



Leica EM PACT2

High Pressure Freezer

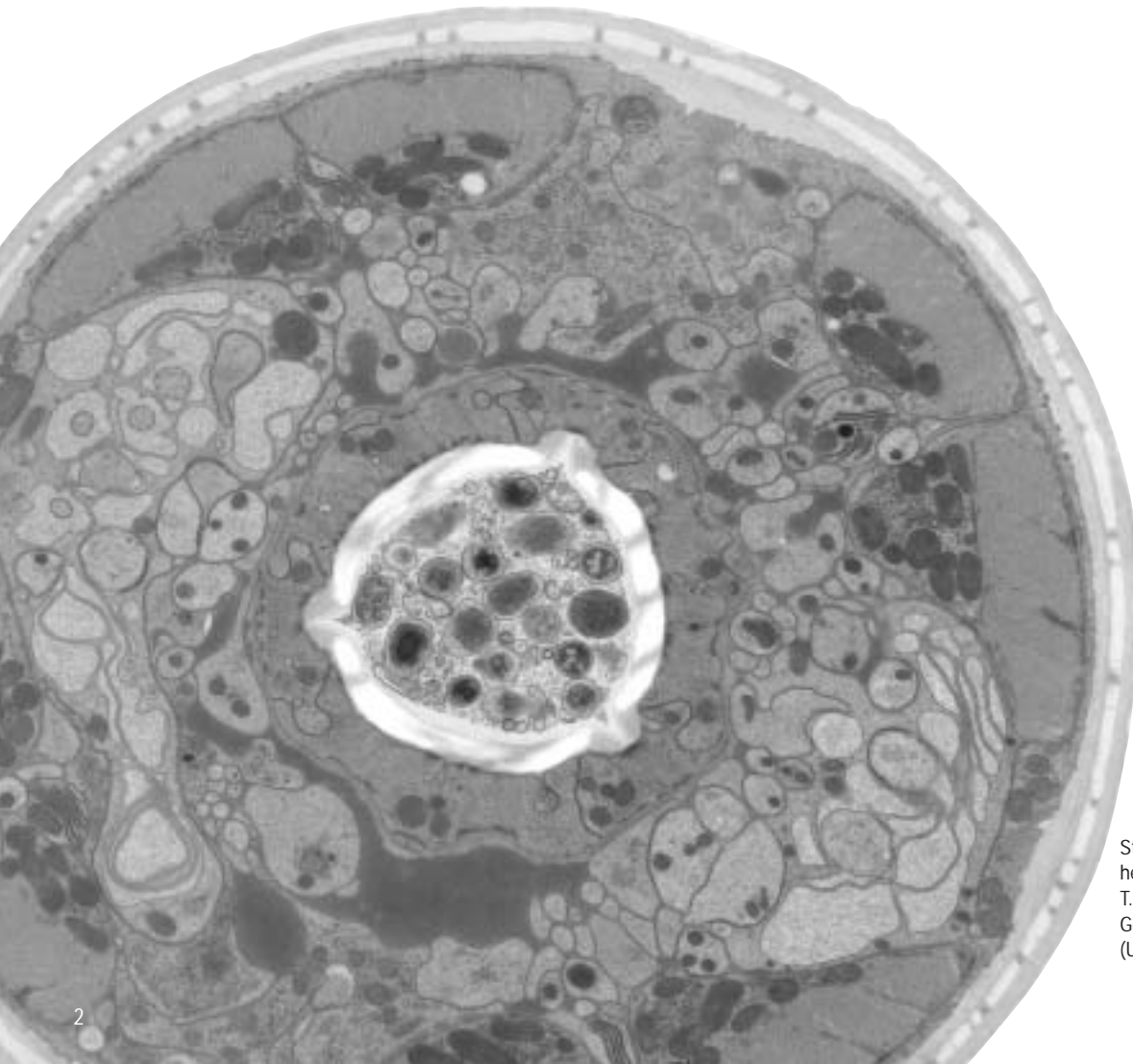
Leica EM RTS

Rapid Transfer System

High Pressure Freezing for Everybody

The Leica EM PACT2 high pressure freezer serves the needs of molecular and cell biologists and all researchers who want an “in vivo” impression of their cellular structures and functions in question – without the artefacts of chemical fixation but with the high resolution information of EM immunocytochemistry, frozen hydrated sections and freeze fracturing.

