

Monday Poster Session: MALDI: Sample Preparation Code: MPN Time Slot/Poster Number: 260

Coupling Thin Layer Chromatography with MALDI-FTMS.

Vera Ivleva¹; Isamu Matsunaga²; Eric A. Berg¹; D. Branch Moody²; Peter B. O'Connor¹; Catherine E. Costello^{*1}

¹Boston University School of Medicine, Boston, Massachusetts; ²Harvard Medical School, Boston, Massachusetts

Novel Aspect: Direct coupling of TLC with newly designed high throughput, high pressure MALDI-FTMS ion source for the simple and accurate analysis of glycolipids from biological sources. **Introduction:**

The newly designed high throughput, high pressure MALDI-FTMS ion source allows direct coupling of thin layer chromatography to MALDI-FTMS. This system allows TLC plates to be taped directly onto the MALDI target, inserted into the high pressure MALDI source, and ions can be desorbed, cooled, trapped, and detected in the FTMS. Beyond the usual resolution, mass accuracy, and sensitivity advantages, the FTMS performance is not limited by the irregular TLC surface as is the case with the TOF. Furthermore, the use of high pressure MALDI is particularly advantageous for labile glycoconjugates which are easily separated by TLC, such as whole brain gangliosides, CD1-presented glycolipid antigens, and glycolipid anchors.

Methods:

Thin layer chromatography plates are used to separate glycolipids. These plates are then attached directly to the MALDI target, and a saturated solution of matrix in ethanol is sprayed on top. The plate is allowed to dry and then inserted into the MALDI source on the FTMS. The FTMS are operated in external trap mode, in which the source hexapoles is used to accumulate the ions over 1-1000 laser shots. The ions are then transferred to the cell (accumulated trapping with a gas pulse at 10V, ramped to 1V for detection) for excitation (430-3500 Da bandwidth, 4 msec, 75 Vpp frequency chirp) and detection (500 kcps, 12 bit samples, direct mode detection).

Preliminary Data:

Leishmania membrane protein anchor glycolipids were separated by TLC followed by MALDI-FTMS analysis, and the resolution of 1:35,000 was obtained. Gangliosides have been desorbed from TLC plates into the MALDI-FTMS and these spectra will be presented. A set of 25 matrices have been tested with the ganglioside desorption in the high pressure MALDI-FTMS, and a comparison of the resolution fragmentation pattern will be presented. Several good high pressure MALDI matrices have been identified and the spectra using these matrices will be presented.

Data will also be presented to show that separation can be tracked and analyzed with the TLC-high pressure MALDI-FTMS. In two other projects, ganglioside rich lipid rafts from neuronal cells and the lipids from the pathogenic mycobacterial species have been isolated. These samples will be tested with the TLC-high pressure MALDI-FTMS

and the results will be presented.

Acknowledgement: This project has been funded in whole or in part with Federal funds from the National Institute of Health, under grant No. P41RR10888-6 from the National Center for Research Resources and from the National Heart, Lung, and Blood Institute, under Contract No.N01-HV-28178.