Aquilos 2 Cryo-FIB

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

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

**Cryo-EM dedication**
The Thermo Scientific™ 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 science 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 the selection of 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 and 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. AS&V allows you to define a milling endpoint and preview the desired subcellular region before viewing ultrastructural detail with higher resolution in a cryo-TEM instrument.

**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 you in 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 and View Software for 3D volume imaging

Instrument features

- NiCoI 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
• 110x110 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
*Cryo-imaging conditions subject to gas flow rates used.

**Vacuum system**
• Complete oil-free vacuum system
• 1x TMP with turbo drag section, 240 l/s
• 1x Scroll pump
• 3x 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°
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
- High-performance ion conversion and electron (ICE) detector for secondary ions (SI) and secondary electrons (SE)
- 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.

System control
- 64-bit GUI with Windows 10, 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 x 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 and 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 DualBeam system operations and applications

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 DualBeam™ 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

Find out more at thermofisher.com/EM-Sales