

Imaging at Atmospheric Pressure by MALDI Mass Spectrometry

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Overview

- An atmospheric pressure (AP) MALDI interface was developed and utilized to map the spatial distribution of peptides and small organic molecules with UV or IR laser excitation
- Spatial resolution of 40 μm was achieved for the IR laser with oversampling
- AP-MALDI imaging of a strawberry skin was demonstrated using native water as the matrix

Introduction

- Matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS)
 - ◆ Powerful tool for the investigation of biomedical and biochemical problems
- MALDI-MS imaging
 - ◆ Rapid and sensitive molecular imaging in vacuum
 - ◆ Information on the spatial distributions and temporal variations of chemical species in biological systems \Rightarrow essential to the understanding of biological processes
- Conventional UV MALDI-MS imaging technique requires
 - ◆ Vacuum environment
 - ◆ Problems: complex sample handling, excludes the use of liquid samples
 - ◆ Possible solution: AP-MALDI
- Sample is coated with matrix
 - ◆ Problem: potentially obscures or distorts the studied component distributions
 - ◆ Possible solution: using native matrix (such as water)
- Advantages of IR-MALDI compared to UV-MALDI
 - ◆ Softer ionization and less analyte ion fragmentation

Potential matrix

◆ Water

Native environment for most biological molecules
Successfully used as a matrix to generate peptide ions^[1]
In vivo analysis of biochemical processes under natural conditions
On-line coupling of MS with liquid separation techniques such as HPLC, CE or electrophoresis

Methods

- MS: Q-TOF Premier mass spectrometer (Waters Co.) with a custom-made AP interface (130 μm ID capillary inlet)
- Laser: Nitrogen laser: 337 nm, 10 Hz, 5 ns, 400 μJ
OPO laser: 2940 nm, 10 Hz, 4 ns, 150 μJ
- Pulsed dynamic focusing (PDF)^[2]: improves sensitivity by a factor of five
- 3-axis precision flexure stage (NanoMax TS, Thorlabs Inc.)
- Ion chromatogram: ion Intensity \sim scanning time (\Rightarrow x-y coordinates)

Results

1. UV-AP-MALDI mass spectrometry and imaging

- ◆ Identification of the origin of unknown fragment ions
- ◆ Analysis of a complex peptide mixtures

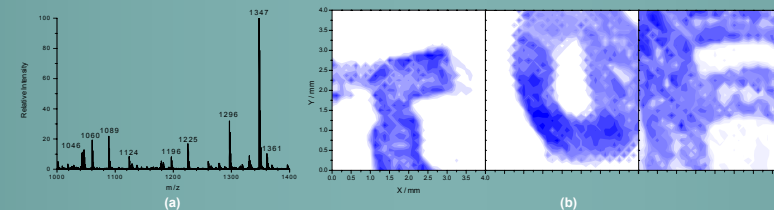


Figure 1: (a) UV-AP-MALDI mass spectrum of a bradykinin, angiotensin I and substance P mixture; (b) AP-MALDI image of three letters, "T", "O" and "F".

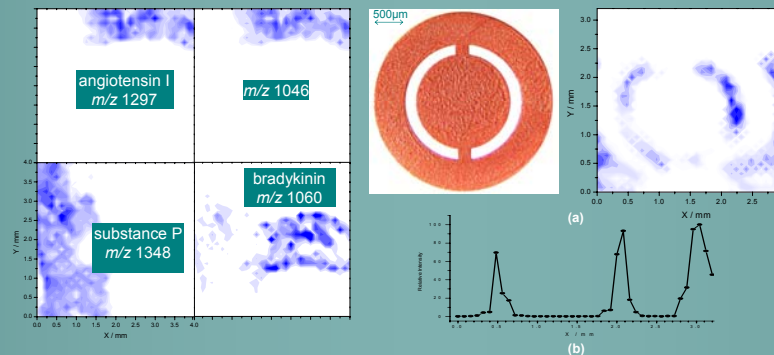


Figure 2: Letter "F" imaged by UV-AP-MALDI. Three strokes in the letter were produced by substance P, angiotensin I and bradykinin solutions. Ion with m/z 1046 is co-located with angiotensin I, thus indicates a contamination or a degradation product.

Figure 3: (a) Optical and AP-MALDI MS images of an electron microscope grid (outer diameter 3050 μm and gap width 150 μm); (b) averaged ion intensity profile along a line through the grid center.

2. IR-AP-MALDI mass spectrometry and imaging

- ◆ Oversampling^[3]: amenable for samples completely ablating from the surface
 \Rightarrow Stepsize of translation stage movement determines spatial resolution
- ◆ Spatial resolution: 40 μm , 5 times smaller than laser spot size
- ◆ Successfully mapped the spatial distribution of peptides
- ◆ Native water of strawberry serves as an efficient matrix
- ◆ Mass spectrum of strawberry: potassium fructose/glucose and sucrose ions

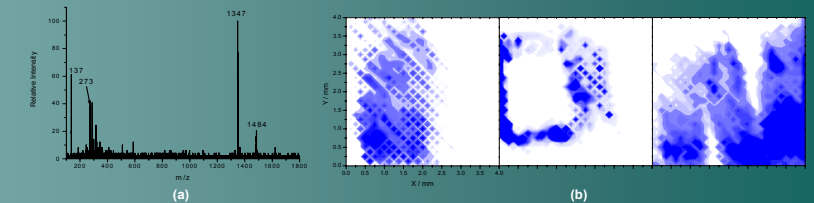


Figure 4: (a) 10 laser shots mass spectrum of the substance P solution with DHB as a matrix; (b) IR-AP-MALDI image of three letters, "I", "O" and "N".

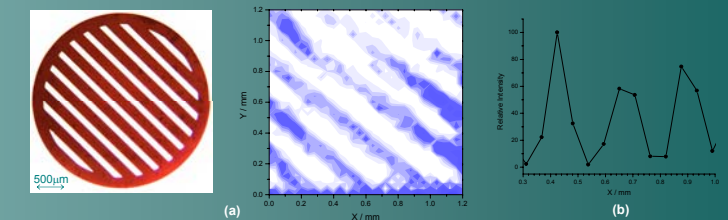


Figure 5: (a) Optical image of an electron microscope grid (outer diameter 3050 μm , hole width 92 μm and bar width 184 μm) and the IR-AP-MALDI image of the grid obtained using the oversampling method; (b) averaged ion intensity profile along the direction normal to the grid bars.

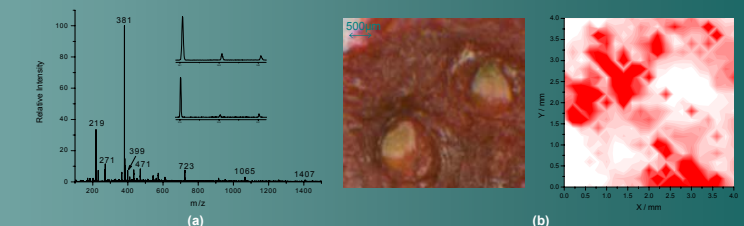


Figure 6: (a) IR-AP-MALDI mass spectrum and (b) optical and false color molecular images of strawberry skin.

Conclusions

- AP-MALDI interface was developed and coupled with UV or IR laser excitation
- "Oversampling" for IR imaging improved spatial resolution
- Native water in a strawberry served as an efficient matrix for IR-MALDI
- IR-AP-MALDI imaging technique
 - ◆ Offers a new approach for *in vivo* studies of complex biological systems in their natural environment
- ◆ A promising tool for biological and medical research

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References

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