

PP3010

CRYO PREPARATION SYSTEM FOR SEM, FE-SEM AND FIB-SEM



Why choose Cryo-SEM?

Cryo preparation techniques for scanning electron microscopy (SEM) are essential for the successful observation of wet or 'beam sensitive' specimens. Cryo-SEM removes the need for specimen-unfriendly conventional preparation techniques, such as critical point drying, allowing observation of specimens in a close-to-life hydrated state.

The limitations of conventional 'wet' processing include:

- Shrinkage, distortion, relocation and extraction of soluble materials
- Mechanical damage of fragile specimens
- Biological material generally requires toxic reagents (fixatives, buffers etc.)

- Long processing times

Advantages of Cryo-SEM

- Specimen viewed in its fully hydrated state
- Soluble materials are retained
- Little or no mechanical damage
- Ideal for time-resolved experiments (i.e. freezing at timed intervals)
- High resolution capability (compared to low-vacuum techniques)
- Extra information obtained by low-temperature fracturing
- Excellent for liquids, semi-liquids, foams and beam sensitive specimens
- Rapid process: typically 5-10 minutes

Recommended applications

- Cryo-SEM or Cryo FIB-SEM samples
- Cryo-FIB lift out and Cryo-FIB lamella preparation
- Biological, materials, foodstuff and polymer applications
- Pre-frozen samples
- Transfer between different imaging systems

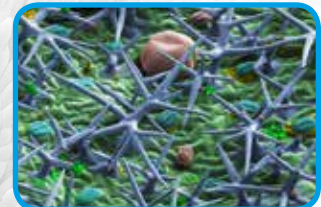
APPLICATION IMAGES



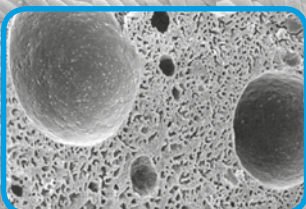
Arabidopsis with virus



Lamella preparation, image courtesy of Jensen Lab CALTECH



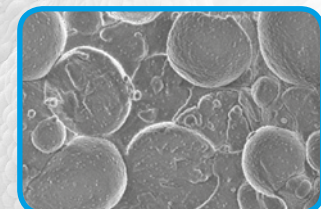
Lavender trichomes with oil droplet and stomata



Ice cream - etched



Predatory mite, image courtesy Gary Bauchan, USDA Beltsville



Oil water emulsion cryo sem, image courtesy of NITTO, Japan

PP3010 Overview

The PP3010 is a highly automated, easy to use, column-mounted, gas-cooled cryo preparation system suitable for most makes and models of SEM, FE-SEM and FIB-SEM.

The PP3010 includes a number of unique but important features, including: off-column cooling (no boiling liquid nitrogen on the microscope) and pumped storage of the cryo transfer device.

The PP3010 has all the facilities needed to rapidly freeze, process and transfer user specimens.

