Discriminating highly similar cancer cell lines using Raman spectroscopy and PLS-DA

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A problem often occurring in cancer research is not only to detect cancer cells, but also to determine the cancer cell type. This study reports that it is possible to distinguish the two highly similar cancer cell lines M-4A4 and NM-2C5 originating from the parent cell line MDA-MB-435. M-4A4 and NM-2C5 show equal tumorigenity. But M-4A4 cells establish easily detectable metastases whereas NM-2C5 cells disseminate to distal organs, remained dormant and do not establish metastases. These cells have previously been investigated by fx Mass Spectroscopy as a way to identify possible connections between the tendency of cancer cells to form metastasis and altered expression levels of proteins[1]. Here, the cells were analyzed with Micro Raman mapping with an excitation of 785nm. Preprocessing included subtraction of a background spectrum and normalization by extended multiplicative signal correction. Subsequently, each map was segmented by k-means cluster analysis. Spectra corresponding to the cytoplasm were averaged and used for classification analysis using partial-least-squares discriminant analysis (PLS-DA). Spectra representing the nucleus were excluded as they showed much smaller differences between the two cell lines compared to cytoplasm spectra. PLS-DA is in principle related to other discrimination algorithms, but retain the advantages of normal PLS regression analysis such as handling highly collinear data well. 22 cells from independent preparations were measured resulting in 52 averaged spectra in the classification analysis. A cross-validated five component PLS-DA was applied resulting in 92% correctly classified samples.

References:

[1] R. Leth-Larsen, R. Lund, H. V. Hansen, A. V.Lænkholm, D. Tarin, O. N. Jensen, H. J. Ditzel, *Molecular & Cellular Proteomics* 8, 1436-1449 (2009).