

ISOTOPE-CODED PROTEIN LABELLING (ICPL) AS A TOOL FOR THE QUANTITATIVE ANALYSIS OF A BACTERIAL PROTEOME

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The development of new quantitative tools for the analysis of whole proteomes (differential proteomics) has been continuous during the past years. Traditional approaches, based on previous fractionation of complex proteomes by 2D-PAGE and comparison of the gel images generated thereby, are being complemented by new techniques that commonly make use of isotopic labeling of the samples to be compared followed by mass spectrometric analysis. Isotopic labeling can be performed *in vivo* (e.g., SILAC), chemically (e.g., ICAT, iTRAQ, ICPL) or enzymatically (e.g., ¹⁶O/¹⁸O). Isotope-coded protein labeling (ICPL) has been described recently as a technique to efficiently label the abundant amino groups present in proteins and allows comparison of up to four samples at the same time. Theoretically, ICPL overcomes some of the disadvantages found in other chemical labeling techniques, such as iTRAQ (poor detectability of the reporter ions in most of the mass spectrometers) or ICAT (low number of potentially labeled residues). However, the number of articles published to date using ICPL is noticeably low and no clear consensus exists on its feasibility as a quantitative tool. Here we describe the results obtained after quantitative comparison between the proteomes of *Salmonella enterica serovar typhimurium* and two derived strains harboring mutations that have been shown to drastically affect the bacterial proteome. Results were analyzed using software specifically designed for differential proteomics (WARP-LC). More than 500 different proteins were identified while about 150 proteins could be quantified as well. Our results demonstrate that ICPL may be a very valuable alternative to iTRAQ when the analysis of iTRAQ specific reporter ions is not possible and clearly overcomes the limitations imposed by other non-isobaric labeling approaches, such as ICAT.