Improved Method for Comparative Proteomics

The University of North Dakota has developed a series of technology that provides a novel method for sample preparation and comparison of proteins in biological samples. This technology offers an excellent opportunity for the development of an efficient and convenient research or diagnostic test.

Current Issues in Comparative Proteomics:

The term, “comparative proteomics” refers to the approaches to determine the changes in protein expressions in biological samples. Defining differential regulation of proteins in response to various biological conditions facilitates our understanding for underlying mechanisms of physiological phenomena as well as disease processes. Current methods for comparative proteomics employ 2-dimensional gel electrophoresis (2D gel) or mass spectrometry (MS). The 2D gel technique involves lengthy procedures and has limited dynamic ranges, while the MS method frequently produces inaccurate results due to inconsistent isotope incorporation during sample peptide labeling. These disadvantages, however, can be avoided by our new methodology.

Our Technology:

To achieve consistent labeling of samples for MS analysis, the number of isotopic atoms, or “labels,” to be incorporated into peptides needs to be regulated. Our new technology takes advantage of the innate property of peptidases and controls the enzymatic condition so that a user can specify the number of $^{18}$O atoms to be incorporated into sample peptides: either only one or only two, but not mixture of both. Controlling the number of isotopic atoms on sample peptides avoids miscalculation of protein amounts. This technology can be applied for the development of a sample preparation kit to make MS-based comparative proteomics simple and reliable for a research or diagnostic use.

Advantages:

- Provides extremely accurate measurements of proteins by minimizing procedural errors
- Circumvents running laborious 2-dimensional gels and offers wide dynamic ranges
- Can be used to develop a protein labeling kit as a simple and dependable comparative proteomics tool

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Mass spectra of peptides that have been labeled with one or two $^{18}$O at their carboxyl termini. Upper panel shows the mass spectra of a peptide labeled by a peptidase, Lys-N, with conditioned or unconditioned reaction buffer. Lower panel the mass spectra of a peptide labeled by a protease, trypsin, or a peptidase, Lys-C, with conditioned or unconditioned reaction buffer. Using the conditioned buffers either one or two $^{18}$O labeling of peptides can be achieved.