The Rapid Transfer System EM RTS allows correlative LM/EM experiments, taking a live specimen from a light microscope (e.g. a confocal microscope) to freezing in less than 5 seconds. In the same way, time resolved experiments are possible. Safety and reproducibility for the specimen are increased while operator mistakes are reduced.



Structural details of the *C. elegans* head in cross-section.
T. Müller-Reichert (MPI-CBG, Dresden, Germany) and Kent McDonald (University of California, Berkeley, USA)





Leica Design by W. Hölbl

Leica EM PACT2 – High Pressure Freezer



Perfect results

- High cooling rates by strong jet of LN₂
- 120 samples can be frozen per hour
- Variety of specimen carriers available for all purposes

Easy to install

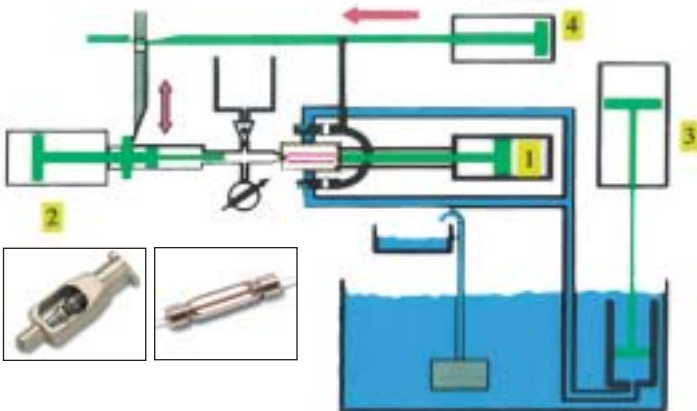
- Compact and mobile
- Standard electrical supply
- Compressor on EM PACT2 trolley

Easy to use

- Touch-sensitive color screen, menu prompts
- Bayonet loading device for automatic orientation
- Specimen ejection into LN₂ bath
- LED bath illumination
- 600 ms temperature/pressure curve displayed for each run
- Internal memory for 8 000 freezing runs
- Data download on memory stick

Safe and convenient

- Low LN₂ consumption
- Dewar with drain outlet
- Universal hydraulic system – can even use water
- Low noise
- Maintenance free “long life pods”



Function: All You Need is LN₂...

The specimen is locked (1) and set under pressure (2) just before freezing (3) via the synchronization mechanism (4). Only the tissue in the specimen holder is under pressure and not the complete specimen chamber. As any fluid can be used in the universal hydraulic system, toxic vapors (e.g. ethanol) are avoided.

Leica EM RTS – Rapid Transfer System

Perfect results

- Specimen carrier loading in less than 5 secs
- Correlative LM/EM
- Rapid biopsy process

Easy to install

- Factory mounted on EM PACT2
- Control via touch sensitive screen

Easy to use

- Rapid loader for inserting specimen
- Automatic and reproducible specimen freezing

Safe and convenient

- Outlet Dewar
- Safety lid
- Low noise
- Funnel for filling LN₂
- Platform for cryopreparation, box, position for accessories



Schematic drawing showing the status of all parameters



Freezing data after ejection of the sample

Leica EM RTS

Application Solutions

Specimen loading made simple with the Rapid Transfer System: insert specimen carrier into rapid loader. Automatically freeze by inserting rapid loader into RTS.

Prepare your sample (eg leaf or a cell monolayer on a sapphire disc) ...

... to fit into one of the various specimen carriers (eg flat carrier \varnothing 1.2 mm, depth 200 μ m or membrane carrier: \varnothing 1.5 mm, depth 200 μ m). Specimen and carrier are held in the rapid loader. The long life pod is connected to the loading device with a bayonet lock.