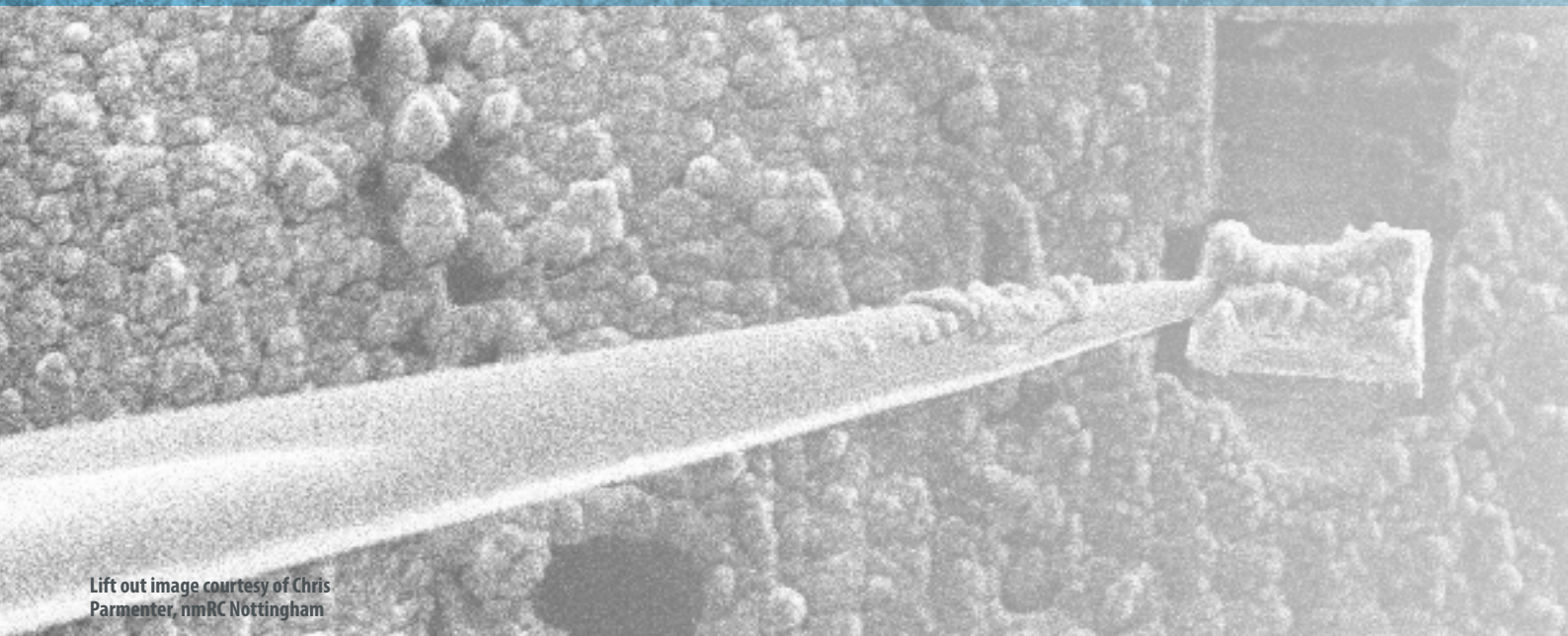
The cryo preparation chamber is turbo-molecular pumped and includes tools for cold fracturing, controlled automatic sublimation and sputter coating.

After processing, the specimen is transferred from the cryo preparation chamber onto a highly stable SEM cold stage for observation.

Cold trapping in the cryo preparation chamber and SEM chamber ensures the whole process is frost-free. Specimen process times are typically between five and ten minutes.

- Recipe driven touch-screen interface
- Fully automated processes and start up
- Gas cooled preparation chamber
- Superb specimen visibility
- Cooling to $-190\text{ }^{\circ}\text{C}$ or better with rapid thermal response
- Off-column cooling and pumping, with minimum mass on the SEM
- Up to 24 hour hold times, with a dedicated off-column 30 litre dewar – no more topping up of on-column dewars
- Single port interface available, if microscope geometry allows
- Beam deceleration compatible up to 5kV
- Full remote control and diagnostics

Lamella lift out using cryo rotate stage



Lift out image courtesy of Chris Parmenter, nmRC Nottingham

PP3010 Features

Specimen holders

The PP3010 comes with a variety of stubs and shuttles designed to accommodate most specimen types. Additional holders are also available, including for high pressure freezing rivets and planchettes.

Handling and transferring specimens

The PP3010 Prepdek[®] workstation is fitted with a combined slushy nitrogen freezing/specimen manipulation system, connected to the rotary pump. Rapid freezing reduces ice crystal damage resulting in enhanced specimen preservation.

The spacious freezing system also allows specimens that have been frozen by alternative methods (or stored field specimens) to be manipulated and mounted onto a suitable holder under liquid nitrogen.

They can then be vacuum transferred into the aQuilo[®] preparation chamber for subsequent processing and observation. The compact cryo transfer device can comfortably be held in

one hand for maximum ease of handling. The sealing mechanism ensures contamination free specimen exchange and the quick-release bayonet connection to the shuttle allows rapid specimen transfer. To ensure the cryo transfer device is maintained in a clean, vacuum compatible condition a pumped storage tube is fitted into the Prepdek[®] work surface. Samples may also be transferred between imaging systems using the new CryoHub.

aQuilo[®] Gas-cooled cryo preparation chamber

The aQuilo[®] cryo preparation chamber is connected directly to the SEM and includes a highly efficient nitrogen gas cooled specimen stage, extensive cold trapping (above and below the specimen) and facilities to fracture, sublimate and sputter coat specimens. Two fully integrated and interlocked gate valves allow transfer into the aQuilo[®] chamber, followed by rapid high-vacuum to high-vacuum specimen exchange to and from the SEM stage.

