 12 Temperature sensor
- 13 Exhaust
- 14 Locking pins
- 15 Cartridge handling system

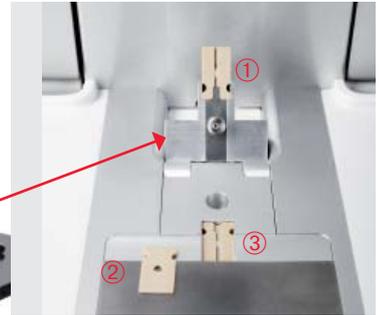
The vitrified sample is automatically transferred to the LN₂-filled Dewar. The freezing process is digitally displayed on a graph showing the actual temperature, time and pressure.

Easy Freezing with the Leica EM HPM100

Sample loading

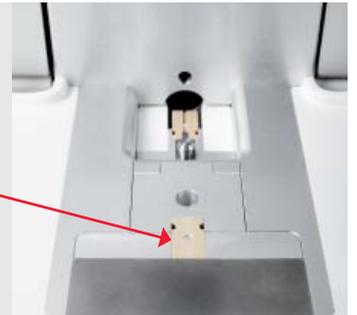
The upper and lower cylinder of the three-piece cartridge are placed in the sample loading assembly.

- ① Upper cylinder
- ② Carrier plate
- ③ Lower cylinder

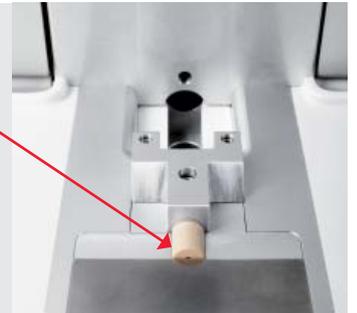


The carrier plate is placed on top of the lower cylinder.

A sample carrier is placed in the center of the carrier plate. The user fills the carrier with the sample to be frozen and places a second carrier on top.



With the cartridge lever the upper cylinder is sandwiched onto the lower cylinder and sample.



One button operation

With a one button operation the enclosed sample is injected for freezing. The sample is automatically frozen in less than 2.5 seconds after injection.



After cryofixation, the frozen sample is automatically released into the LN₂ Dewar.

The vitrified frozen sample is transferred to the carrier release device. The sample is expelled from the carrier plate and is ready for follow-on procedures.



Complete. Convenient. Preparation.

Not only is the Leica EM HPM100 compact and mobile, it also offers a variety of unique features which enables the user to quickly, ergonomically and conveniently prepare samples for freezing.

Process parameters can be stored on a standard memory stick for each run using the USB interface.

The touch screen displays process parameters, temperature, pressure curves, bake-out cycle, and is also used for service.

All freezing processes are enclosed by a safety cover for user protection.



The vitrified sample is automatically released into a conveniently located LN₂-filled Dewar. The sample stays well frozen at all times.



Well-positioned cartridge lever for quick manoeuvring of the sample cartridge.



The integrated stereomicroscope can slide left and right allowing better sample viewing and access during preparation and processing. It comes complete with adjustable LED illumination.



The two-coloured workstation table is cleverly designed for improved sample visualisation, such as when working with cellulose microcapillaries.

