LESA FAIMS mass spectrometry for the spatial profiling of proteins from tissue

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Overview

Here, we demonstrate LESA FAIMS mass spectrometry imaging of proteins in thin tissue sections from mouse brain and liver, and lamb brain.

Introduction

- We have shown previously that coupling of high field asymmetric waveform ion mobility spectrometry (FAIMS) with liquid extraction surface analysis (LESA) mass spectrometry of tissue results in significant improvements in the resulting protein mass spectra.
- Here, we demonstrate LESA FAIMS mass spectrometry imaging of proteins in thin tissue sections from mouse and lamb brain. The results are compared with LESA images obtained in the absence of FAIMS.
- The results show that the number of different protein species detected can be significantly increased by incorporating FAIMS into the workflow.
- The spatial distributions of proteins identified in both LESA mass spectrometry imaging and LESA FAIMS mass spectrometry imaging were in good agreement indicating that FAIMS is a suitable tool for spatial profiling.

Results: LESA & LESA FAIMS MSI of mouse liver

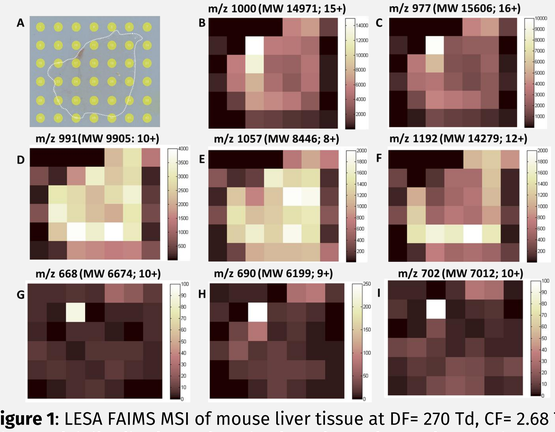


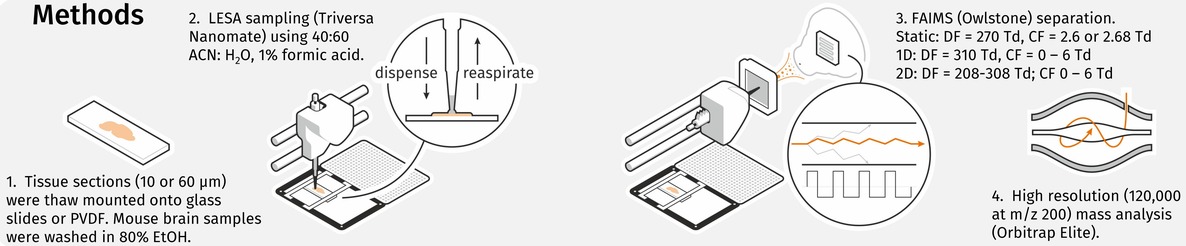
Figure 1: LESA FAIMS MSI of mouse liver tissue at DF= 270 Td, CF= 2.68 Td. The spatial distributions of a selection of protein species are shown . B) and C) correspond to α- and β-globin; F) corresponds to FABP1.

➢ A total of 40 separate protein species were detected in the mass range 4 to 16 kDa across the tissue section profiled by LESA (static) FAIMS, 29 of which were not detected in the LESA analysis. Twenty four proteins (13 unique) were detected by LESA mass spectrometry imaging .

Conclusions

➢ FAIMS is a suitable tool for inclusion in MSI workflows.
➢ LESA FAIMS results in detection of greater numbers of proteins than LESA alone.
➢ NMF may be applied to the interrogation of 2D FAIMS data and thus inform choice of static FAIMS conditions.

Methods



Results: LESA & LESA FAIMS MSI of mouse brain

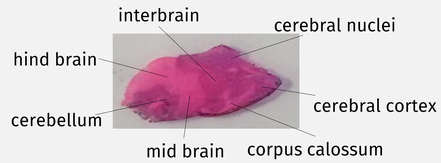


Figure 2: H&E stained sagittal section of mouse brain

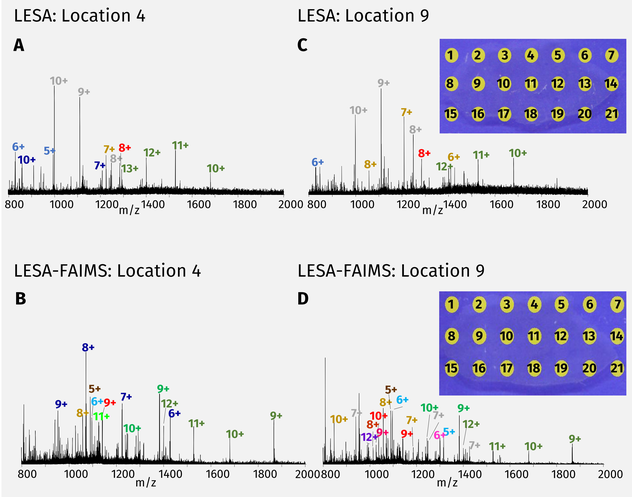


Figure 3: LESA and LESA-FAIMS profiling:. Mass spectra acquired at A) location 4 following LESA profiling; B) location 4 following LESA-FAIMS profiling; C) location 9 following LESA profiling; D) location 9 following LESA-FAIMS profiling. Colours indicate multiple charge states of individual proteins.