Specimen and carrier are held in the rapid loader.

The long life pod is connected to the loading device with a bayonet lock.

The primed loading device then waits for freezing in the RTS.

By gently pushing the rapid loader into the RTS the carrier is tightened securely and then frozen automatically in less than 2.5 seconds.

After cryofixation the carriers are collected in the LN₂ bath of the EM PACT2 before freeze substitution in the Leica EM AFS2.





Leica Design by W. Hölbl

The Tube Holder for every fluid you can think of ... blood, milk, cell suspensions ... nematodes – and more!

Directly suck the suspension into copper tubes (inner \varnothing 350 μm) already mounted in their holder.

Alternatively, take up sample into cellulose microcapillary.

With the loading device the specimen can be inserted into the EM PACT.

After cryofixation the samples are stored under LN_2 in the transfer box.

Sample Punch: punching out of the central part of the specimen tube ready for Frozen Hydrated Sectioning or ...

... peeling away of the top of the copper tube for Freeze Substitution.

Storage of the copper tubes is carried out under LN_2 .

The specimen tube holders are recycled by reloading them with copper tubes.



Leica EM RTS

Application Solutions

Taking microbiopsies for EM is now faster than ever before!

Prepare the Microbiopsy Transfer Station for RTS under the Optical Workstation...

... before taking a biopsy with the biopsy gun.

Insert the biopsy gun into the transfer station ...

... and transfer the tissue into the biopsy carrier.

The specimen and carrier are held in the rapid loader ...

... while the primed loading device waits for freezing in the RTS.

By gently pushing the rapid loader into the RTS the carrier is tightened securely and then frozen automatically in less than 2.5 seconds.

After cryofixation the carriers are collected in the LN₂ bath of the EM PACT2 before freeze substitution in the Leica EM AFS2 or cryosectioning in the EM FC6.





Frozen Hydrated Sectioning made easy.

Alternatively, the carriers can be trimmed with a cutter ...



... for Frozen Hydrated Sectioning in the cryoultramicrotome.



Preparation for follow-on procedures.

All necessary steps for follow-on procedures (eg. Freeze Substitution) can be conveniently performed in the cryopreparation box supplied with the Leica EM PACT2.



The Freeze Fracture Holder for everything that can be fractured ...

Prepare the Freeze Fracture Station under the Optical Workstation ...



... and preload the freeze fracture carrier. A copper ring is put on top of the loaded carrier.



The carrier and copper ring are sandwiched securely in the pod with the supplied torque wrench.