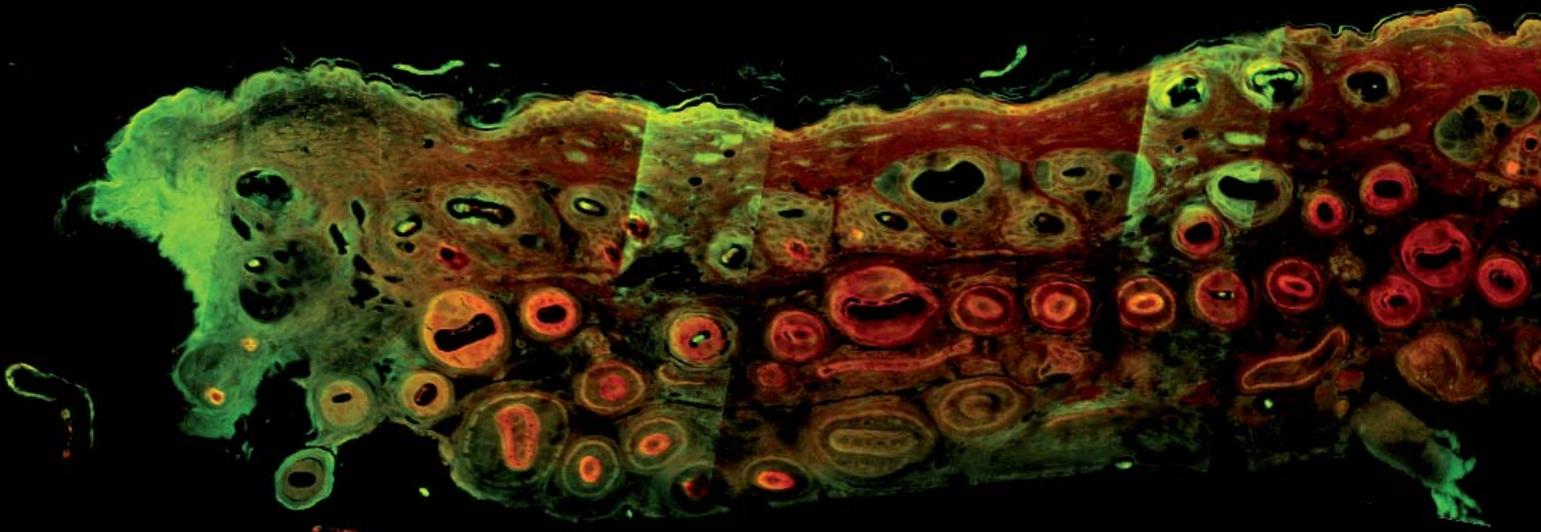
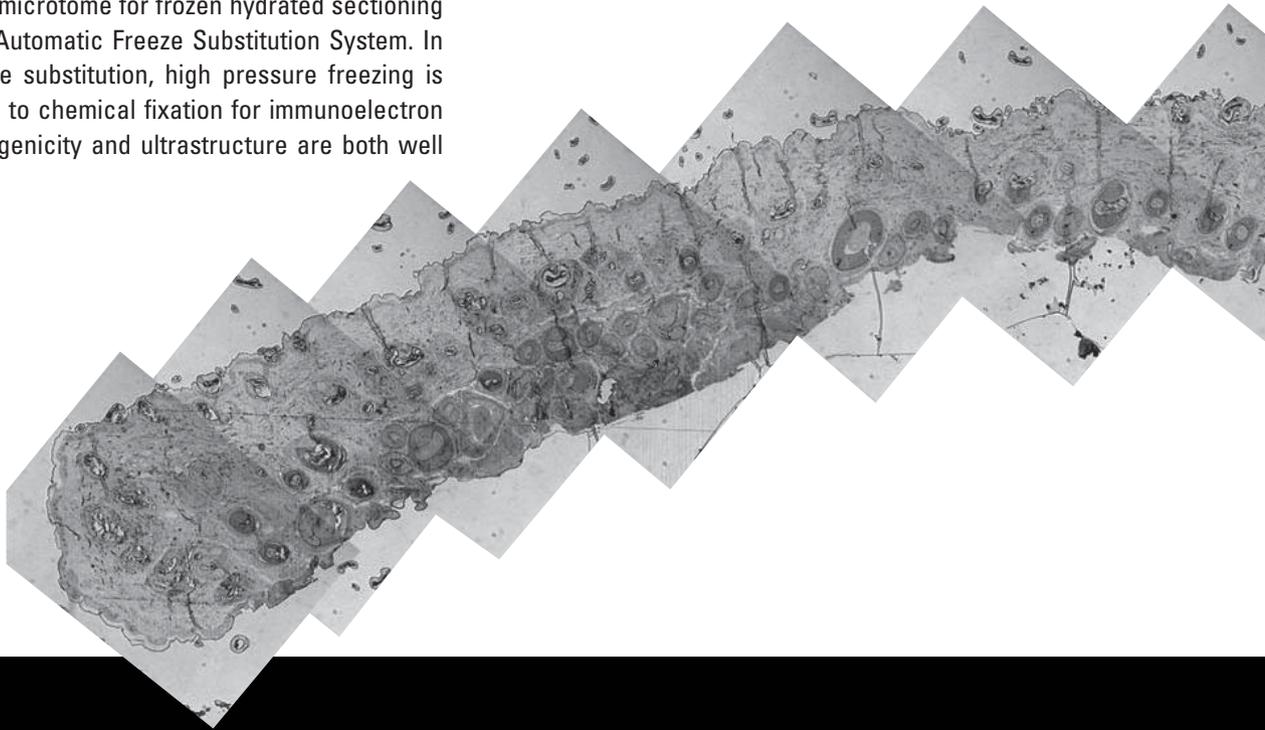


