Systems biology approach to compare gene expression in Caco-2 and MCF-7 cells following exposure to silver nanoparticles

Hans Bouwmeester[1], Richard Helsdingen[1], Evelien Kramer[1], Peter Hendriksen[1], Ad Peijnenburg[1], Ruud Peters[1], Hans Marvin[1], Stefan Weigel[1], Meike van der Zande[1].

[1]RIKILT - Wageningen University & Research Centre, P.O. Box 230, 6700 AE, Wageningen, The Netherlands

To study the effects of NPs, often only one cell type is used, raising the question whether these results can be extrapolated to other cell types. Nowadays large amounts of data linking genetic information and molecular pathways are available (i.e. connectivity map). To exploit this, we compared the effects of silver (Ag)NPs (*i.e.* 20, 30, 60, and 110 nm) and silver nitrate on the transcriptome of Caco-2 intestinal and MCF-7 breast cancer cells. Cells were exposed for 6 and 24h at a non-cytotoxic concentration of 25 µg/ml. This resulted in pronounced effects on gene expression in both cell lines. Principle component analysis showed distinct classification of MCF-7 and Caco-2 cells. Effects in MCF-7 cells were size-dependent with the smallest AgNPs affecting the highest number of differentially regulated genes. In MCF-7 cells, 20 and 30 nm AgNPs affected a larger number genes than $AgNO_3$, while Caco-2 cells were most sensitive to $AgNO_3$ exposure. In MCF-7 cells, AgNPs affected more genes after 24h than after 6h, while the reverse was the case in Caco-2 cells. In contrast, $AgNO_3$ induced more genes after 6 h than after 24 h in both cell lines. Pathway analysis (using MetaCore software) revealed that similar processes were affected in the two cell lines. These processes included stress responses, apoptosis, immune responses, and negative regulation of the cell cycle. To support the outcome of the gene expression experiment we quantified the uptake of silver (NPs) by ICP-MS and TEM analysis and assessed affected pathways by functional cell imaging.