

**P039** A proteomic approach to identify proteins involved in endoplasmic reticulum stress in INS-1E cells

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It has been suggested that endoplasmic reticulum (ER) stress plays an important role in  $\beta$ -cell death associated with the development of type 1 diabetes. In this study we investigated the impact of the ER-stress inducer cyclopiazonic acid (CPA), a selective and reversible inhibitor of the sarcoplasmic reticulum  $\text{Ca}^{2+}$ -pump, on the protein profile of insulin-producing INS-1E cells. Quantitative changes in protein expression were investigated using 2-dimensional difference gel electrophoresis, after incubation of the cells during 6h, 6h+3h recovery or 12h with CPA. This treatment induced apoptosis, as measured by Hoechst/Propidium Iodide staining (11.5%, 6.1% and 18.2% after 6, 6+3h and 12h, respectively,  $n=4$ ,  $p<0.01$ ). Differential analysis revealed 51 spots in the pH4-7 range ( $p\leq 0.01$ ,  $n=4$ ). By now 24 of these spots have been identified by MALDI-TOF/TOF, revealing proteins involved in ER stress and in insulin processing. Initial results point out to important differences in the regulation of these proteins as compared to cytokine-treated INS-1E. Use of a proteomic approach will enable us to clarify the impact of 'pure' ER-stress on apoptosis induction in  $\beta$ -cells, and compare the results with previously unravelled pathways in cytokine-treated INS-1E cells.