New from Hardy Diagnostics

MALDI-TOF

For fast and accurate ID of bacteria and fungi

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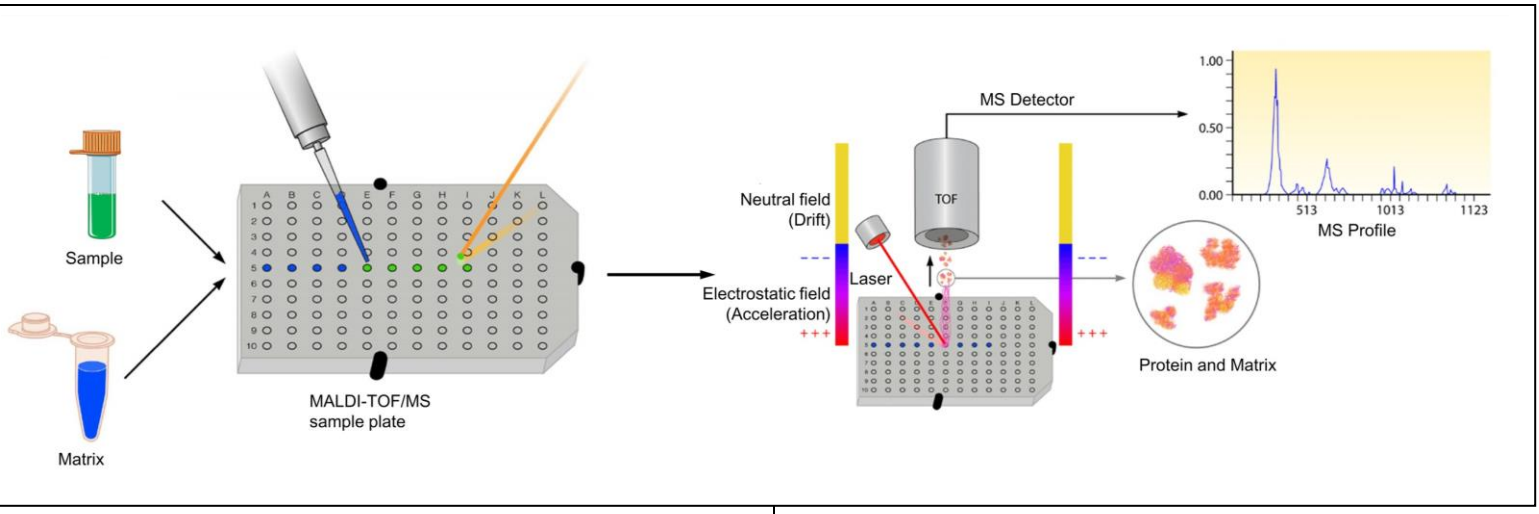
The Matrix-assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry (MALDI-TOF MS) is a critically acclaimed and widely utilized technology that services a multitude of applications in industry and research.

Discovered in the early 1900's, Mass Spectrometry was limited in application to assay small chemical compounds, but the advent of MALDI-TOF technology has increased the breadth of suitability to render the analysis of macromolecules, such as proteins,(1).

Applications in proteomics research have paved the way for MALDI-TOF utility in Microbiology applications for the identification of microorganisms down to the species level through Peptide Mass Fingerprinting (PMF), also known as proteomic fingerprinting. (2) MALDI-TOF technology has developed good rapport within the Microbiology community. The Clinical

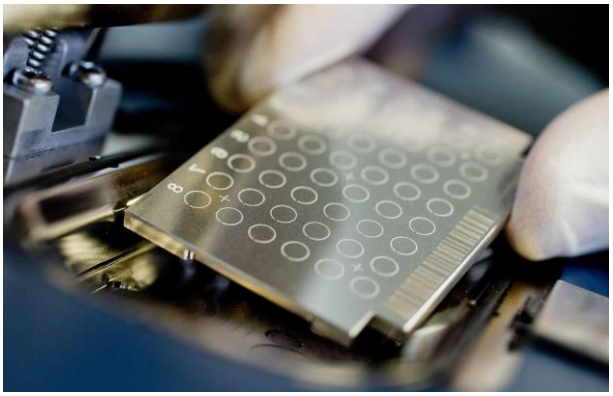


and Laboratory Standards Institute (CLSI) even published a guideline, the M58, to provide recommendations for laboratories looking to implement these systems—a vote of confidence for this technology and its applications. (3)

Mass Spectrometry is an analytical technique that measures the mass (m) to charge (z) ratio (m/z), which can be used to determine exact molecular weights, of one or more molecules from a sample. (4,5) MALDI is a method of "soft ionization" which minimizes the degree of molecular fragmentation, preserving sample integrity, and is paramount for microbial identification. This unique feature is enabled by the utilization of a matrix, an organic acid capable of absorbing ultraviolet light at the emission wavelength from the laser during irradiation; furthermore, the matrix ensures ion formation of the analyte through protonation.

When the matrix and sample are added together and allowed to dry, the mixture crystallizes, leading to the formation of co-crystals. The degree of homogeneity of these co-crystals confers the "shot-to-shot" reproducibility of the acquired mass spectra, and removes the need for manual intervention, allowing for automation. (6)

Prepared samples are introduced into a high-vacuum environment, typically about 1×10^{-8} to 1×10^{-9} bar, to ensure free flight of the molecules following laser irradiation, allowing for separation based solely on mass (m) and charge (z). Pulse laser irradiation of the co-crystallized sample and matrix mixture causes rapid heating of the mixture, which transitions into a gaseous state along with the microbial proteins. Gaseous-state peptides are converted to ions, electrically charged particles, through proton transfer with the matrix (1).