Efficient gas cooled specimen stage and cold traps

At the heart of the aQuilo[®] chamber is a nitrogen gas cooled specimen stage which can be precisely controlled over a temperature range from 100°C to below -190° . Large gas cooled cold traps, located above and below the specimen stage, ensure clean, high vacuum conditions are maintained in the chamber. Both cold stage and cold traps are fed by the unique CHE3010 off-column cooling system, which typically gives hold times of up to 24 hours between fills.

High visibility

The aQuilo[®] chamber has superb chamber visibility. In addition to the large front window (75 x 150mm) there are two top viewing ports and the chamber is lit by high intensity LEDs.

Prepdek® workstation

The Prepdek® is an ergonomically designed preparation and control centre.

It includes the freezing and pre-frozen specimen manipulation devices, an LED viewing light and the cryo transfer device vacuum storage tube.

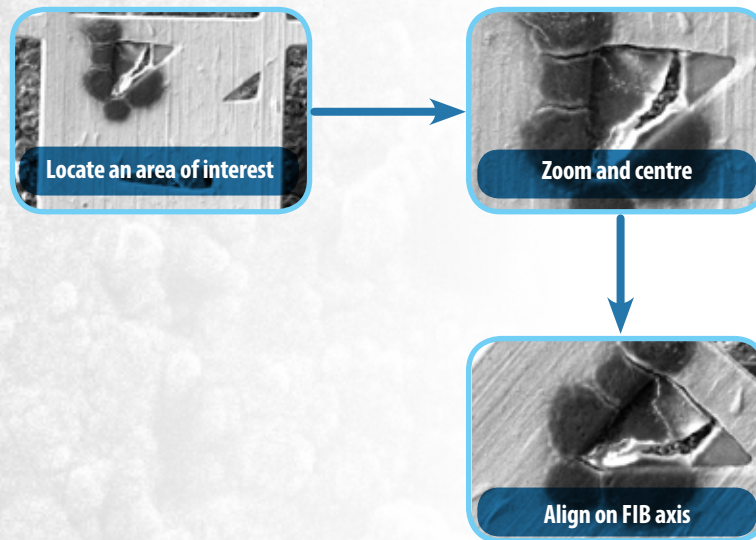
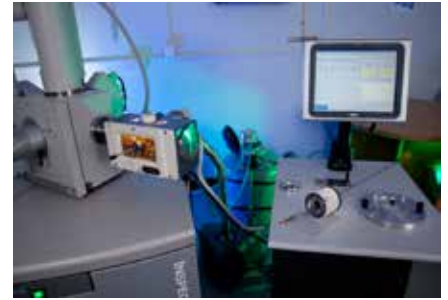
A shuttle mounting jig gives a solid base for specimen mounting.

The control electronics are mounted in a sealed but accessible cabinet beneath the Prepdek®.

The PP3010 is controlled using a large touch screen panel PC, mounted on the Prepdek®.

User-defined 'recipes' can be entered and stored for instant future access. The screen can be set to suit operator preferences; for example, vacuum measurement can be displayed in millibar, Pascal or Torr.

Users can train using the Prepdek® software interface using the demo mode, without any connection to hardware.



Cold fracturing

Actively cooled, twin fracturing tools/manipulators are available and allow a range of specimen types to be cold fractured. Fitted as standard is a front mounted fracturing and manipulation device. The ball-jointed mount offers flexible movement, allowing the blade to be used both as a surface pick (probe) and a fracturing knife. An optional micrometer advanced fracturing tool with rigid blade is available. Fractured fragments are captured in the large cold trap located below the specimen stage.

Automatic sublimation and sputtering

Sublimation temperatures and times can be pre-set and stored for easy retrieval. The process is fully automatic and graphically displayed on the control screen, showing the actual and predicted temperature curves. The high resolution sputter coater will give fine grain films essential for FE-SEM applications. A platinum target is fitted as standard - other metals available include gold, gold/palladium, chromium and iridium. Options

include a carbon fibre evaporation accessory and a terminating film thickness monitor (FTM).

