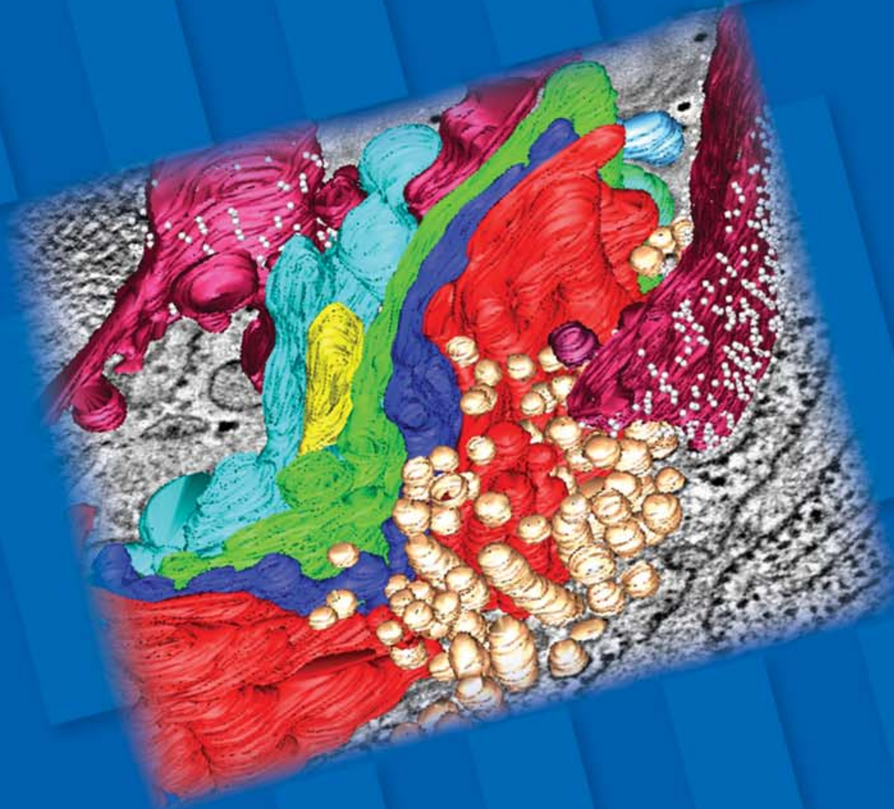


Model 2000 Series

Tomography Specimen Holders

The Model 2000 TEM Tomography Holder Series brings TEM into the three-dimensional world. These advanced TEM specimen holders are from the tomography experts at Fischione. The series includes specimen holders that allow high tilt and extended field of view. They are available in single-, dual-, and on-axis versions and are compatible with even the narrowest pole piece geometries.

FISCHIONE INSTRUMENTS

EXCELLENCE...MAGNIFIED

Model 2020 Advanced Tomography Holder (U.S. Patent 7,219,565)

Model 2020 Advanced Tomography Holder
(shown in stand)

- ***Ideal for room temperature electron tomography.***
- ***High tilt angles.***
- ***Maximized field of view.***
- ***Optimized specimen clamping.***
- ***Easy, accurate specimen loading and centering.***

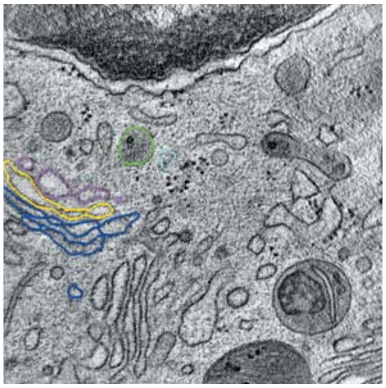
Fischione Instruments Model 2020 Advanced Tomography Holder for transmission electron microscopes (TEMs) features high tilt and extended field of view. The revolutionary holder allows room temperature data collection over wide tilt and translation ranges even in restrictive pole-piece-gap geometries.

The Model 2020 is for life sciences and physical sciences as well as any other applications requiring high specimen tilt and simultaneous large field of view.

Three-dimensional information

Advances in microstructural characterization require the ability to analyze structure and chemistry in three dimensions. However, most TEM techniques are limited to producing two-dimensional information. Tomography, on the other hand, combines two-dimensional data sets taken at various tilt angles to produce three-dimensional information.

Biological research has benefited from the use of electron tomography for many years; however, the physical sciences have been limited by the inability to tilt the specimen to high angles within the confines of the narrow-gap pole pieces necessary for atomic resolution imaging. In addition, there was an increasing desire to use HAADF-STEM to reduce diffraction contrast in physical science applications. Now, Fischione's advanced specimen holder technology enables room temperature tomography for both the life and the physical sciences.