The protonated ions are then accelerated through an electrical field and separated by a voltage potential (V_0) difference based on each ion's mass-to-charge (m/z) ratio. The ions accelerate through the flight tube until they reach the terminus where they are detected and their Time-of-Flight (TOF) is recorded. These measurements are used to generate a mass spectrum, or proteomic fingerprint.

The resulting mass spectrum is then compared against a reference database containing well-characterized type strains of specific genera, species, and subspecies to identify the microbial sample. (1)

While advancements in nucleic acid sequencing technologies have enabled highly specific detection rates of microorganisms, these technologies are too time-consuming and costly to be commonplace. (7) Numerous studies have demonstrated the higher accuracy, faster time-to-result, and lower cost provided by MALDI-TOF technology when compared to classical methods. (8,9,10)

With a promising record and a multitude of potential applications, MALDI has proven to be a swift and reliable method for the identification of microorganisms, both bacterial and fungal.

Due to its complex nature, the initial cost of MALDI equipment is a substantial investment, but the overall cost of identifying an individual species is significantly lower than other standard methods. (11) The time to result is also significantly shorter than conventional techniques, with an estimated mean preparation and wait time of six minutes compared to 5-48 hours. (12)

Studies have shown that the high initial cost of the MALDI-TOF MS can be offset within a few years of use. (11) Rapid identification of microorganisms and cost saving abilities make MALDI-TOF MS an invaluable piece of laboratory technology.

Hardy Diagnostics is proud to introduce our own benchtop MALDI system, the Autof ms 1000. The Autof ms 1000 provides automated, high-speed and high-confidence identification and taxonomical classification, and has the most expansive database in the industry. For more information on this exciting new technology, visit our website here.

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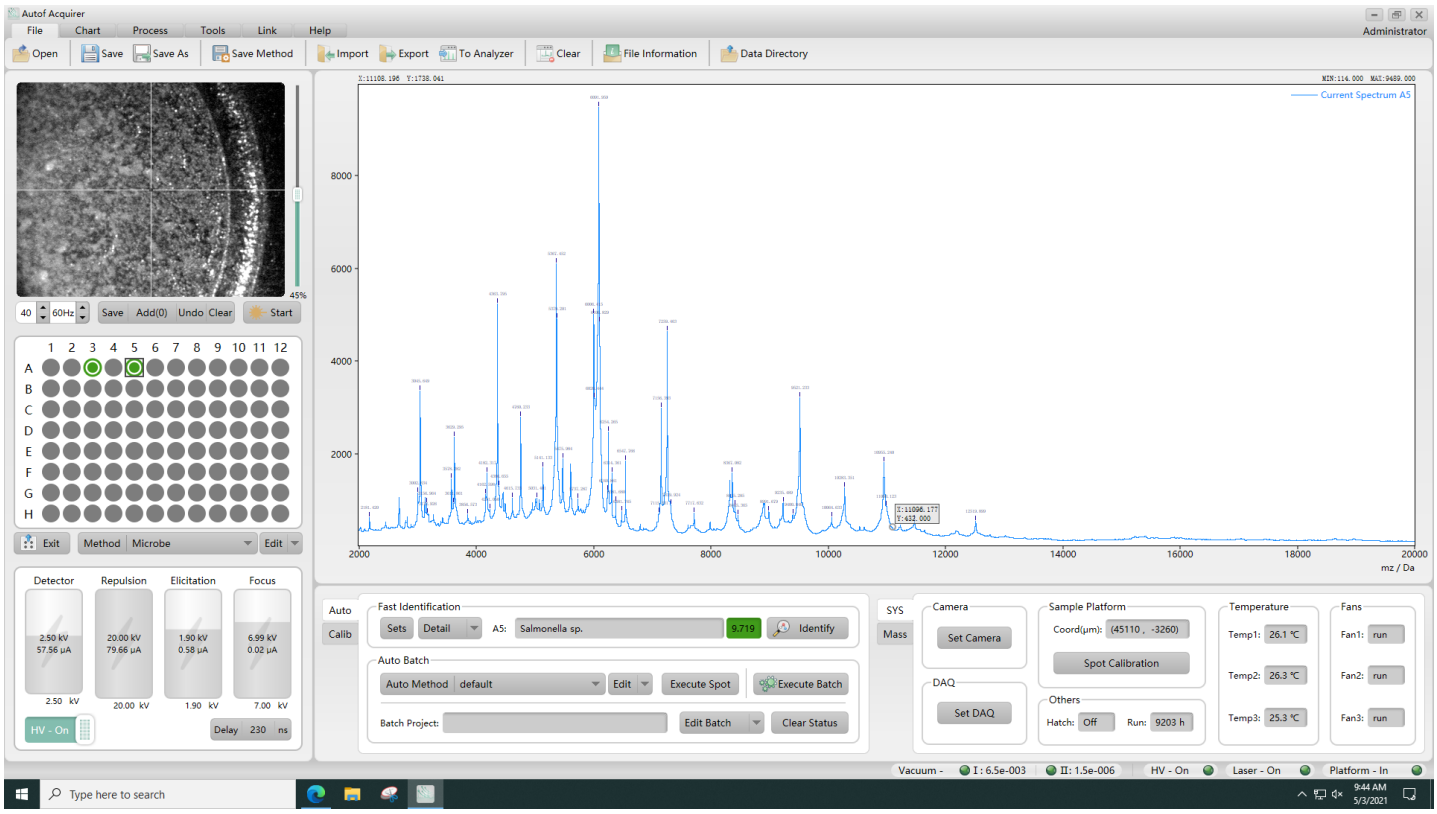

Figure 1: This is a sample print out from the Autof ms 1000. It shows the graphical results of testing a sample of Salmonella.