

INSTRUMENT DEMONSTRATION SUMMARY

Topic: TOF & TOF/TOF – lipid, peptide, and protein imaging

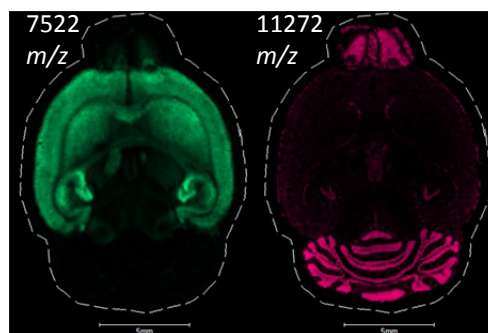
Time: Wednesday 3:00 pm

Background

Matrix-assisted laser desorption/ionization time-of-flight (MALDI TOF) mass spectrometers are the instruments of choice for many imaging experiments. Due to their sensitivity, speed, and high mass range, TOF mass analyzers are advantageous for a wide variety of analytes including small molecules, lipids, peptides, and proteins. While a linear geometry provides the sensitivity required for the analysis of higher molecular weight species, the higher resolving power afforded by the use of a reflector is preferred for the analysis of lower molecular weight species. This workshop will demonstrate the acquisition of protein and peptide images using MALDI TOF mass spectrometers as well as the specificity afforded for MS/MS lipid imaging by MALDI TOF/TOF mass spectrometers (see the MALDI LTQ XL – small molecule imaging demonstration for more information on MS/MS imaging).

Highlights

- Calibration and optimization of an acquisition method
- Key instrumental parameters for sensitivity & resolution
- Alignment of plate to optical image using fiducials
- Setup of an automated method for data collection
- Setup of an image acquisition of brain (lipid and protein) and kidney (peptide) tissue sections
- Visualization of acquired image data
- Demonstration of high-speed TOF/TOF imaging
- Discussion of instrument cleaning and maintenance



Summary

Peptide & Protein Imaging: Peptide and protein images will be acquired from separate 12 μm sections of fresh frozen tissue (peptide: rat kidney; protein: mouse brain) that have been coated with matrix (peptide: via a robotic spraying protocol; protein: via a sublimation/rehydration protocol). Data will be collected in positive ion mode on a Bruker Autoflex Speed mass spectrometer. The images will be collected at 100 μm spatial resolution (protein: linear mode over the m/z range 3,000-25,000; peptide: reflector mode over the m/z range 800-5,000) and visualized using FlexImaging.

High-Speed TOF/TOF Imaging: High-speed TOF/TOF lipid images will be acquired from a 10 μm section of fresh frozen mouse brain that has been coated with matrix via a sublimation protocol. High-speed imaging is facilitated by a high laser repetition rate (5 kHz) and continuous raster sampling. TOF/TOF imaging is facilitated through the use of a precision timed ion selector (TIS), a collision cell, and a reacceleration region. Ion images will be collected in TOF/TOF mode on a SimulTOF 300 tandem mass spectrometer at 100 μm spatial resolution and then visualized using the SimulTOF Viewer.