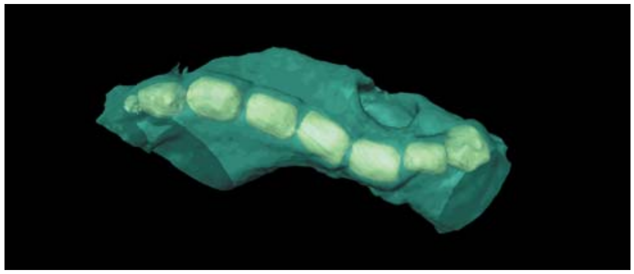
Reconstruction of dendritic cell golgi and lysosomal structures. 3D manual contouring model (right) taken of high-pressure-frozen and freeze-substituted dendritic cell. Angular tilt range from –65° to +65° with 1° increments. The tomogram shows part of a Golgi stack with different cisternae and the presence of endosomal structures.

Image courtesy of W.J.C. Geerts, B. Humbel, and A.J. Koster from the Department of Molecular Cell Biology, Institute of Biomembranes, Utrecht University, The Netherlands, who assisted with the specimen preparation, recording, and processing the tilt series to generate the 3D images. J-L. Murk and M. Kleijmeer from the Department of Cell Biology, University Medical Center Utrecht, The Netherlands, provided the dendritic specimen.

Maximized field of view at tilt angles above 70° with no shadowing

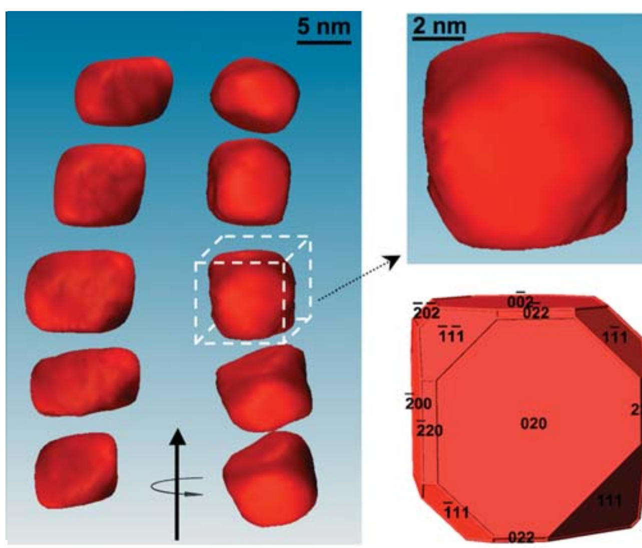
The Model 2020's streamlined specimen clamping mechanism eliminates the shadowing associated with most holders at high tilt angles.

Model 2020 Advanced Tomography Holder



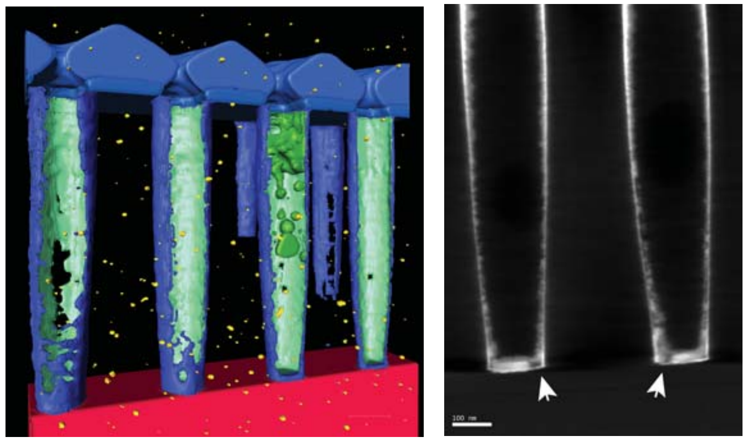
Tomographic reconstruction of magnetic particles from magnetotactic bacteria. Tomographic tilt series taken using Fischione Model 3000 high angle annular dark field (HAADF) STEM detector. Angular tilt range from -76° to +76° with 4° increments. The quality of the reconstruction allows both the crystals (yellow) and the exterior cell wall (green) to be resolved. These intracellular chains of magnetic crystals are of interest to astrobiologists, as similar structures have been observed in Martian meteorites, and to the magnetic media industry, as these biogenic magnets are of higher quality than those currently used for data storage.

Image courtesy of M. Weyland, P.A. Midgley and Rafal Dunin-Borkowski from the Department of Materials Science and Metallurgy, University of Cambridge, United Kingdom, who supplied the HAADF STEM tomographic data series. Dennis A. Bazylinski from Microbiology Faculty, Iowa State University and Richard Frankel from the Physics Department, California Poly State University, U.S.A., supplied the magnetotactic bacterial specimen.



Three-dimensional electron tomography of colloidal cerium oxide (CeO2) nanocrystals prepared by hydrothermal synthesis. CeO_2 is used as an oxygen conductor in solid oxide fuel cells. Electron tomography observations were conducted with an FEI Tecnai F20 operated at 200kV in STEM mode using the Fischione Model 3000 HAADF STEM detector. A series of projections was acquired by tilting from –70° to +74° with an image recorded every 2°. These rendered images indicate the presence of {111} facets at each corner and {220} facets at each edge.