➢ A total of 34 separate protein species were detected in the mass range 4 to 17 kDa across the tissue section profiled by LESA (static) FAIMS, 26 of which were not detected in the LESA analysis. Fifteen proteins (7 unique) were detected by LESA mass spectrometry imaging.

Acknowledgments

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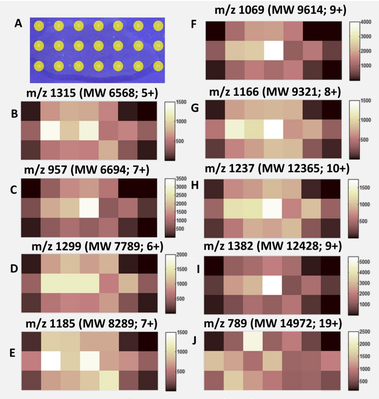

Figure 4: LESA FAIMS MSI of mouse brain tissue at DF= 270 Td, CF= 2.6 Td. The spatial distributions of a selection of the species unique to the LESA (static) FAIMS experiment are shown.

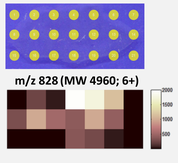

Figure 5: LESA MSI of mouse brain tissue. The spatial distribution of β-thymosin 4 is shown.

➢ β-thymosin 4 was detected in most on-tissue locations in the LESA experiment, but in only two in the LESA FAIMS experiment. A LESA 1D FAIMS MSI experiment was performed with DF = 270 Td and CF -1 to 4 Td. β-thymosin 4 was detected at each location; however the optimum CF for its transmission was 3.6 Td.

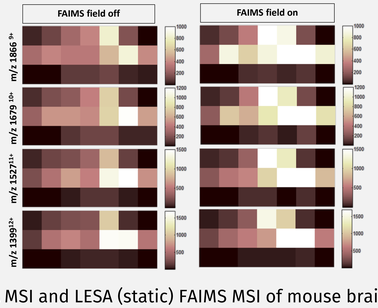

Figure 6: LESA MSI and LESA (static) FAIMS MSI of mouse brain acquired following single extractions from the same tissue section. The spatial distributions of the 9+ to 12+ charge states of calmodulin (MW 16780 Da) are shown (left) in the absence of FAIMS field and (right) in static FAIMS field (DF= 270 Td, CF= 2.6 Td).

Results: LESA & LESA FAIMS MSI of lamb brain

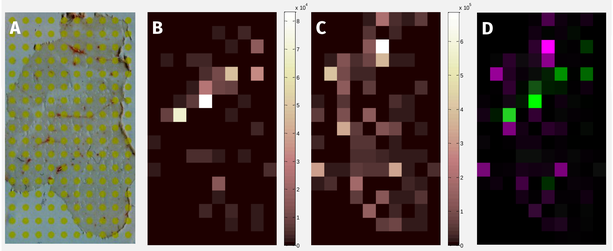

Figure 7: LESA 1D FAIMS MSI of lamb brain tissue at DF= 310 Td, CF= 0-6 Td. A) Photograph of section; B) Ion image of m/z 602 (3+) observed in white matter only; C) Ion image of m/z 1208 (8+) observed in grey matter only; D) Overlay of m/z 602 (green) and m/z 1208 (purple).

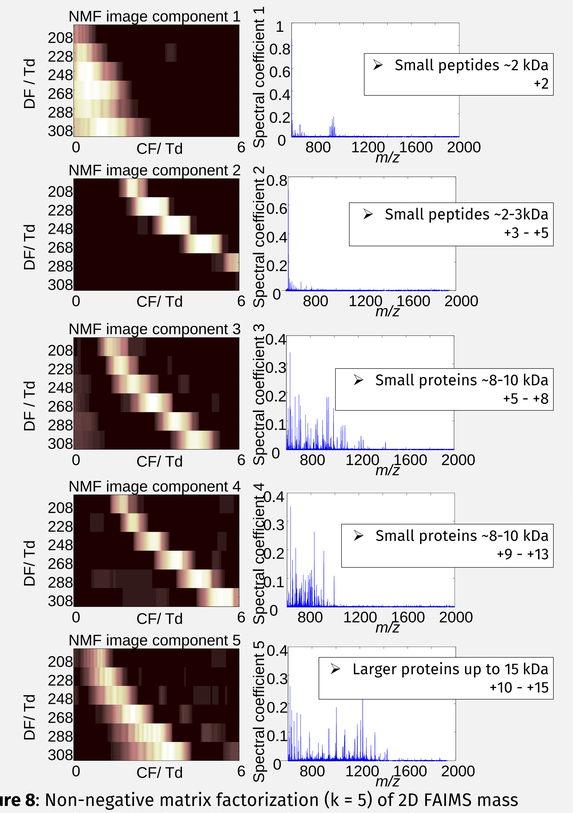

Figure 8: Non-negative matrix factorization (k = 5) of 2D FAIMS mass spectra obtained following LESA sampling of lamb brain.

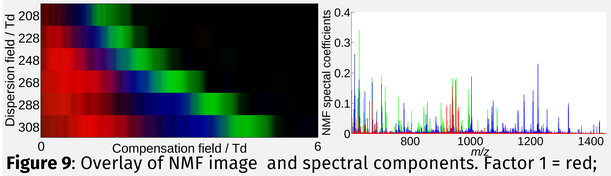

Figure 9: Overlay of NMF image and spectral components. Factor 1 = red; Factor 3 = green; Factor 5 = blue.