

## Aquilos 2 Cryo-FIB

### A second-generation, dedicated cryo-FIB microscope to prepare cryo-lamellas for cryo-electron tomography

The Thermo Scientific™ Aquilos™ 2 Cryo-FIB is a cryo-dedicated system that delivers exceptional sample preparation for high-end cryo-TEM tomography.

#### Cryo-EM dedication

The Aquilos 2 Cryo-FIB (Cryo-Focused Ion Beam) allows you to actively gain control over your sample thickness and prepare thin *in situ* cryo-lamellas from cellular specimens grown on EM specimen supports. The microscope has an integrated, fully rotatable cryo-stage and adjacent cryo-hardware that protects the frozen-hydrated samples from contamination, ensuring that delicate life sciences cryo-samples will be kept at vitrified temperatures.

#### New versatile features

The Aquilos 2 Cryo-FIB's extended operation time allows you to work longer with the system under cryogenic conditions and enables automation of the lamella manufacturing process.

Thermo Scientific™ AutoTEM™ Cryo Software facilitates automated batch milling of cryo-lamellas to increase productivity. The guided software allows you to select multiple points of interest and then automatically prepares several lamellas autonomously in unattended runs, including overnight. Thermo Scientific™ AutoScript™ Software is also included, allowing you to create your own lamella manufacturing and imaging scripts independently and in a flexible manner via advanced programming and scripting interfaces.

The Aquilos 2 Cryo-FIB is available for the first time with the new optional Thermo Scientific EasyLift NanoManipulator cryo-lift-out system, enabling you to prepare lamellas from target regions within bulk-frozen specimens. With the EasyLift NanoManipulator, site-specific regions, even from high-pressure frozen samples, can be extracted and placed inside AutoGrids for further imaging in a cryo-TEM.

Thermo Scientific™ Auto Slice & View™ Software (AS&V) allows you to acquire 3D images under cryogenic conditions by sequentially milling then imaging a cross-sectioned area, such as the interior of a vitrified cell. Auto Slice & View Software allows you to define a milling endpoint and preview the desired subcellular region before viewing ultrastructural detail with higher resolution in a cryo-TEM.

#### Key benefits

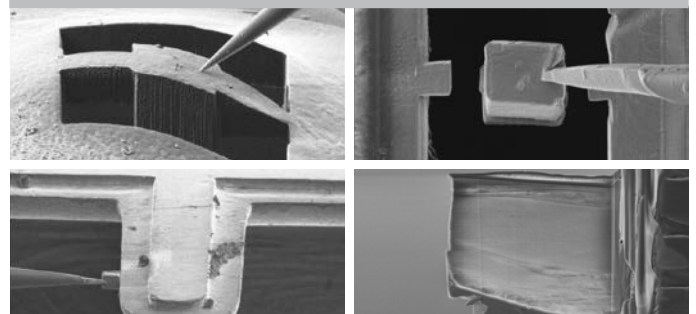
Produce cryo-lamellas for cryo-tomography using autoloader TEM systems. Cryo-dedicated hardware inside the chamber ensures minimal sample contamination, sample damage, and loss of correlation accuracy.

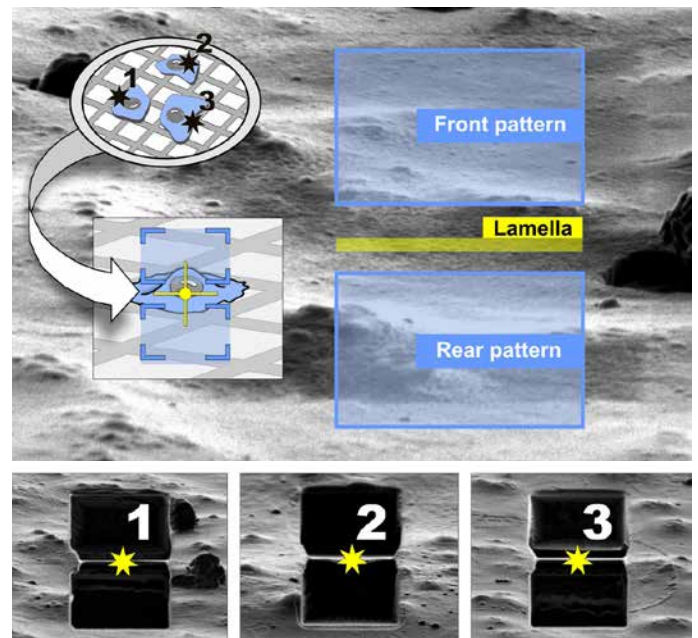
Create thinner cryo-lamellas without cutting artifacts. Ion beam milling enables the creation of compression-free cryo-lamella samples for TEM tomography imaging. Using a cryo-FIB avoids artifacts, such as mechanical compression, that are inevitable for mechanical cryo-sectioning using a cryo-ultramicrotome.