Image courtesy of K. Kaneko from the Department of Materials Science & Engineering, Kyushu University, Japan, K. Inoke from the FEI Company Japan Ltd., Japan, B. Freitag from the FEI Company, The Netherlands, A.B. Hungria and P.A. Midgley from the Department of Materials Science and Metallurgy, University of Cambridge, United Kingdom, T.W. Hansen from the Department of Inorganic Chemistry, Fritz-Haber-Institute, Germany, J. Zhang, S. Ohara, and T. Adschiri from the Institute of Multidisciplinary Research for Advanced Materials, Tohoku University, Japan.

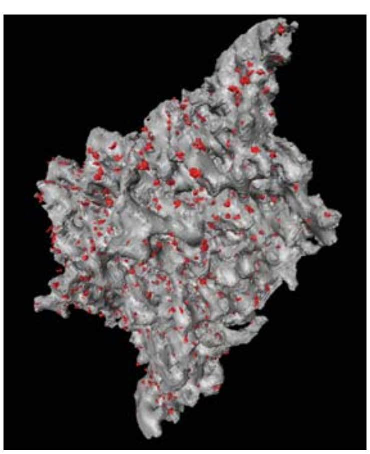


3D imaging of high aspect-ratio VIAs. Surface rendering of two rows of VIAs imaged on top of each other (left) and a digital slice through the 3D volume at higher magnification (right). The shape and uniformity of the barrier and the copper seed layer can be estimated based on the tomographic reconstruction and small defects detected in 3D (two examples marked by arrows). In the surface rendering, the barrier layer is shown in blue and the copper seed layer and some larger copper crystals in green. The silicon substrate is rendered in red and the gold markers in yellow.

Images of this 300nm thick, FIB prepared section were acquired on an FEI Tecnai F20ST using the Fischione Model 3000 HAADF STEM detector. 129 images were recorded automatically over a tilt-range of ±75° using the Xplore3D Tomography Suite. The 3D volume was reconstructed using the SIRT algorithm implemented in Inspect3D with 20 iterations. The visualization was performed in Amira after semi-automatically segmenting the 3D volume.

Images courtesy of C. Kübel, Fraunhofer Institute for Applied Materials Sciences and Production Technology, Bremen, Germany.

Reprinted with permission from C. Kübel, J. Kübel, S. Kujawa, J.-S. Luo, H.-M. Lo, J.D. Russell "Application of Electron Tomography for Semiconductor Device Analysis" in '8th International Workshop of Stress-Induced Phenomena in Metallization, edited by E. Zschech, K. Maex, P.S. Ho, H. Kawasaki, T. Nakamura, AIP Conference Proceedings 817, page 223-228, American Institute of Physics, Melville, New York, 2006.

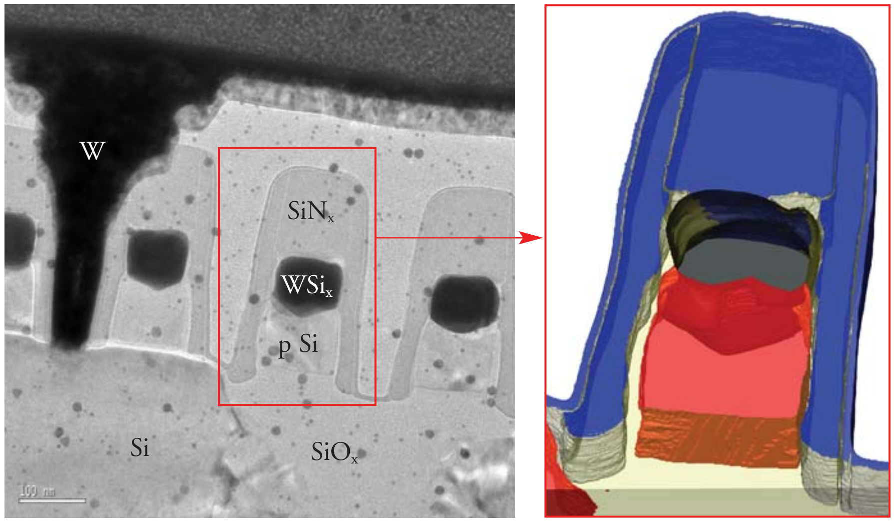


Surface rendering of a Ru-Pt catalyst supported on and within disordered porous silica. The Ru-Pt particles are seen in red, and the silica is rendered as solid gray.

Images were acquired with an FEI Tecnai F20 field emission gun transmission electron microscope (FETEM) at 200kV in scanning transmission electron microscope (STEM) mode using the Fischione Model 3000 HAADF STEM detector. Images were recorded every 1° from +78° to –78°. Image acquisition and reconstruction were accomplished using the FEI software suite, Xplore3D, with visualization performed by Amira.

