

Streamlining MALDI Workflows in the Microbiology Lab

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Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) has been used as a microbial identification technique for more than a decade, overcoming the limitations of classical biochemical methods that can take days for a result and often lack specificity. MALDI is a soft ionization approach used in mass spectrometry (MS) that allows the analysis of biomolecules (such as DNA, ribosomal proteins, peptides and sugars) and large organic molecules (such as polymers and other macromolecules). MALDI-TOF MS is nowadays the state-of-the-art technique for species identification of bacteria, yeast, and fungi, using proteomic fingerprinting.

Speed and accuracy of analysis, straightforward sample preparation, and simple generation of results make MALDI-TOF MS particularly suitable for routine and high-throughput microbial identification. For this purpose, microbial samples are spotted onto MALDI-TOF MS target plates and overlaid with a specific MALDI matrix before analysis. The subsequently acquired mass spectra are then matched against a library of reference spectra. The MALDI-TOF MS target plate, commonly metal, plastic or ceramic, holds the MALDI-TOF MS samples on individual spots for analysis. Two main types of MALDI-TOF MS target plates can be used: disposable and reusable plates. The key difference is that reusable plates require thorough cleaning and checking in between each run, whereas disposable target plates are ready to use. This article discusses the advantages of disposable target plates for cost-effective, convenient microorganism identification workflows.

Rapid, robust workflows

In mid- and high-throughput laboratories, for example in the pharmaceutical or food industry, rapid microbial identification is required for quality control (QC) and timely delivery to market, hence efficient, convenient, and fast workflows are vital. Additionally, in highly regulated industries such as pharma, QC methods must be verified by the lab. Commonly used reusable steel MALDI target plates place that responsibility on the microbiology lab, as they require an additional cleaning step which must often be documented as evidence that cleaning procedures are being followed in case of an audit. However, using disposable target plates can support workflows in regulated environments, such as ISO 15189.

Disposable target plates relieve the burden of validation of the washing procedure and eliminate the risk of contamination and memory effects from previous isolates. Therefore, when using disposable MALDI target plates, the quality management (QM) part of the MALDI-based workflow is much easier and saves time for the management and laboratory team to focus on their core tasks.

In addition, this ready-to-use concept is particularly advantageous in cases where urgent samples require testing; for example, in the food industry, to identify microbial contamination quickly and make decisions to recall or withhold a product from the market.

Aside from the additional cleaning step required for reusable target plates, the workflow of disposable and reusable MALDI-TOF MS target plates is identical, making it simple to make the switch while keeping identification protocols the same. Moreover, disposable target plates offer the same level of performance as reusable target plates.

On the MALDI-TOF market, a range of disposable MALDI target plates are available, in different sizes and materials. One important factor contributing to the robustness of MALDI-TOF MS microbial identification is the flatness of the target plate surface. Some materials, such as plastic, are more susceptible to minor variations in the surface, which can cause mass shifts in the spectra and require additional calibration spots. For example, target plates built on a ceramic base (Figure 1), have a perfectly flat surface that eliminates the need for predefined calibrant positions and offers flexibility to apply the calibrant anywhere on the target plate, allowing use of the whole disposable target plate. Only one calibration position is needed for excellent mass accuracy over the whole target plate, which means more spots are available for sample measurements. In addition, the ceramic material is very robust so target plates do not break if they, for example, fall from the bench to the floor. There are no restrictions in terms of software and workflow to use one target plate for several MALDI-TOF acquisitions when the target plate is not yet completely filled with samples. For example, the user can spot only a few samples, complete the MALDI run, and come back later to use the free positions of the respective target plate for a new spotting (and run).

