

107c Sample Compatibility Screening and Optimization of Separation Protocols Using Mini Two-Dimensional Gels

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High-resolution two-dimensional gel electrophoresis in combination with sensitive protein staining techniques routinely allows the visualization of more than a thousand protein spots from complex protein mixtures in quantities that are sufficient for subsequent identification by mass spectrometry. Despite the known limitations these qualities still make two-dimensional gel electrophoresis the gold standard for the comparative analysis of protein expression patterns in cells, tissues and other biological samples. However, the high demands on the sample preparation often make the successful analysis of non-standard samples such as biofluids or immunoprecipitates by two-dimensional gel electrophoresis challenging. Another requirement that is often difficult to meet is the need for comparatively high protein amounts in high-resolution two-dimensional gel electrophoresis. To address some of these challenges, we are using mini two-dimensional gel electrophoresis as a pre-screening and optimization tool for the subsequent analysis of protein mixtures by high-resolution two-dimensional gel electrophoresis. Although these mini two-dimensional gels do not offer the resolution necessary for an in-depth analysis of complex protein mixtures, their preparation requires significant less sample and time, and these gels offer valuable information on the compatibility of the prepared protein samples with two-dimensional gel electrophoresis protocols. The results of these mini two-dimensional gels also allow the optimization of first and second dimension focusing ranges in subsequent high-resolution two-dimensional gel electrophoresis experiments.