Image courtesy of T.J.V. Yates, E.P.W. Ward, and P.A. Midgley from the Department of Materials Science and Metallurgy, University of Cambridge, United Kingdom.

Model 2020 Advanced Tomography Holder



Electron tomographic analysis of a DRAM device. TEM reference image of the overall DRAM device and surface rendering showing the detailed structure of a selected transistor (Si - red, SiNx - blue, SiOx/SiOxNy - yellow, WSix - black). The tomographic reconstruction reveals the 3D structure of the transistor and even thin, ~1.5nm wide, silicon oxynitride layers are visible in the 3D reconstruction – surrounded by silicon nitride and other oxynitride layers as well as in the direct vicinity of the tungsten plug. The surface rendering reveals a minimal roughness for these thin oxynitride layers, but shows a significant surface roughness of the tungsten silicide and the polysilicon.

Images were acquired on an FEI Tecnai F20ST in TEM mode close to Lichte focus without an objective aperture. 133 images were recorded automatically over a tilt-range of ±75° using the Xplore3D Tomography Suite. The tilt-series was aligned in Inspect3D by a combination of cross-correlation and marker tracking and the 3D volume reconstructed using the SIRT algorithm with 20 iterations. The visualization was performed in Amira after manually segmenting the 3D volume.

Images courtesy of C. Kübel, Fraunhofer Institute for Applied Materials Sciences and Production Technology, Bremen, Germany and J.-S. Luo, H.-M. Lo and J.D. Russell, Inotera Memories, Taoyuan, Taiwan.

Reprinted with permission from C. Kübel, J. Kübel, S. Kujawa, J.-S. Luo, H.-M. Lo, J.D. Russell "Application of Electron Tomography for Semiconductor Device Analysis" in '8th International Workshop of Stress-Induced Phenomena in Metallization, edited by E. Zschech, K. Maex, P.S. Ho, H. Kawasaki, T. Nakamura, AIP Conference Proceedings 817, page 223-228, American Institute of Physics, Melville, New York, 2006.

Accepts wide range of specimen thicknesses

The Model 2020's clamping mechanism accepts specimen grids, standard 3mm diameter TEM specimens, or FIB lamella. The Model 2020 accommodates specimen thicknesses up to 250 microns.

Evenly distributed force from fully retractable clamps

Specimens are secured with two clamps that produce an evenly distributed force on opposing edges of the specimen. For convenience during loading and unloading, the clamps are spring-loaded to lift them off the specimen surface. Then, they can be fully retracted. Positioning the clamps is done without contacting the specimen, eliminating the possibility of specimen damage. This is far superior to typical clamping mechanisms that limit the specimen size or interfere with viewing at high tilt angles.

Easy, accurate, self-centering positioning

The tapered self-centering specimen receptacle guides the specimen into position. The fully retractable clamps make it easy to rotate the specimen manually for a dual-axis tilt series.

Model 2020 Specifications*

Holder type	Room-temperature, single-tilt. For most TEMs.
Specimen size	3mm diameter. Wide range of thicknesses up to 250 microns.
Resolution	0.34nm (in all directions)
Drift	<1.5nm/min
Maximum tilt range	up to +/- 80°
Field of view	up to 1.6mm at 70°

*All specifications depend on the microscope model, pole piece type, and aperture position.
For ultimate resolution and drift performance, the TEM must meet the manufacturer's specifications.

Model 2030 Ultra-Narrow Gap Tomography Holder

Model 2030 Ultra-Narrow Gap Tomography Holder
(shown in stand)

- ***Room temperature tomography in ultra-high resolution microscopes.***
- ***Extended field of view at high tilt angles.***
- ***Can tilt up to 90°.***
- ***Suitable for ultra-narrow gap (<3mm) pole piece geometries.***
- ***Optimized specimen protection during insertion into the TEM.***

Tilts up to 90° in pole piece gaps less than 3mm

The ability to collect three-dimensional information over large ranges of tilt in room temperature transmission electron microscopy reaches a mechanical limitation dictated by the combination of specimen thickness, size, specimen support, pole piece geometry, aperture, and specimen holder configuration.

To obtain high tilt angles in transmission electron microscopes, the pole piece gap should be large and unrestricted. However, for some ultra-high resolution materials science microscopes, this gap can be less than 3mm. Hence, high tilt with traditional 3mm specimen geometry is not possible.

The Model 2030 Ultra-Narrow Gap Tomography Holder is capable of tilting up to 90° while providing a maximized field of view.

Advanced clamping and retracting mechanisms

The Model 2030 accepts a 1.5mm square or round TEM specimen or grid into a cartridge, securing it with a single clamp. The clamp can be fully retracted from the specimen receptacle for convenience during loading and unloading. When engaged, the clamp provides a uniformly distributed force on the edge of the specimen.

To protect the specimen during holder insertion into and removal from the TEM goniometer, the Model 2030 features an advanced mechanism that retracts the specimen cartridge into the body of the specimen holder.

