# The optimization of a laboratory protocol through Design of Experiment statistical methodology

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The Design of Experiment (DoE) is a statistical methodology, very useful in the case of multivariable assays, that permits to evaluate simultaneously the influence of different factors on a specific output and to analyse the interactions among them in order to identify their optimal combinations. Moreover, DoE allows to select from the huge numbers of combinations only a limited number, in order to cover the whole frame, allowing to save both time and money. DoE was mostly used in industrial field in order to maximize robust processes, but recently it has been also applied in biomedical research field. Different studies have demonstrated the advantages of using a DoE approach compared to the classical one (a single parameter is tested in each assay) in the context of automated experiments, determination of cell media [1] compositions or HPLC tuning [2].

In the present study, we utilize for the first time the DoE methodology to optimize the transfection protocol of neural cells, as an example of DoE application to a laboratory procedure. Neural precursor cells are hard to transfect and refractory to lipidic reagents [3], for this reason we choose as transfectant reagent the cationic not-lipidic Poliethylenimmine (PEI). The DoE approach allowed us to identify the main variables (factors) affecting the transfection efficiency and to discover their optimal combinations, developing a protocol that let us to triplicate the transfection efficiency respect to the initial conditions. Moreover, the covariance analysis unmasked significant variable interactions impossible to calculate in one factor-variation tests used in normal laboratory practices. Our results indicate that DoE might be very helpful also in research for the identification of the better experimental conditions and the analysis of interactions between different variables.

*References*

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