A Multidimensional Separation and Visualization Strategy for Proteomic Profiling of Human Tissue and Cell Lines

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Introduction

The accurate fractionation, recovery, quantitation and characterization of sets of individual proteins from complex proteomes are capabilities that are increasingly essential to the growth and success of biomedical research and education. The ability to characterize a complex protein sample by mass spectrometry depends on the power and sensitivity of the separation techniques employed prior to the MS analysis. A critical component of such studies is the design and implementation of a multidimensional separation strategy. Until recently, the analysis of whole proteomes has been heavily dependent on 2-Dimensional Gel Electrophoresis (2DGE) methods. Currently, however, there is a growing interest in the use of gel-free multidimensional separation techniques, which promise to gain higher sensitivity and resolution through automation.

OGE pH Gradient

Low pH High pH

2nd Dimension fraction collecting

Reversed-phase Separation

RP operational Temp = 80 C

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MDLC Visualization Software

MDV is an Agilent prototype software tool for providing a visualization environment for two-dimensional protein data. It was recently developed for the integration of Off-Gel isoelectric focusing and Reverse-phase separations and to provide enablement of multiple proteome comparative analysis.

Conclusions

Until recently, the analysis of whole proteomes has been heavily dependent on 2-Dimensional Gel Electrophoresis (2DGE) based approaches. New MS-based technologies have made it possible to replace gel-based 2D separations with a single gel-free multidimensional separation strategy.

References


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