A specimen loading station allows easy positioning and clamping of the specimen into the cartridge.

Plasma Cleaning

Cleaning the specimen and TEM specimen holder with the Fischione Model 1020 Plasma Cleaner before use is highly recommended.

During collection of tomographic data, the electron beam will be on the same area of the specimen for an extended time. As a result, organic contamination may build up on the specimen. A cleaning time of ten seconds to two minutes in the Fischione Model 1020 Plasma Cleaner removes the contamination. Longer cleaning times can remove contamination spots caused by previous TEM viewing of non-plasma cleaned specimens.

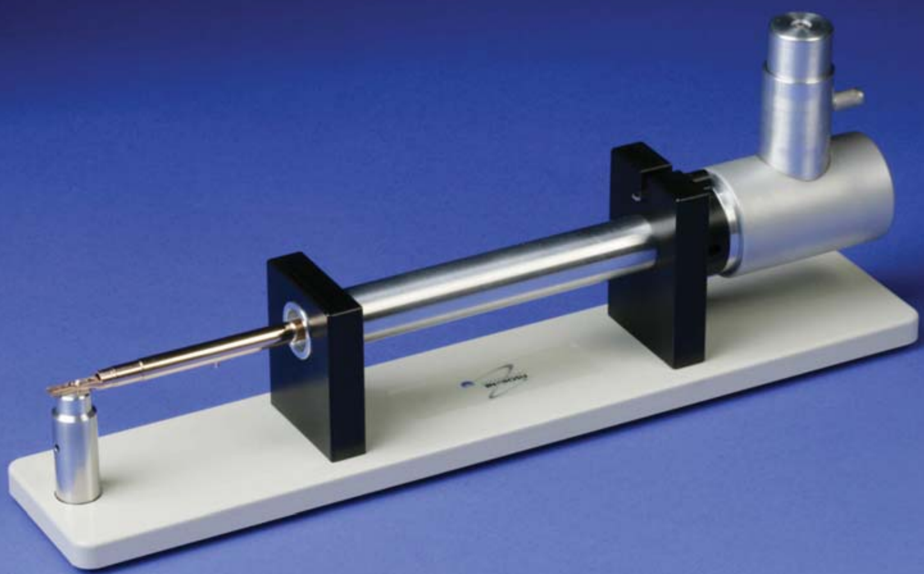
When not in use, the holders should be stored under vacuum in Fischione Model 9010 Vacuum Storage Containers or the Model 9020 Vacuum Pumping Station.

Model 2030 Specifications*

Holder type	Room-temperature. Single-tilt. For most TEMs.
Specimen size	1.5mm square or round. Wide range of thicknesses up to 100 microns.
Resolution	0.34nm (in all directions)
Drift	<1.5nm/min
Maximum tilt range	up to +/- 90°
Field of view	Not limited by holder

*All specifications depend on the microscope model, pole piece type, and aperture position. For ultimate resolution and drift performance, the TEM must meet the manufacturer's specifications.

Model 2040 Dual-Axis Tomography Holder



Model 2040 Dual-Axis Tomography Holder
(shown in stand)

- ***Fully-jeweled mechanism for ultra-precise planar specimen rotation.***
- ***Optimized tilt in pole piece gaps as small as 5mm.***
- ***Ideal for room temperature electron tomography.***
- ***Maximizes tomographic data obtained from the specimen.***
- ***Extended field of view.***
- ***FlexiClamp provides an easy, secure means of specimen retention.***

Precise, in situ planar rotation

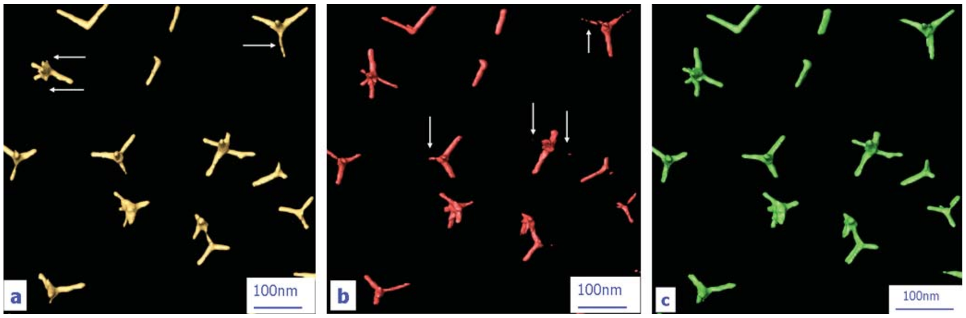
The Model 2040 Dual-Axis Tomography Holder is for TEM imaging or analysis that requires in situ specimen rotation. Acquiring a dual-axis tilt series enhances the information contained in the tomogram.

The Dual-Axis Tomography Holder features an optimal tilt angle range in narrow gap (~5mm) pole piece geometries, while maintaining microscope resolution.

