A comparative performance characteristics of AP and low-pressure (SubAP) MALDI sources

Overview
A comparative study was conducted on the analytical performance of MALDI source operating at either low pressure (0.6-20 Torr) or atmospheric pressure. In both cases, the source was attached to LTQ mass spectrometer equipped with the ion funnel assembly.

- The detection limits achieved in the peptide MALDI MS analysis were significantly lower under subatmospheric than atmospheric conditions (e.g., 0.05 ppm vs. 0.1-1 Torr pressure conditions).
- Working at 1-Torr pressure one observes a decrease in the signal intensity at lower fluences compared to that observed in the AP MALDI source.
- MALDI sources for Arg-rich peptides were barely detectable at 1 Torr. With the increase in the pressure, the adduct peaks start to rapidly grow.

A substantial sensitivity improvement can be explained by the fact that the ion composition and cooling rate of the matrix clusters ablated from the MALDI sample at 1 Torr are very different compared to atmospheric conditions. It is hypothesized that at atmospheric pressure, very high cooling rate of ablated cluster impacts the ion temperature. This trend was confirmed by the fact that the ion funnel to initially higher temperature using higher laser fluences compared to those used in low-pressure or vacuum MALDI sources.

Method
The MassTech AP MALDI source was attached to the chamber comprising the ion funnel with the orthogonal capillary inlet. The face of 0.75 μl (2.0 mm.o.d.) heated capillary (250°C) was placed 1.9 mm from the MALDI plate. The ion funnel assembly was mounted onto LTQ (ThermoFisher) replacing the standalone skimmer-needle assembly.

The low-pressure (SubAP) MALDI source design followed that of the MassTech AP source, yet a chamber accommodating the sample plate was made an tight-fitted evacuated with a separate forepump system. The target plate in SubAP MALDI source was mounted 10 mm from the ion funnel entrance. Up to 50 volt was applied across the gap between the target plate and funnel entrance.

The described ion sources were transported into the LTQ. The laser spot diameter was the same in both the AP and AP sources (≈1 μm, laser intensity dependent). AP MALDI and SubAP MALDI matrix samples were prepared using standard dried-droplet method.

RESULTS

Ion yield vs laser energy: Intensities of three major peptide peaks were summed up and plotted against selected level of laser energy, LTQ operated in the Normal Scan mode (low mass resolution).

At atmospheric conditions, the ablation dynamics of matrix material is greatly different from that at 1-Torr pressure: cluster size is generally larger (Ref.1) and the cooling rate of matrix clusters is much higher in a model where the analyte ions escape from the surface of hot matrix clusters, the ion yield is to a steep (Armenian) function of cluster temperature Tc. So given the limited time for the ions to escape the cluster (before the cluster becomes too cold), the initial temperature Tc of the cluster should be much higher compared to that at 1 Torr due to significantly higher cooling rate r.

It was noticed in many studies that a mild thermal activation of peptide molecular ions in a presence of chemical noise background increased the decomposition rate of ions of chemical noise to a larger extent compared to decomposition of peptides. Assuming that chemical noise comprises various species of the non-organic origin, higher internal temperature in the laser pulse brings to the more fragments of those background species.

AP MALDI ZoomScan mode

Ion yield vs electric field near the MALDI plate

SubAP MALDI

Peptide: Bradykinin H1-7 300 fmol
Ang II 100 fmol
Sub P 50 fmol
Phe/Val 100 fmol
CHCA: 0.5 mg/mL
Dried-droplet spot 1.8 mm in diameter
Laser: 11 μl in the laser spot of ~250 μm (the threshold energy was 4.7 J for AP MALDI source)

1-Torr MALDI

ChCA

Laser: 7 μl in the spot of ~250 μm (the threshold energy was 4.7 J for AP MALDI source)

Relative Intensity of Sub P (x2=5x) peak increased by a factor of 2.5; matrix region signals (100 - 500 m/z) dropped ~50 fold at 1 Torr

MALDI threshold

Activation of peptide ion on the electric field is associated with the gas pressure:

- 5.4 kV/19 cm - 3171/FPM
- Sub AP 45 Torr / 750 J/cm

In AP MALDI source, light ions are loss when they hit the capillary edge because the ions fail to follow the electric field lines. On contrary, larger ions are ion-optically focused by the field boosting those ions into the capillary inlet.

In the approach developed in the original paper, ion neutralization rate K does not depend on metastable velocity v* of the ions. When such a dependence is introduced, the model starts to fit a non-linear charge of ion survival rate at (v) at small accelerating field E.

The use of enhanced laser beams in MALDI can be done by a process of the charge neutralization/field-driven charge separation in a gas phase.

Conclusion
- SubAP MALDI source that utilizes the ion-funnel technology offers a 6-20 fold sensitivity increase (field dependent) in MALDI MS analysis of a wide mass range.

The gas pressure in the SubAP MALDI source can be easily changed from ~0.8 Torr to 1.0 Torr to study MALDI-PSD at different conditions. The laser can be connected to other source of gas (argon, nitrogen, etc.)

The dependence of ion yield on the applied electric field can be described by a process of the charge neutralization/field-driven charge separation in a gas phase.

References