Improve sample preparation precision. Guided User Software and Maps Software for correlation makes the Aquilos 2 Cryo-FIB easy to use, even for new users. Import light microscopy data into Maps Software for identification of features of interest, targeting, or correlation over different imaging modalities.

Automate milling and discover cryo-lift-out. Explore state-of-the-art automation software for cryo-lamella production and work with challenging bulk-frozen samples that require cryo-lift-out sample preparation.

Run overnight operations. Longer running times at cryogenic temperatures enable overnight use of the system for further automation of cryo-lamella production.





### Cryo-tomography workflow connectivity

The Aquilos 2 Cryo-FIB is specifically designed for use within the cryo-tomography workflow. This workflow includes a Thermo Scientific™ Vitrobot™ System and an autoloader-supported cryo-transmission electron microscope, such as the Thermo Scientific™ Krios™ Cryo-TEM. Using cross-system hardware and software solutions, including Thermo Scientific™ Maps™ Software, cryo light microscopy data can be imported for targeting and then used to calculate milling positions. The finished lamella positions can, in turn, be imported into the TEM.

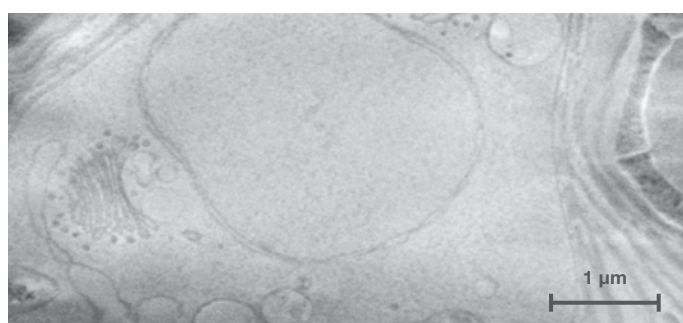
The Aquilos 2 Cryo-FIB offers an integrated link to the correlative cryo-light microscopy solutions from Leica Microsystems. A software import solution and a common sample carrier with a dedicated shuttle facilitate the import and correlation of data between Leica Cryo-LM and Cryo-FIB for a simple and straightforward correlative workflow.

### Integrated sputter coater

A retractable sputter coater is integrated within the Aquilos 2 Cryo-FIB chamber to allow for deposition of a nanometer-thin inorganic platinum layer onto the milled cryo-lamellas. This sputter coater ensures efficient coating operations and that the vitrified sample does not need to be transferred to an external sputter coating device. The coating renders the cryo-lamellas conductive, preventing charge-up during cryo-tomography. In particular, charging can obstruct tomographic image acquisition when using a Volta phase plate. All sputter coater controls are embedded in the cryo-FIB user software.

### Flexibility and ease of use

The Aquilos 2 Cryo-FIB supports the preparation of cryo-lamellas by providing dedicated software guidance and easy-to-use setup and alignment procedures. This support enables you to quickly familiarize yourself with the system and get up to speed in cryo-sample preparation and lamella milling.



