

Evaluation of the biological and therapeutic properties of mesoangioblasts-vessel associated progenitor cells- in the cell based therapy of the cystic fibrosis disease

Vezzali C¹, Celesti G¹, Bonfanti C¹, Antonini S¹, Barone C², Pesce E⁴, Tomati V⁴, Maiuri L³, Egan M², Pedemonte N⁴, Bruscia E², Messina G¹

¹University of Milan, Dept. of Biosciences, Milan, Italy; ²Yale University, School of Medicine, New Haven (CT), USA; ³IERFC, Ospedale San Raffaele, Milan, Italy; ⁴Gaslini Institute, Genova, Italy. (Grant No. FFC#6/2015) [[doi.org/bwjv](https://doi.org/10.1007/978-88-470-2111-1_10)]

Keywords: Cystic Fibrosis, Mesoangioblast progenitor cells, Cell-based Therapy

Background. Since the CFTR gene was cloned in 1989, several strategies for correction of CF lung disease have been explored. Among these, cell-based approaches are under investigation. Endogenous lung stem and progenitor cells have been studied although their contribution in the amelioration of chronic lung diseases is still debated. Stem cell-based approaches to treat CF have not achieved the efficiencies of delivery and engraftment needed for therapy. The reasons rely in the low cell engraftment in the lungs after systemic administration, in the small percentage of differentiated cells in airway epithelia and in the even less percentage of CFTR expression.

Hypothesis and objectives. During our recent works on a class of mouse progenitor cell derived from vessel, named mesoangioblasts4 (mMABs), we observed that mMABs, when systemically transplanted in healthy mice distribute throughout lung, trachea and intestinal epithelium for up to 2 months. The major aim of this study was the evaluation overtime of the engraftment of adult mMABs transplanted in CF mice, in terms of percentage of engraftment and persistence in time. As a parallel purpose, mMABs have been studied in terms of their ability to differentiate in epithelial cells and to express functional CFTR in transplanted CF mice, thus resulting in a general amelioration of the pathology. The whole study aimed to develop a cell therapy approach for patients affected by CF.

Methods. The therapeutic properties of mMABs were tested in F508del-CFTR and KOCftrtm1UNC mouse models. Functional rescue of CFTR was evaluated in vivo by measuring the nasal potential difference across the nasal epithelium (NPD) and ex vivo at intestinal level, by Ussing chamber. Inflammatory status after MAB transplantation was investigated in CF mice by

ELISA and qRT-PCR. Moreover, MAB capability to differentiate in different epithelial cells was studied, both in vitro and in vivo, by immunofluorescence and qRT-PCR. Human derived MABs were also tested for CFTR expression and activity, by WB and Ussing chamber, this latter in co-culture with CFTR-mutated Human Bronchial Epithelial cells (HBE).

Results. We observed that mMABs engraft lung, tracheal and intestinal epithelium for up to 6 months in F508del CFTR mice after a single systemic injection. We also verified that mMABs are able even in vitro to potentially express a functional CFTR channel. Most importantly, in vivo, mMAB transplanted KOCftrtm1UNC mice express a functional CFTR. Notably, when transplanted and engrafted in the epithelium, mMAB express typical epithelial markers, thus demonstrating mMAB ability to differentiate in epithelial cells. Finally, the first studies on human MABs demonstrated their ability to express a functional channel.

Spin-off for research & clinical purposes. This project confirmed the therapeutic value of MABs for the development of an efficacious in vivo cell therapy for the cure of CF.

Acknowledgment. FFC#6/2015: funded by FFC, supported by Delegazione FFC di Lucca, Delegazione FFC di Lecce, Gruppo di Sostegno FFC di Tremestieri, Gruppo di Sostegno FFC di Magenta Milano, Delegazione FFC di Fermo, Delegazione FFC di Olbia Tempo.

References

1. Riordan JR, Rommens JM, Kerem B, et al. Identification of the cystic fibrosis gene: cloning and characterization of complementary DNA. *Science* 1989;245:1066-73.

<https://doi.org/10.1126/science.2475911>

PMid:2475911

2. Vaughan AE, Brumwell AN, Xi Y, et al. Lineage-negative progenitors mobilize to regenerate lung epithelium after major injury. *Nature* 2015;517:621-5.

<https://doi.org/10.1038/nature14112>

PMid:25533958 PMCID:PMC4312207

3. Zuo W, Zhang T, Wu DZ, et al. p63(+)Krt5(+) distal airway stem cells are essential for lung regeneration. *Nature* 2015;517:616-20.

<https://doi.org/10.1038/nature13903>

PMid:25383540

4. Diaz-Manera J, Touvier T, Dellavalle A, et al. Partial dysferlin reconstitution by adult murine mesoangioblasts is sufficient for full functional recovery in a murine model of dysferlinopathy. *Cell Death Dis* 2010;1:e61.

<https://doi.org/10.1038/cddis.2010.35>

PMid:21364666 PMCID:PMC3032521

5. French PJ, van Doorninck JH, Peters RH, et al. A delta F508 mutation in mouse cystic fibrosis transmembrane conductance regulator results in a temperature-sensitive processing defect in vivo. *J Clin Invest* 1996;98:1304-12.

<https://doi.org/10.1172/JCI118917>

PMid:8823295 PMCID:PMC507556

6. Clarke LL, Grubb BR, Gabriel SE, et al. Defective epithelial chloride transport in a genotargeted mouse model of cystic fibrosis. *Science* 1992;257:1125-8.

<https://doi.org/10.1126/science.257.5073.1125>

PMid:1380724