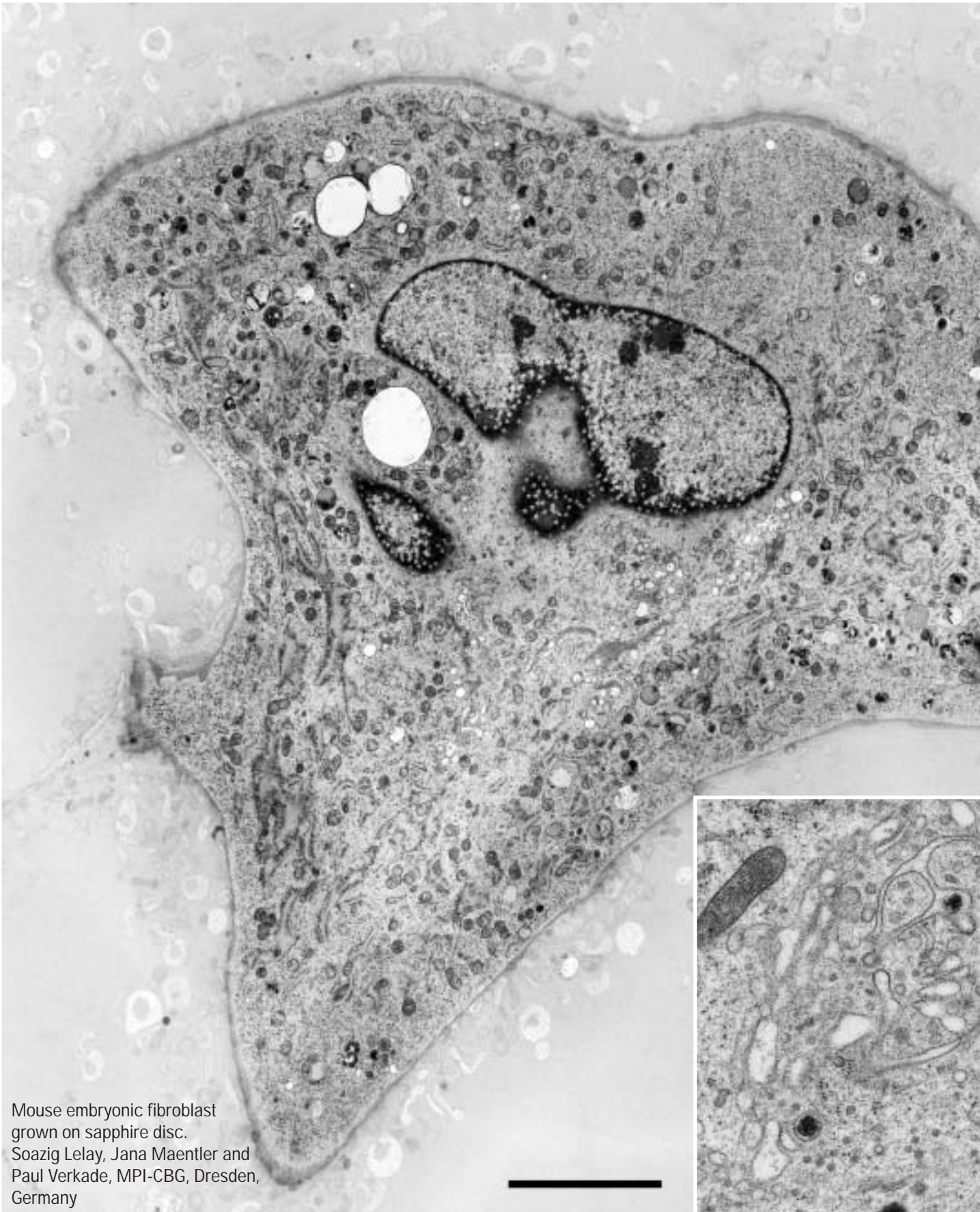
With the loading device the specimen can be inserted into the instrument.



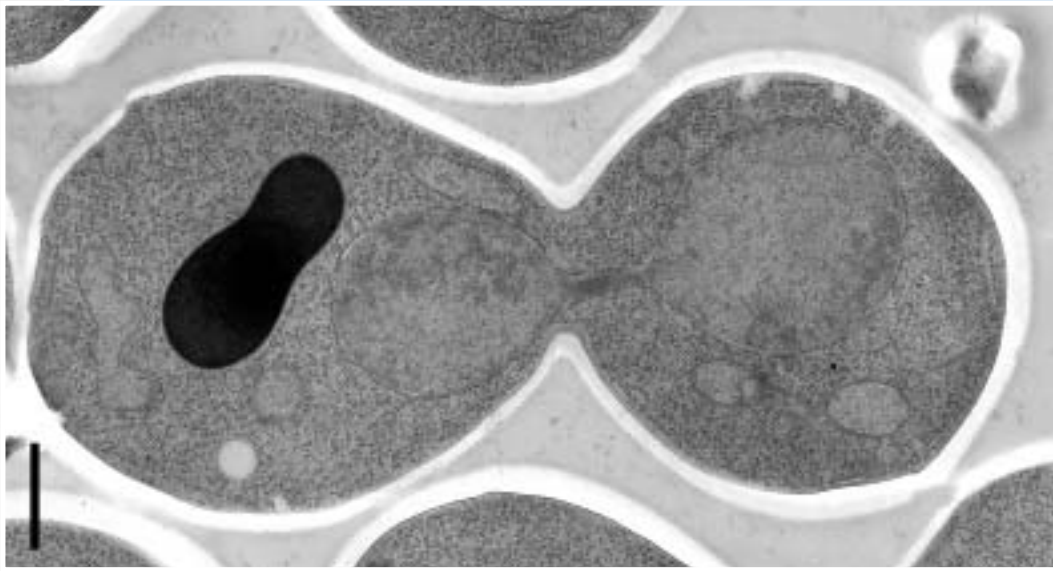
After cryofixation the carriers are collected in the LN₂ bath of the EM PACT2 before Freeze Fracturing.