### System features

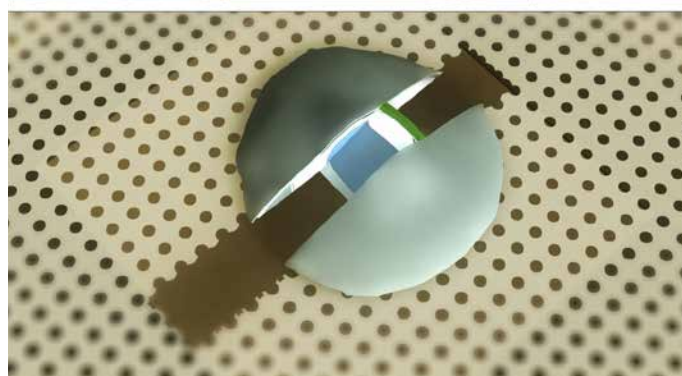
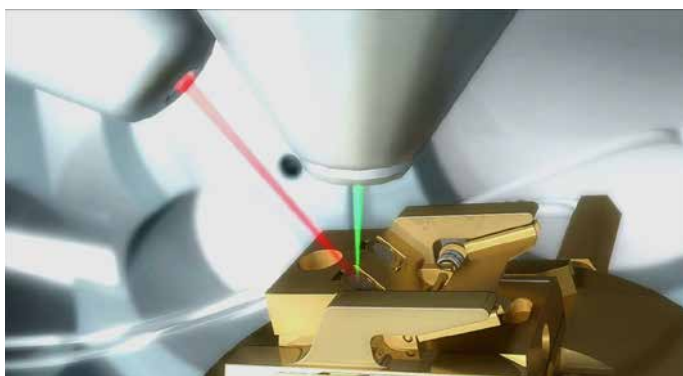
- Complete cryo-infrastructure: large-capacity liquid nitrogen Dewar for extended runtimes, heat exchanger, flow controller, load-lock system, sample preparation station, controller, and transfer device
- Fully rotatable cryo-stage (<-170°C at the sample), keeping samples at vitreous temperatures
- Special sample holders customized for shallow-angle milling of EM grids, referred to as Cryo-FIB AutoGrids (autoloader approved)
- Sample shuttle for AutoGrids: Cryo-FIB shuttle with integrated shutter system during cryo-transfers
- In-chamber retractable sputter coater for applying conductive coatings
- Gas injection system (GIS) for applying protective coatings
- Includes cryo-FIB consumables kit: tweezers, clipping and grid box tools, AutoGrid boxes, C-Clips and Cryo-FIB AutoGrids
- Seamless compatibility and connectivity with our cryotomography workflow
- Optional EasyLift NanoManipulator for cryo-lift-out

### Included software

- Maps Software, including image correlation and lamella preparation functions
- AutoTEM Cryo Software for milling automation
- AutoScript Software for custom scripting
- Auto Slice & View Software for 3D volume imaging

### Instrument features

- NiCol UHR non-immersion field emission-SEM column
- In-lens detection system: segmented lower (T1) and upper detector (T2)
- Everhart-Thornley SE Detector (ETD)
- Sidewinder ion column
- Workstation with Windows 10 + two 24-inch LCD monitors



- Support computer with Windows 10 + one 24-inch LCD monitor
- Large table top with support
- xT software with more dedicated cryo functionality
- 110×110 mm eucentric stage
- IR camera for viewing sample and chamber
- In-chamber Thermo Scientific™ Nav-Cam™ Camera for sample navigation
- Complete oil-free pumping system
- Integrated beam current measurement
- Automatic aperture system
- Optional extra acoustic enclosure for XDS pump

### Cryo package features

- Rotatable cryo-stage (cooldown time: <20 minutes)
- Cryo-preparation station (including AutoGrid clipping insert)
- Controller for preparation station
- Sample transfer device (transfer rod)
- Cryo-loader (SEM load lock for transfer rod)
- Two standard Cryo-FIB AutoGrid shuttles
- Optional Leica CLEM shuttle/kit
- Standard shuttle (for SEM alignment specimens and pin mount stubs)
- Large-capacity liquid nitrogen Dewar
- Electronic integrated nitrogen gas flow controller
- Heat exchanger (delivers cooling gas)
- Oil-free pump for heat exchanger
- Dry diaphragm pump for cryo-preparation station
- Cryo-consumables kit with 20 AutoGrid boxes, 100 Cryo-FIB AutoGrids, 100 C-Clips, 2 clipping tools, 2 AutoGrid tweezers, 2 grid box openers, 2 soft grip tweezers, forceps, crossover and stub tweezers
- Platinum deposition gas injection system (GIS) for deposition of protective layers
- In-chamber and automated retractable sputter coater (target: platinum) for deposition of conductive layers
- Sample bake-out box for cleaning shuttles and samples
- Safety wear kit for maintenance
- Integrated temperature logger
- Acoustic enclosure for heat exchanger scroll pump

