Microbial Classification/Identification and Authentication Using MALDI-TOF Intact Cell Mass Spectrometry

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The analysis and characterisation of macromolecules and their complexes which are the core of life, as well as the spectral typing of microbial cells for their taxonomic classification/identification and authentication using spectral analysis by Matrix Assisted Laser Desorption Ionisation – Time Of Flight Mass Spectrometry (MALDI-TOF MS) are both modern approaches for the life sciences and biotechnology studies. MALDI-TOF MS emerged in the late 1980s as a sound technique to investigate the mass spectrometry of molecular high-mass of organic compounds through a soft ionisation of the molecules resulting in minimum fragmentation. This technique has contributed to increasing scientific knowledge about the microorganisms and is now used as a reliable tool for rapid tests in hospitals and health centres. In this case the interest of the art in question is the analysis of the intact cell. The spectrum generated is analysed as fingerprint and the technique is called MALDI-TOF IC (Intact Cell) MS. In the MALDI-TOF ICMS technique the microbiological sample is covered with a UV-absorbing organic compound called matrix leading to a crystallised mixture. Then the crystallised sample is submitted in a vacuum system that is targeted and irradiated with a pulsed light from a nitrogen laser (337 nm). The charged matrix molecules and/or clusters, transfer protons onto the sample molecules (e.g. peptide or proteins) in the expanding plume. The generated ions are accelerated into the TOF analyser, in which ions are separated according to their "time-of-flight" which is a function of molecular mass to charge. The TOF analyser determines the molecular mass to charge (*m/z*) ratio of ions by measuring velocities from accelerating ions to defined kinetic energies after calibration of the instrument with molecules of known molecular mass. In MALDI-TOF ICMS technique the linear mode set covers the mass range of 2 - 20 kDa. Nowadays, MALDI-TOF ICMS technique has been contributed for a great increase of knowledge in the microbial field. Information about each microorganism (e.g. morphological description, physiological and biochemical properties, ecological roles, and societal risks or benefits) is the key element in this process. Identifications can been a long and seemingly never-ended process with frequent revisions of the taxonomic schemes. These changes make identifications even more complicated for the non specialised researchers as each taxonomic group has specialised literature, terminology and characters. Taking it in account MALDI-TOF ICMS is already been used as sound technique in microbial classification/identification and authentication and, as a matter of consequence for quality control programmes for culture collections.

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