

OF4 ETISEQ – an algorithm for automated elution time ion sequencing of concurrently fragmented peptides for mass spectrometry-based proteomics

Jason W. H. Wong¹, Alexander B. Schwahn², Kevin M. Downard²

¹ UNSW Cancer Research Centre, University of New South Wales, Sydney, Australia

² School of Molecular and Microbial Biosciences, University of Sydney, Sydney, Australia.

In-source CID, shotgun CID, parallel CID, MS^E, peptide mixture fragmentation

Development of an algorithm to automatically define parent-fragment peptide ion lineage.

Data-dependent acquisition of LC-MS/MS data has been the principal method for collecting peptide fragmentation data for protein identification and quantification. However, the method is not without limitations including dynamic range restrictions and reduced selected ion chromatographic resolution for protein quantification. Proteomics data acquired by concurrent peptide fragmentation, which has been variably termed, shotgun CID [1], parallel CID and MS^E [2] has been demonstrated to overcome such limitations. Nevertheless, concurrent peptide fragmentation data acquisition remains to be widely adopted due to the lack of direct compatibility with existing MS/MS spectra identification software. An algorithm called Elution Time Ion Sequencing (ETISEQ), has been developed to enable automated conversion of concurrent peptide fragmentation data to data dependent LC-MS/MS-like data. ETISEQ generates MS/MS-like spectra based on the correlation of parent and fragment ion elution profiles. The performance of ETISEQ is demonstrated using concurrent peptide fragmentation data from tryptic digests of standard proteins and whole influenza virus. It is shown that the number of unique peptides identified from the digests is broadly comparable between ETISEQ processed concurrent peptide fragmentation data and the data dependent acquired LC-MS/MS data. The ETISEQ algorithm can be easily integrated into MS/MS analysis platforms, it is anticipated that it will popularize concurrent peptide fragmentation data acquisition in proteomics laboratories.

[1] S. Purvine, J.T. Eppel, E.C. Yi, D.R. Goodlett *Proteomics* 2003, 3, 847-850.

[2] A.B. Chakraborty, S.J. Berger, J.C. Gebler *Rapid Commun. Mass Spectrom.* 2007, 21, 730-744.