HTX SubliMATE™

MALDI Matrix Nano-Coating System

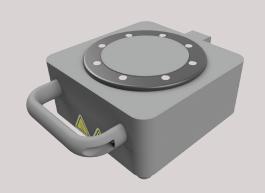




HTX SubliMATETM MALDI Matrix Nano-Coating System

The HTX SubliMATE™ is an easy-to-use

MALDI matrix deposition system offering speed,
throughput, reproducibility, and the smallest
available matrix crystals for sub-micron resolution
mass spectrometry imaging.



The HTX SubliMATE $^{\text{TM}}$ is a robust and easy-to-use sublimation system offering a standardized and controlled method for sample preparation for matrix assisted laser desorption/ionization (MALDI) mass spectrometry imaging. The sublimation chamber is designed for speed, multiplexed sample preparation, and reduced matrix consumption.

A fully integrated ceramic heater is used to quickly heat MALDI matrix to sublimation temperatures up to 300° C. Slides are cooled with ice or dry ice bath for temperatures ranging from 15 to -78 °C to produce ultra-small matrix crystals. The high capacity slide holder (4 standard microscope slides, 25 x 75 mm each) allows for high throughput sample preparation.

MALDI matrix solution is pipetted onto a stainless steel Matrix Wafer before each sublimation cycle (Figure 1). The HTX Matrix Wafer (patent-pending) ensures homogeneous distribution of matrix crystals across a large area of up to 4 glass slides.



Figure 1. HTX Matrix Wafer

Key Characteristics

- Proprietary sublimation chamber producing ultra-small matrix crystals (<1 micron)
- 4 slide capacity for increased throughput
- Fast heating and cooling for quick sample preparation
- Highly consistent matrix deposition across entire sample area (+/- 4% by fluorescence gray values values and slide weight)
- Proprietary Matrix Wafer template design for even distribution of matrix prior to sublimation
- Validated protocols for most MALDI matrices (ex: CHCA,DAN, DHAP, DHB, NEDC)
- Continuous matrix coverage as needed for high spatial resolution MALDI imaging

Benefits of Sublimation

- 1. Spatial Resolution: Sublimation provides minimum crystal size in the nanometer range
- 2. Minimum Delocalization: By skipping the liquid phase in the matrix deposition process, more spatial information is preserved in samples
- 3. Efficient: The matrix layer can be applied in one single process

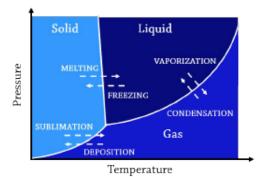


Figure 2. Phase State Diagram. The sublimation process occurs when solid matrix undergoes a direct phase transition to a gas and then deposits in a homogeneous layer onto the sample mounted at the cool top of chamber.

Case Study #1: Imaging of Lipids in the Human Eye

Experimental Summary

Tissue Type Human Eye
Preservation Fresh frozen

Tissue Cut $10\text{-}12~\mu\text{m thickness}$

Time to Ready Vacuum 120 seconds
Sublimation Time 10 minutes
Heating Temperature 130°C
Cooling Temperature -78°C

Instrumentation and Supplies

MALDI Plate ITO coated glass slides

Matrix DAN or DHAP

Matrix Deposition Device HTX SubliMATE™

MALDI MS tims TOF fleX MS

(Bruker Daltonics)

Laser Raster $5 \mu m$

Imaging Software SCiLS Lab (Bruker Daltonics)

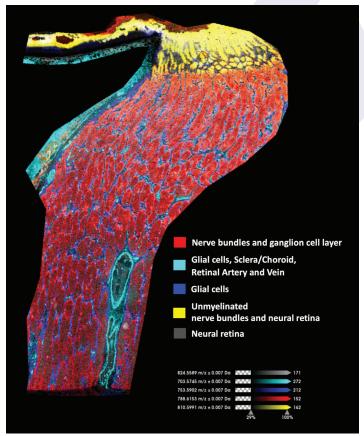


Figure 3. MALDI IMS data of optic nerve and surrounding tissue. Imaging was performed in positive ion mode at $5\mu m$ with sublimated matrix DHAP.

