

MALDI-TOF based Metabolomic approach

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Abstract- Metabolomics is the study of metabolite profiles in biological and pathological samples (urine, plasma, saliva, blood fat biopsies, plant cells, bacteria viruses and fungi. The metabolome is the most predictive phenotype: consequently, the comprehensive and quantitative research study of metabolites is a desirable tool for diagnosing disease, identifying new therapeutic targets and enabling appropriate treatments. In addition to other Mass Spectrometry (MS) methodologies used for determining the localization of small molecules in tissue, MALDI-TOF/TOF can also be used to detect drugs and their metabolites in tissue without the burden of matrix interference.

Keywords- Metabolomics, MALDI-TOF, Mass Spectrometry

Introduction

Metabolomics is concept, which has been defined as the science of the comprehensive monitoring of the metabolic complement in biological systems [2]. Metabolomics provides information about biological systems that cannot be obtained by the classical 'omics' approaches transcriptomics, genomics and proteomics. Metabolomic approaches are under dynamic development and several synonyms have been suggested, such as metabonomics [3], metabolite profiling or fingerprinting. Metabolomics potentially holistic approach to metabolome analysis is driven primarily by recent advances in mass spectrometry (MS) technology and by the goals of functional genomics research. The broadest overview of metabolic composition is very complex and entails establishing a multifaceted and fully integrated strategy for optimal sample extraction, metabolite separation, identification, detection, automated data gathering, handling, analysis and lastly quantification. computational developments and analytical techniques are essential to achieve this goal [4]. Owing to their soft nature of ionization, MALDI- MS and electrospray ionization- mass spectrometry (ESI-MS) have been at the forefront of bioanalytical research with far reaching applications in genomics, proteomics, biological imaging and metabolomics. In MALDI-MS, biomolecules mixed with matrices (small, UV-absorbing compounds) and exposed to laser pulses form gas-phase ions that are typically measured in time-of-flight (TOF) mass analyzers [1].

MALDI-TOF in bioremediation

MALDI-TOF-MS techniques are used to identify proteins of interest from 2-D gels, as well as to detect and identify bacteria, viruses, fungal spores and low-mass compounds in environmental samples. The complex mass spectra of environmental samples can be used to create characteristic fingerprinting databases to detect many site-specific microorganisms. Bioremediation, MALDI-TOFMS can detect

specific bacterial signature proteins and biomarkers (primary and secondary metabolites) from site-specific samples for the taxonomic identification of potential microorganisms [5]. A form of direct sample analysis on a microchip using MALDI-TOF-MS – SELDI-TOF-MS – is another promising analytical technique for site-specific samples and of differentially expressed signature proteins in blue mussels (*Mytilus edulis*) exposed to PAHs and heavy metals were analyzed using SELDI-TOF-MS. Although SELDI analysis has been useful for identifying potential biomarkers in clinical research, some have questioned its reproducibility and specificity [6].

Metabolomic Analysis of mammalian tissue

MS-based metabolomics research demonstrates a surprisingly large effect of the gut microbiome on mammalian blood metabolite is studied. Plasma extracts from germ-free mice were compared with samples from conventional (conv) animals by using various MS-based methods. Many of features are detected in only 1 sample set, with the majority of these being unique to the conv animals, whereas approximately 10% of all features observed in both sample sets showed significant changes in their relative signal intensity. Amino acid metabolites are particularly affected. Multiple organic acids containing phenyl groups are also greatly increased in the presence of gut microbes. A broad, drug-like phase II metabolic response of the host to metabolites generated by the microbiome is observed and suggesting that the gut microflora has a direct impact on the drug metabolism capacity of the host. These results suggest a significant interplay between bacterial and mammalian metabolism [7]. Other analysis study the identification of specific metabolites as well as mechanisms of their increase using MALDI-TOF illustrates the potential of mass-based metabolomics to address problems in CNS biochemistry and neurovirology, as well as neurodegenerative diseases [8]. MALDI metabolomics is also used

to identify genetic variants associated with heart failure by using a rat model of the human disease [9]. Metabonomic method for the investigation of abnormal metabolic process in both serum and liver tissue of liver transplanted rats is studied. Thirty-four metabolites in serum and 29 metabolites in liver are identified. Results of correlation analysis illustrated metabolites with similar function exhibited similar variations in liver and serum. The data processed by principle component analysis (PCA) showed time-dependent biochemical variations. The present study may offer specific putative pathways in the pathophysiological mechanism of orthotopic liver transplantation [10]. The present study investigated small molecule analysis of urinary samples as a noninvasive method to detect acute cellular renal allograft rejection. Matrix-assisted laser desorption/ionization Fourier transform mass spectrometry (MALDI-FTMS) is used to analyze 15 urinary samples from transplant patients with different grades of biopsy showing improved clinical acute cellular rejection (ACR) and 24 urinary samples from 8 transplant patients without evidence of rejection [12].

Metabolomics in understanding cellular responses in plants

Natural shift is taking place in the approaches being adopted by plant scientists in response to the accessibility of systems-based technology platforms. Metabolomics is one such field, which involves a comprehensive non-biased analysis of metabolites in a given cell at a specific time. Although microbes may prove to be the richest overall source of metabolites, plants are the source of the most complex individual mixtures. Mariet van der Werf (TNO-Food, Zeist, The Netherlands) reported that it has been predicted that bacterial genomes already sequenced can support the biosynthesis of just a few hundred metabolites (e.g., 580 for *Bacillus subtilis* and 800 for *Escherichia coli*), but for individual plants, this value is likely to be in the tens of thousands [14]. These metabolic richness comes not just from the number of genes present (20,000 to 50,000) but also from multiple substrate specificities for many enzymes, subcellular compartmentation and the occurrence of nonenzymic reactions. Approximately more than 50,000 different compounds have been elucidated in plants and it is predicted that the final figure for the plant kingdom will approach or even exceed 200,000 [13]. Denise Jacobs, presented data, which correlated proteome changes with alkaloid accumulation in periwinkle (*Catharanthus roseus*) cell cultures and showed that as many as one-third of the 2000 proteins visualized by two-dimensional gel electrophoresis were correlated with alkaloid accumulation. Several protein identifications via matrix-assisted laser-desorption ionization (MALDI)-TOF are

presented and represents a considerable challenge for plant scientists [11].

Conclusion

The future of metabolomics rests with its ability to monitor subtle changes in the metabolome which occur prior to the detection of a gross phenotypic change reflecting disease. The integrated analysis of metabolomics and other omics may provide more sensitive ways to detect changes related to disease and discover new novel biomarkers. Knowledge regarding these multivariant characteristics is critical for establishing validated and predictive metabolomic models for cancer prevention. Understanding the metabolome will not only provide insights into the critical sites of regulation in health promotion, but will also assist in identifying intermediate or surrogate cancer biomarkers for establishing preemptive and preventative or therapeutic approaches for health. While unraveling the metabolome will not be simple, the societal implications are enormous.

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