A fully jeweled mechanism provides ultra-precise, in-plane specimen rotation, while maintaining eucentricity.

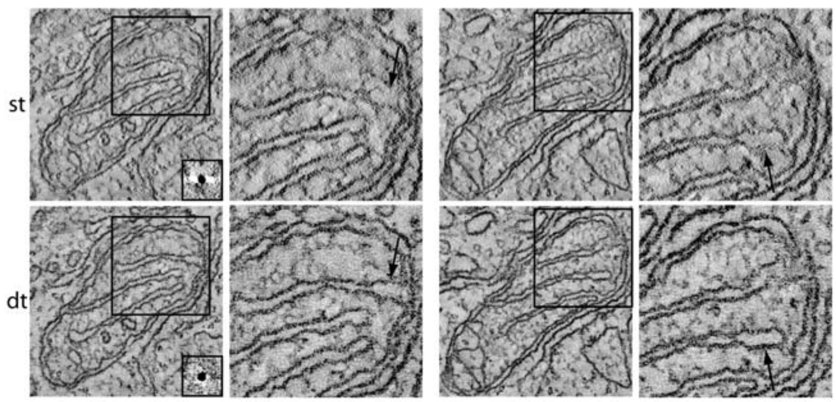
The FlexiClamp is a spring-type, annular ring which securely clamps the specimen into the specimen cup. It maximizes specimen visibility, even at high tilt angles. A dedicated tool facilitates the use of the FlexiClamp.

Initially, the specimen can be fully rotated through 360° to orient either the grid bars or a specimen feature to the alpha tilt axis. Once the specimen is properly oriented, the first tilt series is acquired. A two-position precision indexing mechanism provides 90° in-plane rotation. These features greatly facilitate the acquisition of a dual-axis tilt series.



Dual-axis tomographic reconstructions of CdTe tetrapods. The HAADF tilt series were taken using the Fischione Model 3000 HAADF STEM detector on an FEI Tecnai F20 operated at 200kV. Image (a) is a reconstruction of a single-axis tilt series (-70° to +70°) and shows that some of the legs of the tetrapods are missing or weak due to the missing wedge of information, as indicated by the arrows. Image (b) is a reconstruction of the perpendicular tilt series (-65° to +70°) showing that while the missing legs in (a) are present in this data set, there are a different set of missing legs, again indicated by the arrows. Image (c) is a dual axis reconstruction of the two data sets (Fourier sum) and illustrates that no legs are missing because the missing information has been greatly reduced. The tilt axes in (a) and (b) are parallel to the direction of the arrows.

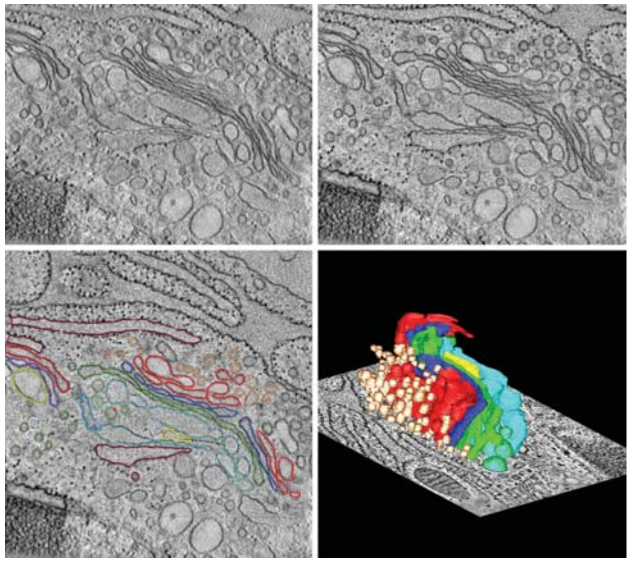
Image courtesy of I. Arslan, J.R. Tong, and P.A. Midgley from the Department of Materials Science and Metallurgy, University of Cambridge, United Kingdom. A.P. Alivisatos from the University of California at Berkeley and Lawrence Berkeley National Laboratory, U.S.A. provided the CdTe tetrapod specimen.



Tomographic slices through the 3D volume of a single (upper row) or dual-axis (lower row) electron tomographic reconstruction of a mitochondrion from chemically fixed cultured Hela cells. Images were acquired on an FEI Tecnai 12 microscope equipped with an FEI Eagle 4K camera. The second and fourth columns represent enlargements of the areas marked in the lower magnification pictures to their immediate left. Arrows point to areas where information is gained in the dual-axis tomographic reconstruction. The small insets in the first column show the reconstructed 10nm gold particles, emphasizing the advantage of performing dual-axis tomography.

Image courtesy of K.M. Valentijn-Estur, K.A. Jansen, J.A. Valentijn, M. Barcena, J. Fernandez-Rodriguez, T. Nilsson, and A.J. Koster from the Department of Molecular Cell Biology, Leiden University Medical Center, The Netherlands.