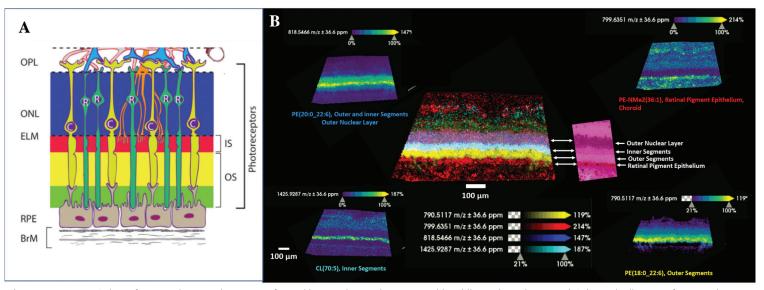


Figure 4. MALDI IMS data of outer retina. Imaging was performed in negative mode at 5 µm with sublimated matrix DAN. A) Schematic diagram of outer retina with layers OPL outer plexiform layer, ONL outer nuclear layer, ELM external limiting membrane, RPE retinal pigment epithelium, C cone, R rods, IS inner segments, OS outer segments. Figure adapted from Anderson et al. 2020 JASMS. B) MALDI IMS data of photoreceptors in peripheral retina. Specific lipid signals correlated with discrete layers include PE(20:0_22:6), OS and IS and ONL; PE- NMe2(36:1), RPE; CL(70:5), IS; PE(18:0_22:6), OS and IS.

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5µm Imaging of Lipids in Mouse Cerebellum

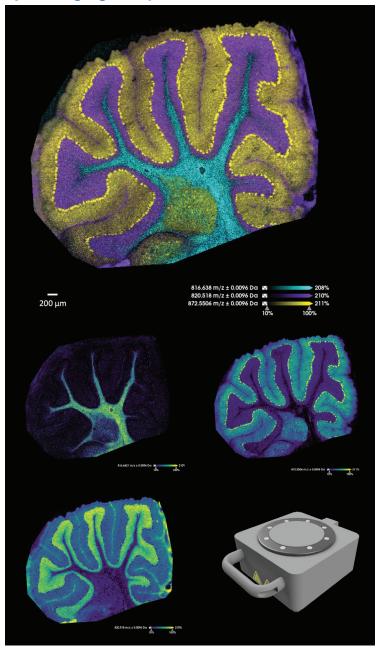


Figure 5. Ultra-high spatial resolution lipid imaging of mouse brain. Frozen brain section was coated with DHAP using the HTX SubliMATE $^{\text{TM}}$. Sublimation was performed at 200°C for 5 minutes. Slide was imaged in positive mode at 5 μ m.

HTX SubliMATE™ Specifications	
Sublimation Chamber	
Dimensions	9.5"x13"x4" (24 x 33 x 10 cm)
Weight	17.5 lbs (8 kg)
Cooling	Ice or Dry Ice
Heating	Ceramic, Max 300°C
Vacuum Connector	NW10
Vacuum Pump Capable of 0.02 mBar Recommended: Edwards E2M28	
Dimensions	23 x 6.8 x 11"(58 x 17 x 28 cm)
Weight	97 lbs. (44 kg)
Inlet Flange	NW25
Vacuum Level for Sublimation	< 40 mTorr

Reproducibility

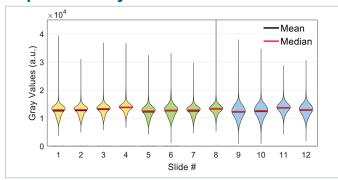


Figure 6. Reproducibility of DHAP matrix coating by HTX SubliMATE™. DHAP-coated slides (n=12) were prepared by 3 different users (yellow, green, blue) via addition of 30 mg DHAP to Matrix Wafer. Sublimation was performed at 130°C for 10 minutes. Autofluorescence gray values were measured from 1 cm² sections on each side.

Tissue images and reproducibility data are courtesy of Drs. David Anderson, Eric Spivey, and Richard Caprioli; Mass Spectrometry Research Center, Department of Chemistry, Vanderbilt University School of Medicine, Nashville, TN, USA.

The HTX SubliMATE $^{\text{TM}}$ is available worldwide exclusively from HTX Technologies, LLC and authorized resellers.

To request further information contact:

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HTX Technologies offers innovative sample preparation systems for advanced analytical platforms. Our integrated workflow solutions include user training, instruments, software, consumables and method development services.



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