

Modified target surface to enhance performance during MALDI analysis

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User Benefits

- ◆ Higher number of samples on the target increases the throughput.
- ◆ Higher sensitivity.
- ◆ Less sample (volume) needed.

Introduction

Miniaturization, parallelization, and automation are crucial for modern approaches in drug discovery and biotechnology in order to maximize the throughput, efficiency, and reducing the massive expenses.

Aquarray developed a Droplet Microarray (DMA) technology for chemical synthesis, characterization and biological screening (1-2). The surface of a target slide / plate is chemically modified and omniphilic-omniphobic patterned resulting in a high contrast in wettability between omniphilic spots and surrounding omniphobic areas.

Thus, aqueous and organic liquids can be applied manually by a pipette or automated using a liquid dispenser on the patterned surface while generating high-density arrays of nanodroplets. Each nanodroplet represents a separated vessel that can be used for different chemical, analytical or biological experiments.

One of the critical steps in the workflow of combining chemistry with biology is the analytical investigation, since most analytical techniques are not designed for a highly miniaturized setup. Shimadzu MALDI mass spectrometers enable fast and sensitive characterization of chemical and biological samples. Here we report a high-throughput workflow using the Aquarray droplet microarray technology for fast sample preparation and high-sensitivity MALDI MS characterization down to the attomole range per spot.

Sample preparation

The surface of Shimadzu stainless steel FleximassTM-SR0 targets were modified by Aquarray with a pattern of omniphilic spots and omniphobic surroundings. The spot size of 900 μm

distance between the spots of 225 μm as well as the circular shape of the spots can be customized depending on the application. Fig. 1 a) shows the behavior of water spread all over the surface of the modified target. Water is pushed away from the omniphobic surroundings and small droplets are formed on the omniphilic sample positions.

Using this pattern, the number of samples on the target could be increased by a factor of 14 from 48 (+3 for calibration) to 705.

Sample preparation was performed by non-contact liquid dispenser, I-DOT Mini AQ Edition (50 nL sample solution and 50 nL matrix solution) or pipetted manually (100 nL sample solution and 100 nL matrix solution). This is approx. 5-10 times less compared to standard sample preparation protocol for non-modified MALDI targets (0.5 μL sample solution and 0.5 μL matrix solution).

To show the increase in sensitivity, Glu1-Fibrinopeptide B (GluFib) was used as analyte because this analyte is also used to specify the sensitivity of the used MALDI instruments. For these experiments, CHCA was used as MALDI matrix.

MALDI analysis was performed with a MALDI-8020 bench-top MALDI time-of-flight (TOF) instrument and a MALDIminiTM-1 bench-top MALDI digital ion trap (DIT) instrument.

Results MALDI-8020

On the MALDI-8020 bench-top TOF instrument GluFib could still be detected in a concentration of 10 amol on target (Fig 2). This is far below the specified sensitivity of 250 amol. The gain in sensitivity is about factor 25.

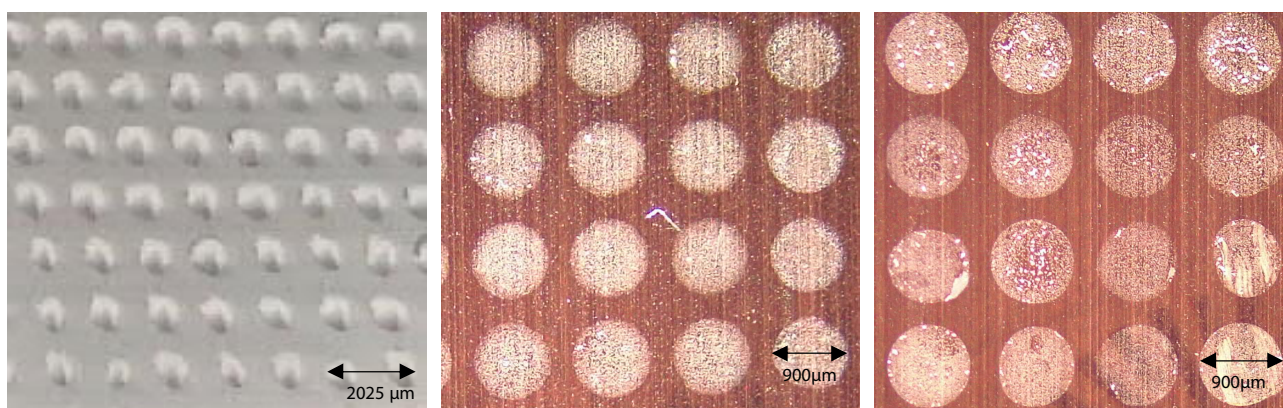
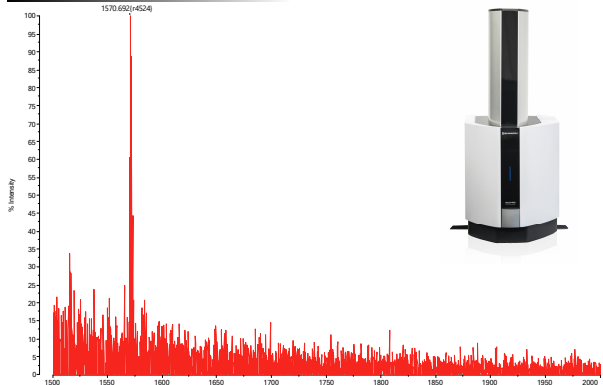