Cryo preparation chamber pumping

The aQuilo® chamber is evacuated by a remotely-positioned 70L/s turbo-molecular pumping system, backed by a suitable rotary pump. Typical vacuum levels during operation are in the region of 10^{-6} mbar or better. Positioning the turbo-molecular pump away from the SEM ensures total elimination of mechanical vibration and significantly reduces the mass connected to the SEM. The vacuum buffer tank backs the turbo-molecular pump and is automatically evacuated by the rotary pump when required – typically for only a few minutes in each hour. A dry pump is available as an option.

SEM cold stage and cold trap

A highly stable, thermally isolated, nitrogen gas cooled stage attaches to the SEM stage using an adaptor. The SEM stage and cold trap are cooled by two separate cold gas circuits - both have rapid response and are capable of reaching temperatures down to -190°C or lower.

Cryo Rotate Stage

The stage offers users 360 degree, bi-directional rotation and specimen temperatures can typically be managed below -165°C . When milling lamella, rotating the sample through 180 degrees allows both sides of the lamella to be imaged. Users can also align sample features prior to a cryo lift-out of the lamella.

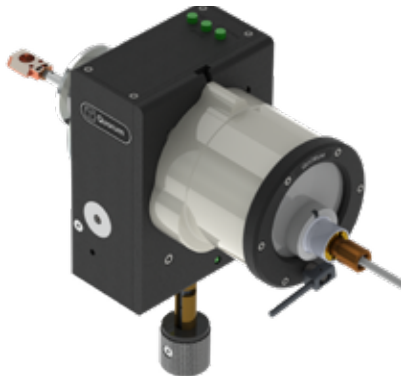
CHE3010 off-column, vacuum isolated gas cooling system

The CHE3010 is a fully integrated, remotely positioned cooling system. The cold nitrogen gas generated is used to cool the entire PP3010T – i.e. SEM stage, SEM cold trap, aQuilo chamber cold stage and cold traps. The CHE3010 delivers temperatures down to -190°C or lower.

A key feature is that the cold gas is carried to the microscope UVA vacuum isolated lines, giving not only superb thermal efficiency but also the option of flexible site location (typically on the floor behind the microscope).

Materials Transfer Systems

P3004 QuickLok - ambient temperature airlock for SEM, FIB-SEM, beam-line and vacuum products.



P3006 CoolLok - non-airlock cooling for SEM, FIB-SEM beam-line and vacuum platforms.



Cryo Cooling System

P3005 SEMCool - cryo transfer system for SEM, FIB-SEM, beam-line and vacuum platforms.



PP3010 Accessories



Cryo Rotate Stage



The new stage offers users 360 degree, bi-directional rotation and specimen temperatures can typically be managed below -165°C.



Glovebox Interface



Specimen stubs & shuttles

Specifications

SEM components:

Nitrogen gas cooled SEM cold stage: -190°C or lower. Temperature stability: <0.5°C
Nitrogen gas cooled SEM cold trap: -190°C or lower
SEM LED illumination

SEM cooling:

CHE3010 off-column, 21 litre gas cooling system, typically with up to 24 hours hold time

Column mounted aQuilo cryo preparation chamber with:

Nitrogen gas cooled cold stage: -190°C, or lower to +100°C.
Temperature stability: < 0.5° / hr
Nitrogen gas cooled upper and lower cold traps ambient to -190°C or lower
Multiple LED illumination and CCD camera (Optional Binoculars Viewer)
Actively cooled fracturing/manipulation tool, micrometer controlled knife (option)
Large front window (150mm x 78mm) plus two top viewing ports
Automated, high resolution sputtering (Pt standard)
Optional film thickness monitor and carbon fibre evaporation attachment
Automated sublimation

Preparation chamber pumping system:

Floor-mounted turbo pumping with stainless steel vacuum connection to the preparation chamber
Base vacuum: 10⁻⁶ mbar or better. Single 5m³/hr⁻¹ or equivalent rotary vacuum pump required
Optional dry pump

Touch-screen control via panel PC:

380mm/15" panel PC
User definable "recipes" can be stored
On-screen data logging, diagnostics and help videos

Prepdek® specimen preparation station:

Slushed nitrogen freezing and specimen handling system – ideal for handling pre-frozen specimens
Includes work area for specimen preparation
Pump storage for cryo transfer device, flexible LED light and specimen mounting jig