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Solving the dilemma of high variability in NADPH oxidase activity assessment

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NADPH Oxidase (NOX) activity has been suggested to be implicated in a plethora of cellular functions contributing to physiological and pathological conditions. Precise determination of NOX activity is not trivial. Several studies have addressed NOX activity in relation to polymorphisms in cis-acting genetic elements, however, providing much conflicting results. To date, no genetic polymorphism pertinent to one of the NOX subunits has been established as biomarker for NOX activity. We set up to determine activity of the phagocytic NOX by a particular sensitive assay in Lymphoblastoid Cell Lines (LCLs) allowing iterative measurements on the same genetic background. A set of 290 LCLs of Caucasian origin with available genotypic data was split into training and test set by a 2:1 ratio. NOX activity determination was performed initially four times in each LCL at different days each with four same LCLs serving as reference for inter-day comparisons. Stringent criteria to identify outliers of the measurement series were applied to obtain a robust value of the NOX activity for each LCL. If these criteria were not met with at least three independent measurements two to four additional repetitions were conducted. If again applied stringent criteria fail, robustness of repetitive NOX activity measurements, the respective LCL was excluded from further analyses. Measurement series of LCLs which matched the consistency criteria elicited a mean variation coefficient of 17.0% (SD±10.6%). An almost identical distribution of the determined NOX activity was noted for the training and the test set of LCLs. These data prove our method suitable for reliable assessment of NOX activity providing a reliable basis for pending association testing with genotypes. As with 290 LCLs a total of 1500 NOX activity measurements, each time assayed in quadruplicates, were performed, this study is the largest conducted in this context so far.

Biography

Tana Takacova has completed her Medical degree at the Commenius University in Bratislava, Slovakia, as well as the Georg-August University in Goettingen and Heidelberg University School of Medicine in Germany. In addition, she frequently gave lectures of Molecular Medicine as an Honorary Guest Student at the Georg-August University in Goettingen. Her research interests involve the role of tumor microenvironment and cancer immunotherapy. Currently, she pursues her Internship in the Department of Haematology, Oncology and Clinical Immunology at the Heinrich Heine University in Dusseldorf, Germany.

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