## LABEL-FREE QUANTIFICATION BASED ON DATA INDEPENDENT ACQUISITION MASS SPECTROMETRY

## K. Aloria<sup>1</sup>, <u>M.J. Omaetxebarria</u><sup>2</sup>, J.P.C. Vissers<sup>3</sup>, A. Fullaondo<sup>4</sup>, J.M. Arizmendi<sup>2</sup>

 <sup>1</sup>Proteomics Unit, SGIker, University of the Basque Country, Leioa, Bizkaia;
<sup>2</sup>Departament of Biochemistry and Molecular Biology, University of the Basque Country, Leioa, Bizkaia;
<sup>3</sup>Waters Corporation, Atlas Park, Simonsway, Manchester M22 5PP, United Kingdom;
<sup>4</sup>Departament of Genetics, Physical Anthrpology and Animal Physiology, University of the Basque Country, Leioa, Bizkaia.

The quantification of differentially expressed proteins is a key area where proteomics technology has made great progress over the last years. The development of a data independent, alternate scanning acquisition, where no precursor is selected and low and high collision energy data is alternatively acquired, affords accurate mass measurements used for both protein identification and label-free based quantification. Furthermore, beyond new experimental approaches, bioinformatics software is in constant development in order to obtain more robust tools for data processing, qualitative analysis and quantification. Recently, an LC-MS based absolute quantification method based on the comparison of the 3 most intense peptides of each protein with a known protein standard has been developed. In this study we have tested this approach using known amounts of protein standards at different ratios. 4 proteins have been spiked in an E.coli lysate background and their absolute and relative protein abundance measured. Furthermore, we have used the same experimental approach to analyse the protein expression pattern of EGF treated and untreated human MDA-MB-468 breast cancer cells. Finally, the same experimental data has been analysed with a probabilistic based quantification algorithm, where relative quantification of all peptides and proteins is performed. The obtained results with both quantification methods have been compared.