USING ETTAN™ DIGE SYSTEM FOR ANALYSIS OF PROTEOME CHANGES IN TRANSFORMED E. COLI

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Two-dimensional electrophoresis (2D electrophoresis) is a well established method used to study differences in protein expression caused by environmental changes induced by for example disease, drug or growth factor treatment etc. The 2D DIGE technology with two different samples and an internal standard per gel labelled with CyDye™ DIGE Fluor minimal dyes, significantly reduces the required number of gels compared to conventional 2D electrophoresis. The internal standard significantly reduces the gel to gel variation and thereby improves statistical validity. 2D DIGE and DeCyder™2D software version 6.5 were used to assess significant differential expression as a result of time and temperature changes in transformed E. coli cultures. Temperature and time clustering was shown with DeCyder2D Extended data analysis (EDA) module. Proteins with significant variation were identified by mass spectrometry using MALDI. The levels of target protein from the 2D DIGE analysis were confirmed with fluorescent Western blotting using ECL Plex™.