The user can operate the Leica EM HPM100 in an ergonomically seated position.

Revolutionary in Application

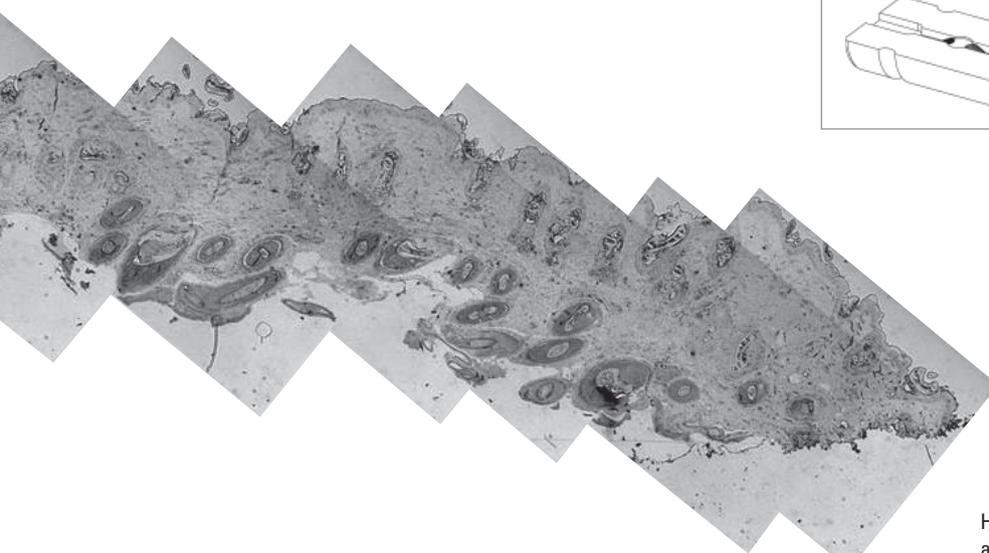
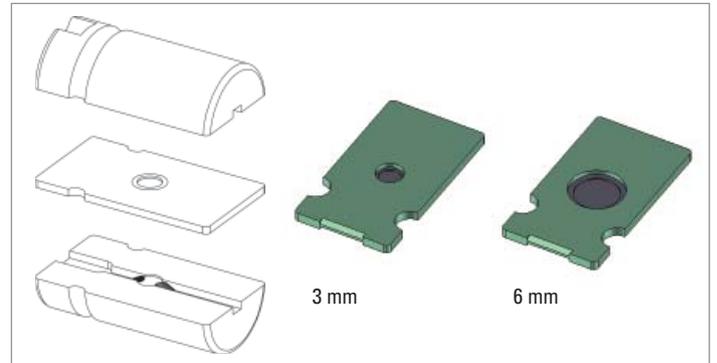
The Leica EM HPM100 offers a variety of sample carriers suitable for almost any type of application. Most notable is the 6 mm diameter sample carrier, which no other high pressure freezing instrument offers.

Once frozen, samples can be placed into the cryo chamber of the Leica EM UC6/FC6 ultramicrotome for frozen hydrated sectioning or the Leica EM AFS2 Automatic Freeze Substitution System. In combination with freeze substitution, high pressure freezing is an excellent alternative to chemical fixation for immunoelectron microscopy as the antigenicity and ultrastructure are both well preserved.