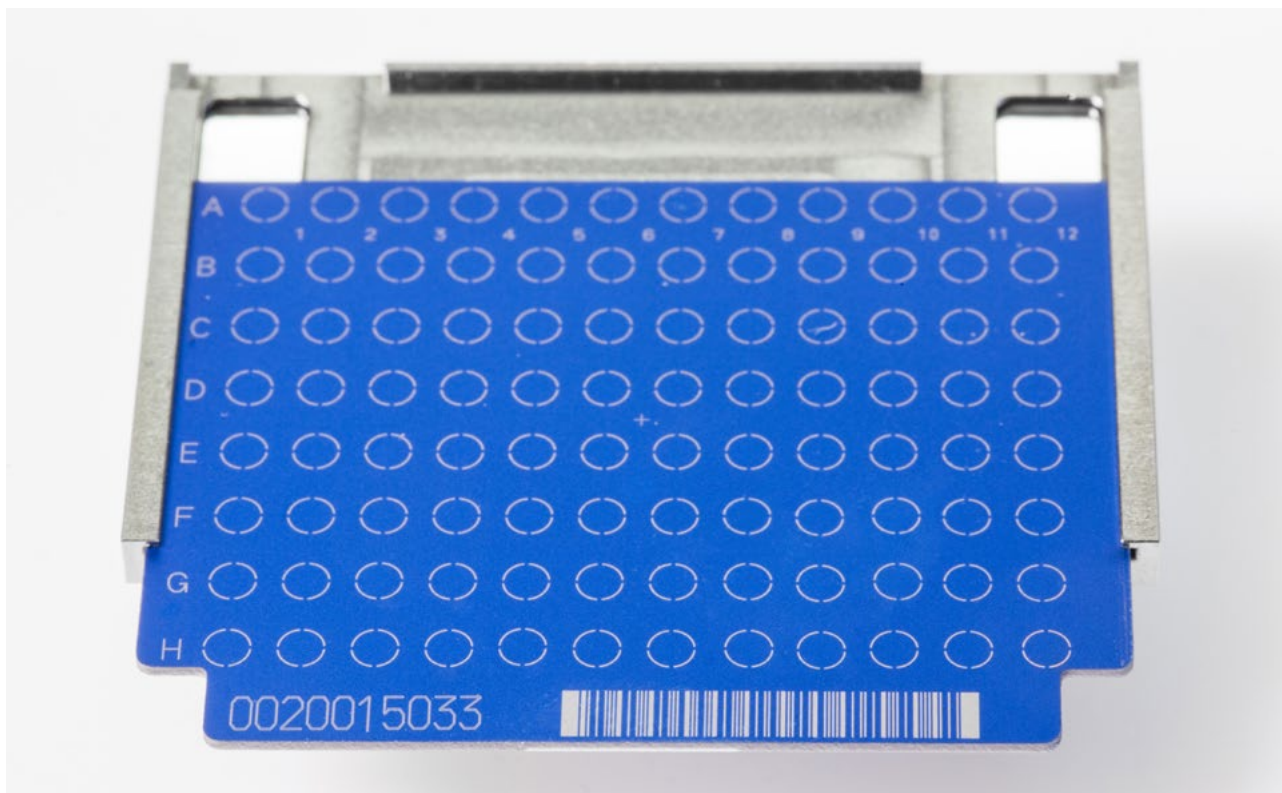


Figure 1: Ceramic-based disposable MALDI-TOF MS target plate. Credit: Bruker Daltonics.

Cost considerations

While disposable MALDI-TOF MS target plates may have a slightly higher cost upfront, there are no hidden costs associated with their use or maintenance. In contrast, for reusable target plates, labs would need to purchase, store, use, and dispose of a range of organic chemicals required for the cleaning procedure. As well as the financial implications, organic cleaning solvents require additional considerations, such as the use of fume hoods and their maintenance, extra safety protocols for staff, and appropriate disposal measures.

In summary, the complete time for the laboratory that is needed to order, maintain and process this “overhead” would need to be considered in a direct cost comparison to reusable target plates. Additionally, the availability and price of organic solvents fluctuates, leading to challenges for those responsible for the lab’s inventory and sometimes making it hard to predict the future costs.

Spot on sample prep

In high-throughput labs, microbiologists can save time waiting for liquid samples to dry by using ceramic disposable target plates, the surface of which dries around 10% more quickly than stainless steel plates.

These disposable plates also incorporate technology designed to provide a hydrophobic surface that surrounds the hydrophilic sample positions or “anchors”. This allows liquid samples to spread evenly on the target spot without cross contamination between positions (Figure 2).

Avoiding cross contamination between spots on target plates is critical for MALDI-TOF MS microbial identification workflows.

This technology is particularly useful for the sample preparation of slow-drying liquid samples, which are more prone to running between spot positions and causing cross contamination. Because the droplet stops spreading at the hydrophobic rim and remains on the hydrophilic anchor, it is much easier for the user to place samples on the target without any risk of droplets running into each other on the target plate. Finally, for special sample protocols which require the addition of larger volume (e.g. 3-5 μ l of sample) the anchor effect keeps the volume concentrated on the respective spot position without any risk for cross contamination.

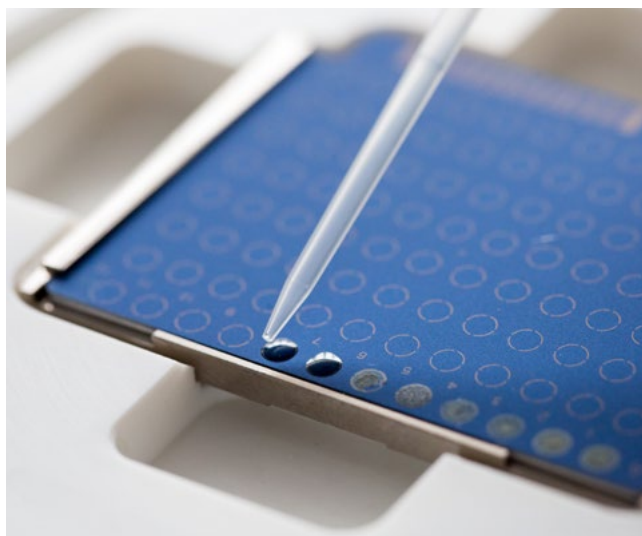


Figure 2: Dedicated technology prevents samples running between spot positions on ceramic disposable target plates. Credit: Bruker Daltonics.

The key to convenience

Their ready-to-use capability and simplified workflow, resulting in a time-saving benefit, make disposable MALDI-TOF MS target plates a very convenient solution for mid- to high-throughput microbiology labs. To add to the enhanced efficiency and time savings, disposable target plates can be used with ready-to-use bacterial test standards (BTS), and pre-portioned HCCA (α -Cyano-4-hydroxycinnamic acid) matrix for simple, safe, quick, and accurate microbial identification.

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