Model 2040 Dual-Axis Tomography Holder



Tomographic reconstruction of high pressure frozen B-lymphocytes. All images were recorded using an FEI Tecnai 20 Lab6 filament TEM, operated at 200 kV. Datasets were recorded from –60° to +60° in 1° increments with a TVIPS GmbH CCD camera. This image contains a representative image of the single tilt tomogram (upper left), the corresponding image of the double tilt series (upper right), the contour model (lower left), and a 3D model (lower right) of a Golgi region of the B-lymphocyte.

For data acquisition, the FEI Inspect 3D data acquisition software was used. For alignment of the images, and reconstruction of the double tilt tomogram, IMOD image processing, modeling, and display programs from the Department of Molecular, Cellular, and Developmental Biology, University of Colorado, U.S.A. was used. The contouring and modeling were also done with IMOD.

Image courtesy of W.J.C. Geerts from the Department of Molecular Cell Biology, Utrecht University, The Netherlands. Specimen preparation was done by D. Zeuschner from the Department of Cell Biology, University Medical Center Utrecht, The Netherlands

Model 2040 Specifications*

Holder type	Room-temperature. Single-tilt. For most TEMs.
Specimen size	3mm diameter. Wide range of thicknesses up to 100 microns.
Resolution	0.34nm (in all directions)
Drift	<1.5nm/min
Maximum tilt range	up to +/- 70°
Field of view	up to 950 microns at 70°

*All specifications depend on the microscope model, pole piece type, and aperture position. For ultimate resolution and drift performance, the TEM must meet the manufacturer's specifications.

Model 2050 On-Axis Rotation Tomography Holder

Model 2050 On-Axis Rotation Tomography Holder
(shown in stand)

- *Accepts either rod-shaped or conically-shaped specimens.*
- *Ideal for specimens prepared by Focused Ion Beam (FIB).*
- *Ideal for Atom Probe Tomography (APT) and Field Ion Microscopy (FIM) specimens.*
- *Allows 360° image acquisition and tomographic reconstruction without the loss of information due to the missing wedge.*
- *Three-position index allows precise axial rotation of the specimen.*
- *Compatible with all pole piece gap geometries.*

Highly innovative holder for rod or cone specimens

The Model 2050 is a highly innovative TEM tomography holder. It accepts either rod-shaped or conically-shaped specimens and rotates them fully through 360° about the axis of the holder.

The preparation of rod or conical specimens is an extension of conventional TEM methodology which has historically been associated with a 3mm diameter disk specimen. With the advent of electron tomography, combined with advances in specimen preparation technology, it became necessary to develop a specimen geometry and holder to optimize 3D tomographic information from the specimen.

On-axis rotation — no information loss

Traditionally, single axis tomography results in a missing wedge of information and dual-axis tomography results in a missing pyramid of information. On-axis rotation tomography yields results without the loss of any information, thus providing the maximum achievable amount of data from the specimen.

Model 2050 On-Axis Rotation Tomography Holder

The atom probe field ion microscope (APFIM) combines the principles of both time-of-flight mass spectroscopy and point-projection microscopy. The APFIM is able to identify individual elements and to locate them within the bulk material at the atomic level.

The atom probe specimen is a small pointed tip with a radius of approximately 100nm or less. In essence, whereas a typical TEM specimen is thin in one dimension, a typical APFIM specimen is thin in two dimensions.

Once the specimen is inserted into the APFIM, a positive voltage applied to the tip creates a very high electric field that strips electrons from surface atoms and accelerates the resultant ions toward the imaging detector.

Lighter ions reach the detector more quickly than their heavier counterparts. To accurately measure the time of ion departure, the positive voltage is typically pulsed. The arrival time of the ion is recorded on the single particle detector. The time-of-flight of the ion yields elemental identity.

This process of field evaporation provides a 3D image of the evaporated specimen. For optimal atom probe tomography, it is advantageous to correlate results with TEM images of the area of interest within the specimen, which means a suitable specimen holder such as Fischione's is important.

Operation

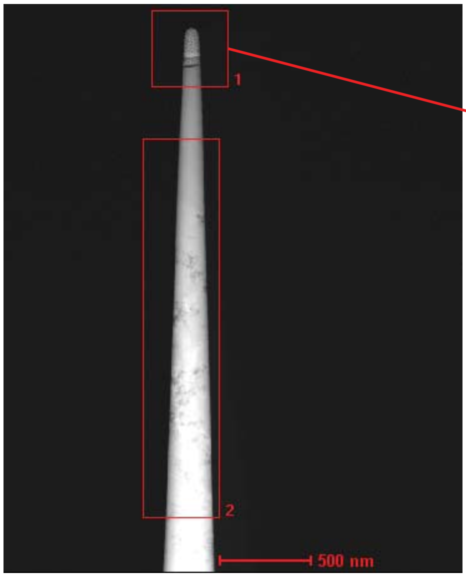
The Model 2050 features a cylindrical specimen cartridge into which a sample post is inserted. Sample posts are available in a diameter of either 1.8mm to accept common APFIM specimen mounts or 1mm to accept FIB-prepared specimens.