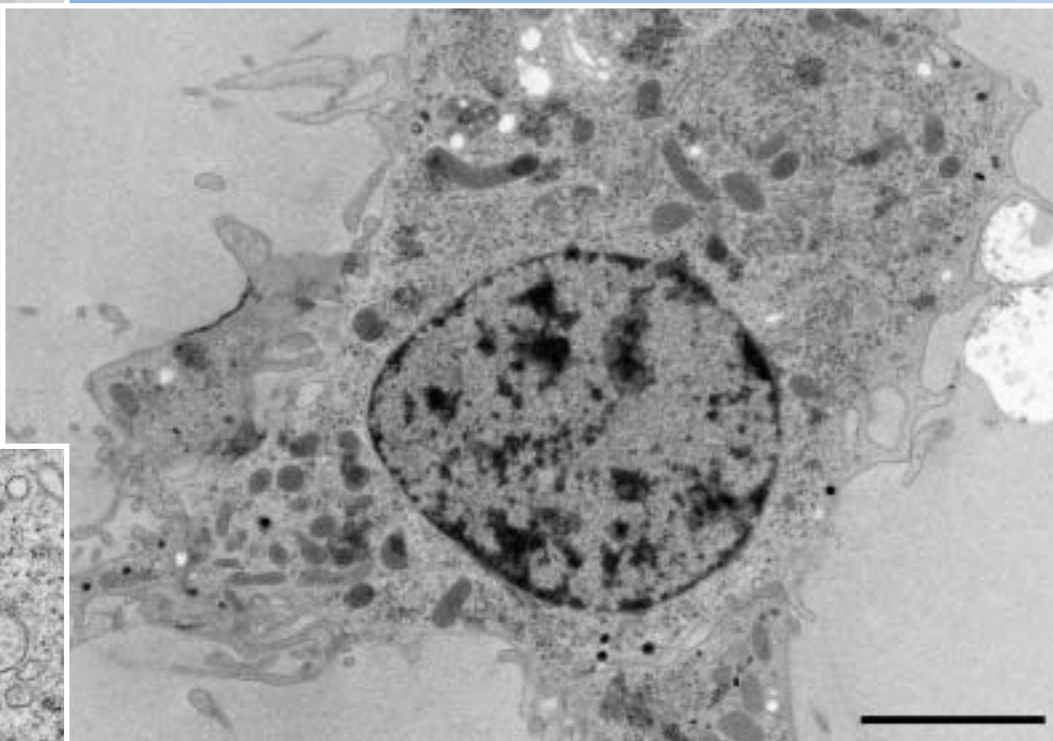
Accessories are also available for loading EM PACT2 without EM RTS.



Mouse embryonic fibroblast
grown on sapphire disc.
Soazig Lelay, Jana Maentler and
Paul Verkade, MPI-CBG, Dresden,
Germany



Yeast frozen in the membrane carrier. Scale bar = 500 nm, 250 nm for insert.
 Courtesy of Mark van Breugel, Jana Maentler and Paul Verkade, MPI-CBG, Dresden, Germany.



Insulin granules producing INS-1 cells. Scale bar = 5 μ m.
 Courtesy of Joke Ouwendijk, Jana Maentler, Melanie Jäger, Michele Solimena and Paul Verkade, TUD and MPI-CBG, Dresden, Germany



Rat, Langerhans cells. Scale bar = 1 μ m
 Courtesy of Joke Ouwendijk, Jana Maentler, Melanie Jäger, Michele Solimena and Paul Verkade, TUD and MPI-CBG, Dresden, Germany

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