

SameSpots







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SameSpots Case Study

Background Information

"Our current work in the Protein and Gene Biomarkers Unit is investigating the characterisation of changes in the proteome profile induced by environmental toxic compounds, with the aim of identifying critical targets and pathways in biological systems that are affected by, and respond to, adverse chemical and environmental exposure."

"We have been using SameSpots in our research since January 2007. The software has drastically simplified our proteomics workflow and opens up new avenues for the exploration of the data obtained."

Our 2D experiment design

"Our current 2D experiment is a time course study over three time points comparing two treatments which are two doses of the same compound. We use 16cm x 24cm gels and stain with Colloidal Coomassie®. Our experiment set up includes four replicates at each time point - we don't require biological replicates as the samples are from the same homogenous cell population. Consequently each of our experiments has a total of 36 gel images."

The challenges we face in our 2D image analysis

"With at least 1500 visible spots per gel, our images are very challenging to analyse! Before we introduced SameSpots into our workflow, we were spending 4-6 hours per gel for an expert user which meant 3-4 weeks work to perform image analysis for each experiment. This didn't include any statistical analysis which was an additional challenge. Univariate statistical analysis was not a correct approach due to our experimental design, but the use of multivariate analysis was compromised due to the problem of missing values introduced during the analysis."

How SameSpots helps us to meet the challenges?

"Since using SameSpots the speed of our analysis has been drastically improved. We align our gels accurately within 5 mins per gel and then run the SameSpots analysis. We don't usually perform any correction of spot detection which results in fast, robust and very reproducible analysis. This has an immediate positive effect on the quality and confidence in our results."

"Statistical tools included allow a rapid analysis of the experimental groupings as intra- and inter-group variability is easily visualised. As a result we can redefine the whole workflow and make corrections to our experiment in a realistic timeframe, for example add new gels or more replicates." "The SameSpots approach also produces data with no missing values, so we can apply PCA and cluster analysis without having to export to any other statistical package. It provides us with the complete solution. The software is extremely user-friendly and can be used by any researcher with a basic knowledge of the main statistical techniques."

"The analysis of our 36 gel experiments is now reduced to a few hours of work to get a first evaluation of the results. Such is the performance of SameSpots we have also been able to retrospectively analyse gel images that had previously proved too difficult to analyse!"

"More importantly, our research is now so much faster allowing us to include more replicates into our experimental design, which means we are producing statistically superior results. We are confident we are finding real changes in our gels."

Future plans

"We have ambitious plans for our future 2D experimental design. We can finalise several 2D projects in parallel and spend less time to find significant changes in proteome profiles that can be sent for subsequent validation using additional techniques. SameSpots has simplified our proteomics research as the software is easy to use and the technical support from the TotalLab team is excellent."

About the Istituto di Ricerche Farmacologiche "Mario Negri"

Founded in 1963, The Istituto di Ricerche Farmacologiche "Mario Negri" is a non-profit independent scientific organisation for biomedical research and education.

"In the Environmental Health Sciences Department we investigate the effect of environmental factors on human health, focussing on the survey of environmental contaminants, the assessment of human exposure with related health risks and the toxicity mechanisms of pollutants."

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