The New 6 mm Diameter Sample Carrier. Only by Leica Microsystems.

The 6 mm carrier opens up new perspectives for correlative microscopy, as it allows a true pre-selection of a region of interest by CLSM within large areas and the EM investigation of the same sample without the drawback of artefacts caused by chemical fixation.

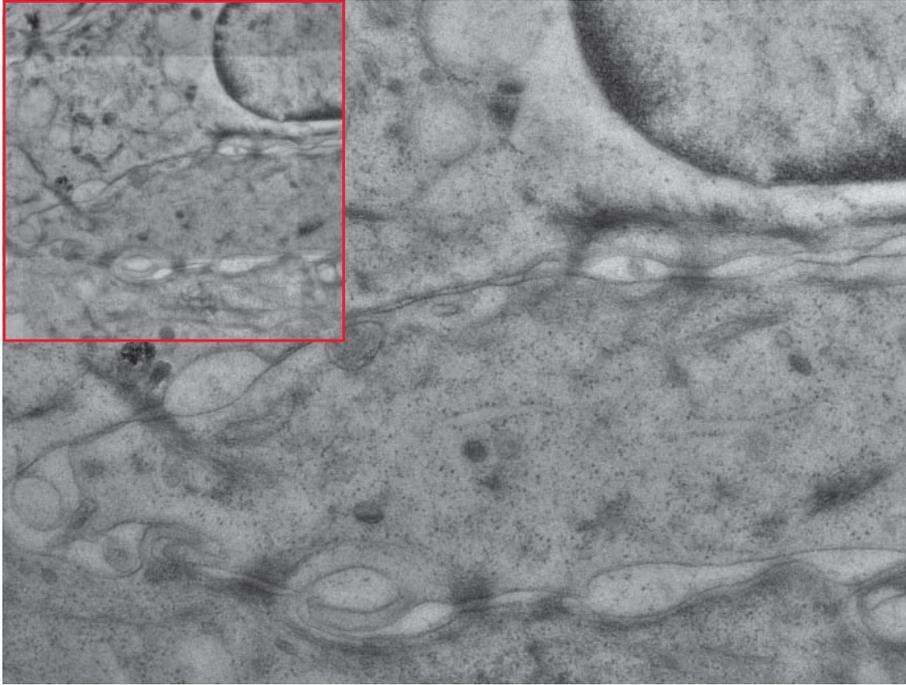


High pressure frozen mouse skin biopsy (diameter 6 mm) after freeze substitution in uranyl acetate/acetone.