## Technical specifications

### Electron Optics

- High-stability Schottky field emission gun
- Minimum source lifetime: 12 months
- Easy gun installation and maintenance: auto bakeout, auto start and no mechanical alignments
- Continuous beam current control and optimized aperture angle
- Double stage scanning deflection
- Dual objective lens, combining electromagnetic and electrostatic lenses
- User guidance and column presets
- Beam current range: 1.5 pA to 400 nA
- Accelerating voltage range: 200 V – 30 kV
- Resolution (with cryo-stage): 1.6 nm at 30 kV, 2.6 nm at 2 kV (at room temperature), 6.0 nm\* at 2kV (at cryo-temperature)

### Ion Optics

- Source lifetime: 1,300 hours
- Voltage: 500 V to 30 kV
- Beam current: 1.5 pA – 65 nA in 15 steps
- Drift suppression mode as standard for non-conductive samples
- Resolution (with cryo-stage): 7.0 nm\* at 30 kV

### Vacuum system

- Complete oil-free vacuum system
- 1× TMP with turbo drag section, 240 l/s
- 1× Scroll pump
- 3× IGP, 25 l/s
- Chamber vacuum at room temperature: <4e-4Pa
- Chamber vacuum at cryo-conditions: <8e-5Pa

### Cryo-stage

- Integrated rotatable cryo-stage
- Rotation: 360° (endless)
- Compucentric rotation and tilt
- Cooldown time: <20 minutes
- XY range: 110 mm
- Z range: 65 mm
- Tilt range at cryo (eucentric WD): -15° to +55°

\*Cryo-imaging conditions subject to gas flow rates used.

## Image processor

- Dwell time range from 25 ns to 25 ms/pixel
- Up to 6144×4096 pixels (up to 64k through Maps Software)
- File type: TIFF (8-, 16-, 24-bit), BMP or JPEG standard
- Electronic scanning rotation: 360° degrees
- Thermo Scientific SmartSCAN System (256 frame average or integration, line integration and averaging, interlaced scanning)
- DCFI (Drift Compensated Frame Integration)

## Accessories

- Seismic restraint kit
- Acoustic enclosure for microscope scroll pump
- Additional preparation station
- Remote control and imaging
- Additional cryo-consumables and accessories, including cryo-FIB AutoGrids and hot plate
- Leica CLEM kit (includes shuttles and tools required to work with the Leica EM Cryo-CLEM kit)
- EasyLift NanoManipulator for cryo-lift-out. Dedicated easy lift variant with -165°C at the needle tip and <200nm/min drift after insertion.
- Thermo Scientific™ iFLM™ Correlative System combines fluorescence light and electron microscopy into one system

## Warranty and training

- 1-year warranty
- Choice of service maintenance contracts
- Choice of operation / application training contracts
- Standard Aquilos Cryo-FIB training for tool users
- Optional advanced training course
- Customer witness acceptance test (CWAT)

## Documentation and support

- Online user guidance
- User operation manual
- Prepared for Thermo Scientific RAPID Remote Diagnostics
- Free access to online resources

## System control

- 64-bit GUI with Windows10, keyboard, optical mouse
- Up to four live images showing independent beams and/or signals. Live color signal mixing.
- Local language support. Check with your local Thermo Fisher Scientific sales representatives for available language packs.

- 3 × 24-inch widescreen monitors with 1920×1200 pixels
- Multifunctional control panel (MUI)

## Supporting and application software

- AutoTEM Cryo Software for on-the-grid batch lamella automation
- AutoScript DB Software for full and flexible control of the system through scripting
- Auto Slice & View Software for 3D volume imaging applications
- Maps 3 Software for tiling and stitching (import 120+ image data formats, up to 64k × 64k), correlation of LM and SEM data (software assists in computation of eucentric position for milling and fast retrieval of ROIs)
- XT software with dedicated cryo-controls
- “Beam per view” graphical user interface concept, with up to four simultaneously active quads
- Thermo Scientific SPI Software (simultaneous FIB patterning and SEM imaging)
- Thermo Scientific iSPI Software (intermittent SEM imaging and FIB patterning)
- Thermo Scientific iRTM Software (integrated real-time monitor) and FIB immersion modes for advanced, real-time SEM and FIB process monitoring and endpointing
- Patterns supported: rectangle, line, circle, cleaning, cross-section, regular cross-section, polygon, bitmap, stream file, exclusion zones, arrays
- Sample navigation on an optical image
- Undo / Redo functionality
- User Guidance for most common system operations and applications



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