Fig. 1 a) Water droplets show the omniphilic / omniphobic pattern due to the surface modification, MALDI samples prepared by b) non-contact liquid dispenser and c) manually.

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Processed data (averaged) : 0.7 mV [sum=138.7 mV], Unsmoothed_profiles # 1 - 200



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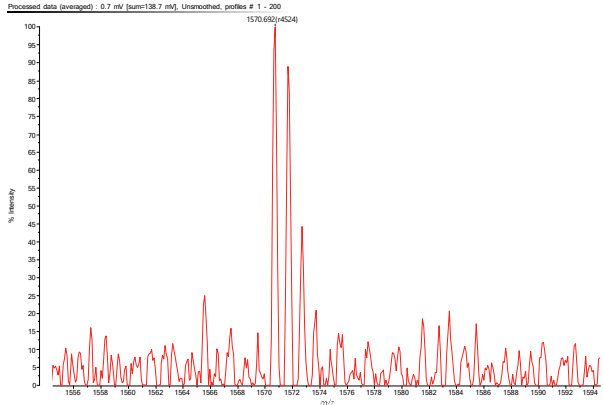


Fig. 2 Mass spectrum recorded by MALDI-8020 bench-top MALDI-TOF instrument of 10 amol GluFib prepared on the target modified by Aquarray.

■ Results MALDImini-1

To compare these results, the sensitivity on the MALDImini-1 bench-top MALDI-DIT instrument was also evaluated. The limit of detection for GluFib was found to be around 50 amol (Fig. 3). Compared to the specified sensitivity of 1 fmol, this corresponds to a gain factor of 20.

■ Conclusion

This evaluation showed that Aquarray's surface modification can enhance MALDI analysis in various dimensions. Due to the omniphilic / omniphobic surface the samples can be applied on a smaller area on the target and in higher density. That way, 14 times more samples can be analyzed.

The use of a non-contact liquid dispenser allowed the automated sample preparation for high-throughput applications.

Both lower solvent volume and higher sensitivity help to save sample and facilitates analysis of rare samples. On two different MALDI instruments with two different mass analysis technologies, the approx. gain of sensitivity was around the same factor i. e. between 20-25. It is assumed that on other MALDI instruments a similar gain in sensitivity can be observed.

Fleximass and MALDImini are trademarks of Shimadzu Corporation in Japan and/or other countries.

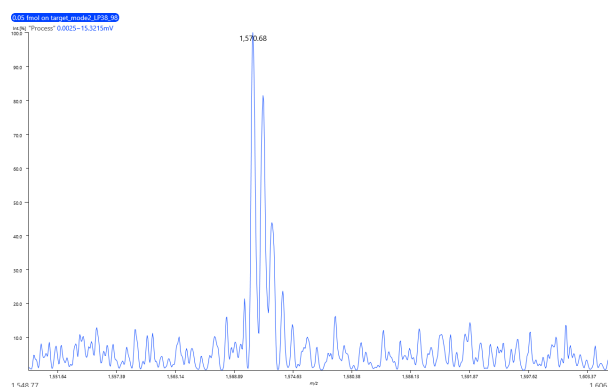
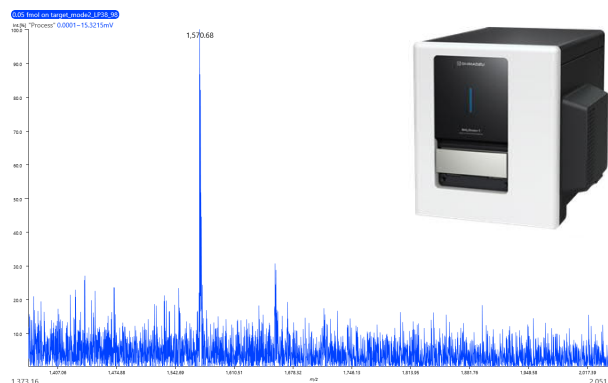


Fig. 3 Mass spectrum recorded by MALDImini-1 bench-top MALDI-DIT instrument of 50 amol GluFib prepared on the target modified by Aquarray.

■ Acknowledgments

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■ References

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