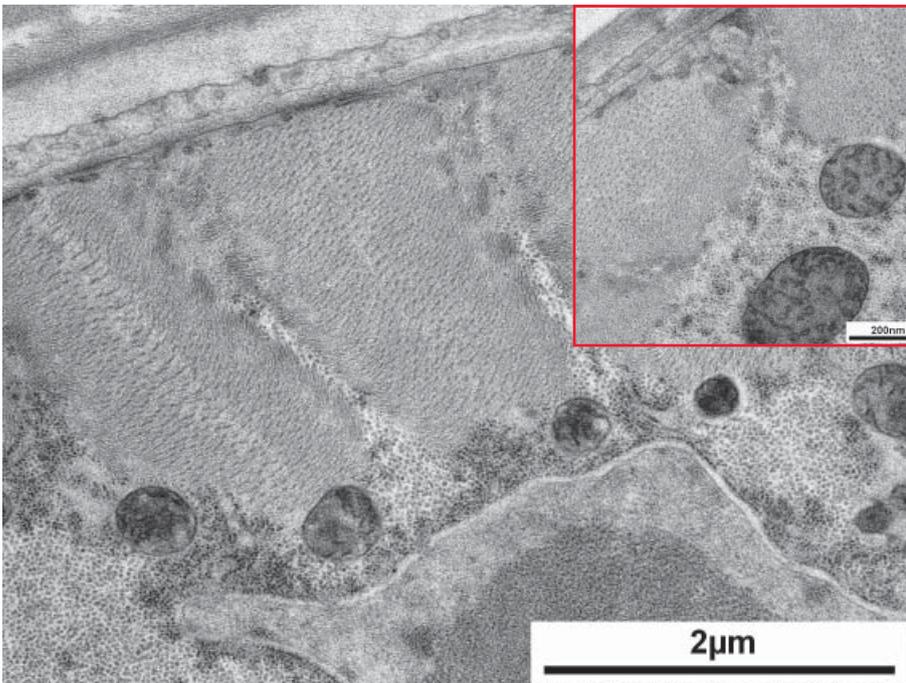


Pressure frozen mouse skin biopsy (diameter 6 mm) after freeze substitution in uranyl acetate/acetone solution stained with safranin O in a confocal laser scanning microscope. Images courtesy of Electron Microscopy ETH Zurich (EMEZ).

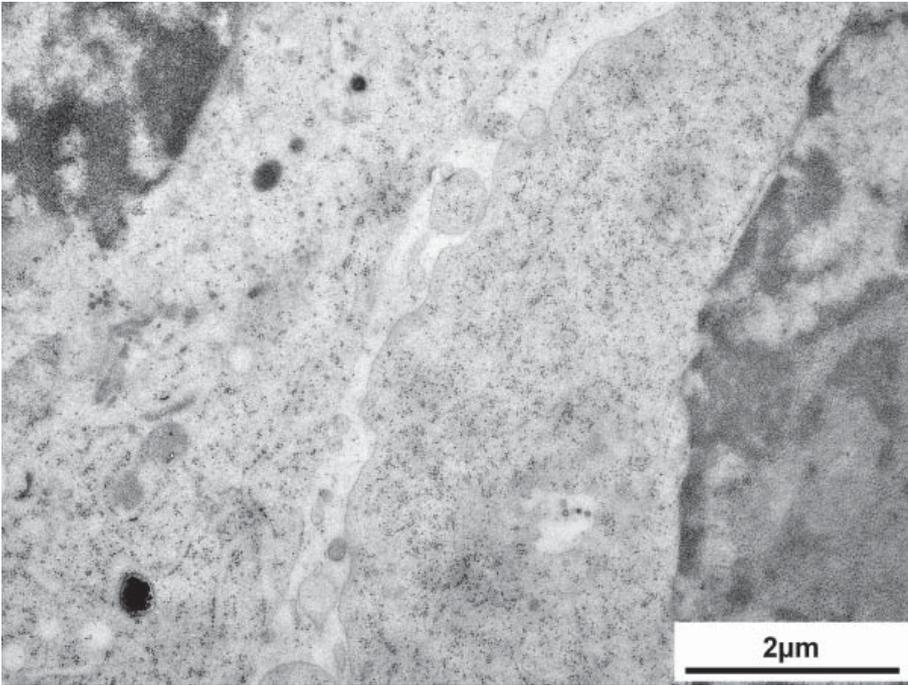
Excellent Preservation of Ultra Structure



High pressure frozen mouse skin biopsy (diameter 6 mm) after freeze substitution in uranyl acetate/acetone solution post-stained with uranyl acetate and lead citrate and sectioned prior to imaging in a TEM. Electron Microscopy ETH Zurich (EMEZ).

