The role of Glucocerebrosidase GBA2 in cystic fibrosis lung inflammation: from molecular mechanism to therapeutic strategies

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Background. Several studies indicate that sphingolipids (SL) play a regulatory role in airway inflammation, the most critical aspect of CF lung disease. Recently, Ceramide (Cer) derived from glycosphingolipids (GSL) has gained more interest since inhibition of the glucocerebrosidase GBA2 and its down-regulation by siRNA, are associated with a significant reduction of IL-8 after P.aeruginosa (PAO) infection, as well as a reduction of the intrinsic inflammatory state in CF human epithelial bronchial cells (CFhEBC).

Hypothesis and Objectives. In our work hypothesis, the aberrant inflammatory response to PAO in CFhEBC starts from alterations in the lipid composition of specific plasma membrane (PM) macromolecular complex by the action of the GSL-hydrolases associated with the cell surface, including GBA2. To figure out the possible molecular mechanism we investigated the effect of PAO infection on specialized membrane area called lipids rafts. Moreover, with the aim to develop a new anti-inflammatory treatment we developed new lipid based-nanoparticles for the delivery of siRNA targeting GBA2.

Methods. In CFhEBC and control cells we evaluated the effect of PAO infection on: i) SL hydrolases activities, ii) SL pattern, and iii) lipid rafts organization. The lipid-based nanoparticles used for GBA2 silencing were characterized both for their biophysical properties and GBA2 knocking down efficacy.

Results. The data obtained further support the role of GBA2 in the inflammatory response upon PAO infection. Moreover, we found that in CFhEBC PAO causes a recruitment of PM-associated GSL-hydrolases into lipids rafts. At this site, the enrichment of enzymes involved in GSL catabolism causes a reduction of the ganglioside GM1 and sphingomyelin, which is paralleled by increased levels of GlucosylCer and Cer both events responsible for the activation of the inflammatory response. In addition we developed lipid-based non viral nanovectors (NP) able to silencing GBA2 in vitro for up 8 days with any toxicity for the cells. By X-ray analysis, we identified the most promising NP structure in order to penetrate CF mucus.

Spin-off for research & clinical purposes. The identification of GBA2 as well as other GSL-hydrolases as possible molecular targets related to the inflammatory response and the development of an innovative NP-based approach for their silencing could represent an important amelioration of the therapeutic strategies in CF.

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Reference:

1. Unravelling the role of sphingolipids in cystic fibrosis lung disease., Aureli M, Schiumarini D, Loberto N, Bassi R, Tamanini A, Mancini G, Tironi M, Munari S, Cabrini G, Dechecchi MC, Sonnino S. Chem Phys Lipids. 2016;200:94-103.PMid:27592248

2. GBA2-encoded β-glucosidase activity is involved in the inflammatory response to Pseudomonas aeruginosa. Loberto N, Tebon M, Lampronti I, Marchetti N, Aureli M, Bassi R, Giri MG, Bezzerri V, Lovato V, Cantù C, Munari S, Cheng SH, Cavazzini A, Gambari R, Sonnino S, Cabrini G, Dechecchi MC. PLoS One. 2014 Aug 20;9(8):e104763 PMid:25141135 PMCid:PMC4139313