Phenotyping Cystic Fibrosis Cells by microFTIR and Principal Component Analysis

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Background. The autosomal recessive Cystic fibrosis (CF) disease is caused by thousands different mutations in the Cystic Fibrosis Transmembrane conductance Regulator (CFTR) gene which codifies for CFTR protein transporting chloride through the plasma membrane of secreting epithelia. CF patients homozygous for a CFTR defect can develop severe multi-organ disease. CFTR-defective epithelial cells can be classified by Fourier Transform InfraRed microspectroscopy (microFTIR) and explorative PCA [1]. **Experimental design**. We applied this approach to classify cells from two monozygotic CF twins with a rare genotype (2183delAAinsG associated with the genetic variant TG15/T5 *in trans*) but with very discordant phenotypes [2]. Cells from their healthy mother bearing the TG15/T5 genetic variant and from two healthy controls with a non-defective wild type CFTR genotype were also considered. Lympho-monocytes of CF patients and controls were immortalized with the Epstein Barr Virus to obtain lymphoblastoid cells (LCs). CFTR expression and functions were assessed by complementary approaches [3-5] and exposing LCs cells to the CFTR corrector VRT-809 alone or in combination with the CFTR potentiator VRT-770. Independent blind experiments were carried out to cross-validate the results of IR analysis. **Results and conclusions**. Experimental evidence was achieved that microFTIR and PCA classified LCs with the different CFTR genotypes as well as distinguished the LCs cells of the twins with different phenotypes. In particular, the spectra of LCs from the two CF twins grouped separately. LCs of twin with more favourable CF phenotype preferentially grouped with LCs of the mother and was sensitive to VRT-802 correction, as suggested by their classification close to LCs healthy controls. These data were confirmed by those of methods assessing CFTR functionality. In conclusion, unsupervised explorative multivariate data analysis performed by PCA and/or HCA techniques in both dataset of SR FTIR of single LCs cells and average FTIR spectra acquired from several LCs cells probed with globar as IR source are useful screening approach to classify different CF phenotypes. Probing LCs cells exposed to CFTR correctors and potentiatiors with microFTIR represent a rapid and convenient approach to test drugs directly in cells expressing the patient's geno/phenotype.

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