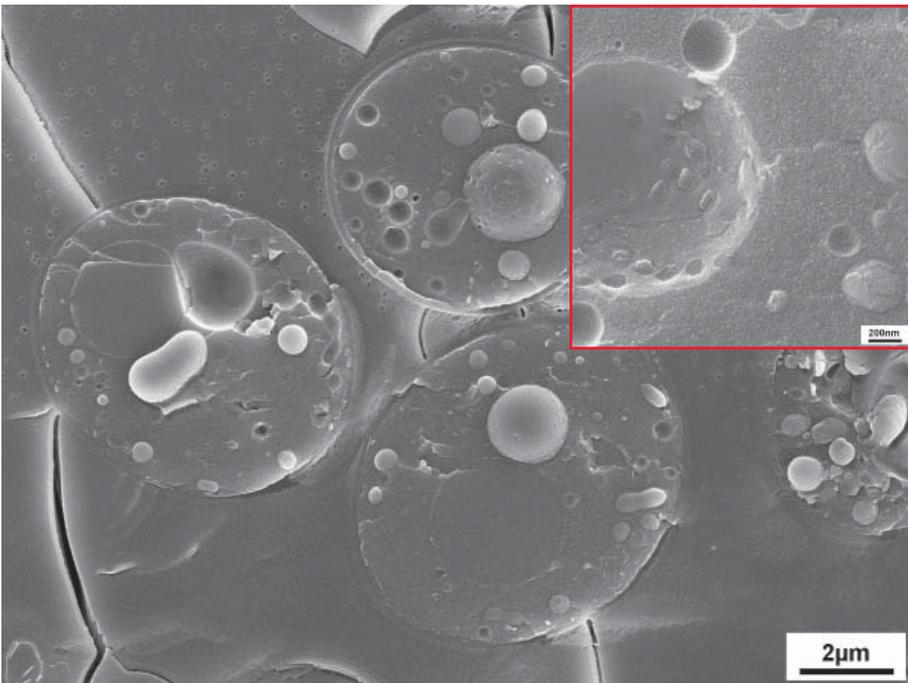


High pressure frozen Nematode. Freeze substituted in water-free acetone containing 2% OsO₄, embedded in Epon/Araldite, thin sectioned and stained with uranyl acetate 2% and Reynolds lead citrate. Hexadecene was used as transmission fluid. Electron Microscopy ETH Zurich (EMEZ). Specimen courtesy of M. Gotta, Institute of Biochemistry, ETH Zurich, Switzerland.



High pressure frozen Macrophage monolayer grown on Sapphire disc. Freeze substituted in water-free acetone containing 2% OsO₄, embedded in Epon/Araldite, thin-sectioned and stained with uranyl acetate 2% and Reynolds lead citrate. Electron Microscopy ETH Zurich (EMEZ). Specimen courtesy of G. Sumara, Institute of Cell Biology, ETH Zurich, Switzerland.

Alternatively, when using the appropriate sample carriers, samples can be transferred to the Leica EM BAF060 Freeze Etch System and processed for high resolution cryo-SEM or to make high resolution replicas for TEM, amongst other applications.



Cryo SEM image of a high pressure frozen suspension of baker's yeast *Saccharomyces cerevisiae*. Freeze fractured in the Leica EM BAF060 at -115°C, etched at -105°C for 5 minutes, coated by electron beam evaporation with 3 nm of Pt/C. Electron Microscopy ETH Zurich (EMEZ).

“With the user, for the user”

Leica Microsystems

Leica Microsystems operates internationally in four divisions, where we rank with the market leaders.

• Life Science Division

The Leica Microsystems Life Science Division supports the imaging needs of the scientific community with advanced innovation and technical expertise for the visualization, measurement, and analysis of microstructures. Our strong focus on understanding scientific applications puts Leica Microsystems' customers at the leading edge of science.

• Industry Division

The Leica Microsystems Industry Division's focus is to support customers' pursuit of the highest quality end result. Leica Microsystems provide the best and most innovative imaging systems to see, measure, and analyze the microstructures in routine and research industrial applications, materials science, quality control, forensic science investigation, and educational applications.

• Biosystems Division

The Leica Microsystems Biosystems Division brings histopathology labs and researchers the highest-quality, most comprehensive product range. From patient to pathologist, the range includes the ideal product for each histology step and high-productivity workflow solutions for the entire lab. With complete histology systems featuring innovative automation and Novocastra™ reagents, Leica Microsystems creates better patient care through rapid turnaround, diagnostic confidence, and close customer collaboration.

• Surgical Division

The Leica Microsystems Surgical Division's focus is to partner with and support surgeons and their care of patients with the highest-quality, most innovative surgical microscope technology today and into the future.

The statement by Ernst Leitz in 1907, “with the user, for the user,” describes the fruitful collaboration with end users and driving force of innovation at Leica Microsystems. We have developed five brand values to live up to this tradition: Pioneering, High-end Quality, Team Spirit, Dedication to Science, and Continuous Improvement. For us, living up to these values means: **Living up to Life.**

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