

Abstract

This doctoral dissertation investigates the application of advanced analytical techniques, particularly MALDI-TOF MS spectrometry, to study antimicrobial resistance mechanisms, identify clinical microorganisms, and monitor antibiotic therapy. The study integrates “-omics” approaches, including proteomics and metabolomics, with modern diagnostic tools, thereby significantly advancing personalized medicine and microbiological diagnostics.

MALDI-TOF MS was employed for the rapid and accurate identification of bacteria isolated from clinical samples, including hard-to-heal wounds and respiratory tract infections. The MBT-STAR-BL and MBT-ASTRA assays were used to assess resistance to various classes of antibiotics, such as beta-lactams, macrolides, and quinolones. Particular emphasis was placed on combining MALDI-TOF MS with complementary analytical methods, enabling a comprehensive evaluation of pathogens, their adaptive mechanisms, and their biological properties.

A key component of the dissertation was the development of innovative microextraction fibers coated with molecularly imprinted polymers (MIPs) for the isolation of antibiotics, including amoxicillin, cefotaxime, and metronidazole, from plasma samples collected from intensive care unit patients. When combined with liquid chromatography–tandem mass spectrometry (SPME-LC-MS/MS), this approach enabled precise quantification of drug concentrations and their metabolites in biological samples. This represents the first application of this methodology in clinical diagnostics and constitutes a significant advancement in the personalization of pharmacological therapy.

The results demonstrated that the developed methods significantly reduce diagnostic time while improving the accuracy of pathogen identification and antimicrobial resistance assessment. The dissertation also presents innovative sample preparation strategies, including the use of silver nanoparticles as an alternative matrix for laser desorption/ionization, which enhances the analysis of low-molecular-weight compounds.

The diagnostic tools developed in this research show strong potential for routine application in microbiological diagnostics and antibiotic therapy monitoring. By integrating mass spectrometry, “-omics” methodologies, and advanced sample preparation techniques, this interdisciplinary study contributes to efforts to combat antimicrobial resistance and supports the development of modern diagnostic and therapeutic strategies, ultimately improving healthcare quality and the effectiveness of bacterial infection treatment.

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