In the case of FIB, lift-out techniques are used to transport and attach a thick specimen to the tip of the sample post. Then the specimen is further FIB milled to electron transparency into a rod or cone shape. The sample post containing the specimen is clamped into the specimen cartridge. The specimen cartridge precisely fits within the body of the holder and is accurately aligned with the eucentric plane of the microscope. The cartridge is rigidly affixed to a mechanism which both moves along and rotates about the axis of the holder.

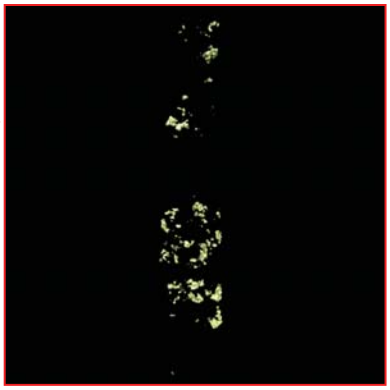
A dedicated loading station facilitates the positioning of the cartridge into the holder. Once the specimen cartridge has been loaded into the holder, longitudinal movement is manually activated in order to retract the loaded cartridge into the body of the holder. This is done to protect the specimen during holder insertion into and removal from the TEM goniometer. Once the Model 2050 is removed from its loading station, the cartridge is mechanically interlocked into the holder so that it cannot become separated while the holder is in the microscope.

Initially, the specimen can be fully rotated through 360° to select the proper specimen orientation. A stop is then engaged which fixes the continuous rotation of the specimen with respect to the holder. A three-position precision indexing mechanism provides the means to orient the specimen in 120° increments. At each increment, while keeping the specimen position fixed with respect to the holder, the microscope's goniometer is tilted to ±60° in order to acquire a tomographic tilt series. Indexing the rotation of the specimen by 120° two more times, combined with the ±60° goniometer tilt, yields data corresponding to 360° of specimen rotation. This procedure maximizes the amount of tomographic data obtained from a single specimen.

Following TEM imaging and analysis, the specimen cartridge can be removed from the holder and securely stored in a storage box which can also be used to safely transport the specimens.



HAADF STEM image of a Ni specimen containing alumina particles. The needle-shape was created using an FEI DB 235 DualBeam Instrument equipped with an Omniprobe nanomanipulator for specimen lift-out.



Reconstruction of alumina particles. An FEI Tecnai F30 TEM was used to collect 19 x-ray spectral images, 500 x 100 pixels by 1024 channels at a rate of 50msec/pixel. The specimen was rotated on-axis in 10° increments from –90° to +90°. Total data acquisition was approximately one million x-ray spectra taken in the course of 4 days. Analysis was conducted simultaneously with the Sandia multivariate statistical analysis software. In the image, Ni is shown as red and the alumina component is shown after image alignment in green.

Images courtesy of P. Kotula, Sandia National Laboratories, U.S.A., L.A. Giannuzzi, FEI Company, U.S.A., and F. de Haas, FEI Company, The Netherlands.

The focused ion beam (FIB) instrument has become an extremely useful tool for TEM specimen preparation. It allows precise positioning of the area of interest within the bulk material. In addition, its sophisticated software control allows the specimen to be extracted from the bulk in the shape of a rod or cone with a substantial area being electron transparent. The Fischione Model 1040 NanoMill can readily provide additional specimen thinning to remove amorphization or Ga implantation.

Model 2050 Specifications*

Holder type	Room-temperature. Single-tilt. For most TEMs.
Specimen size	1.8mm cartridge: Rod or conical with electron transparent tip. Base diameter up to 1mm. Length up to 20mm.
	1.0mm cartridge: A rod or conically-shaped specimen is affixed to the tip of the sample post.
Resolution	0.34nm (in all directions)
Drift	<1.5nm/min
Maximum tilt range	Full 360° rotation, as well as indexing in three 120° increments

*All specifications depend on the microscope model, pole piece type, and aperture position. For ultimate resolution and drift performance, the TEM must meet the manufacturer's specifications.

Touch Protection

Fischione's Advanced Tomography Holders are compatible with the TEM's touch-alarm that stops goniometer movement in the event that a pole touch occurs. Always be aware of the TEM's pole piece configuration and follow the microscope manufacturer's recommendation for operating the goniometer at high tilt angles.

Ordering information

All Fischione Advanced Tomography Holders come with a dedicated loading station for secure specimen handling, tools to assist in specimen clamping, and a Fischione Model 9010 Vacuum Storage Container for storing the Holder in a clean, vacuum environment.

Cover image: Tomographic reconstruction of high pressure frozen B-lymphocytes.
Image courtesy of W.J.C. Geerts, Utrecht University (The Netherlands).

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