**PROGETTO**

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| **Supervisore:**: | Andrea Annoni |
| Titolo/*Title*: | Definizione di una terapia genica diretta al timo per controllare lo sviluppo di cellule T allo-reattive e promuovere l'accettazione di trapianto allogenico / *Engineering Thymic Selection to Control the Development of Alloreactive T Cells to promote acceptance of allogeneic transplantation.* |
| *Curriculum*: | Terapia genica e cellulare |
| Link alla pagina personale del sito web di Ateneo o del polo ospedaliero di riferimento: | https://research.hsr.it/en/institutes/san-raffaele-telethon-institute-for-gene-therapy/immune-core/andrea-annoni.html |

**Descrizione del progetto (max 3.000 caratteri spazi inclusi)**

Transplantation represents a valid therapeutic option in case of organ failure but also for reconstructive purposes, although the required lifelong multi-drug immunosuppressive therapy counterbalances its benefits. Minimization or even avoidance immunosuppression via immune modulatory regimens would be the key to a wider applicability of vascularized composite allotransplantation (VCA).

The only clinically successful avenue of transplant tolerance induction to date has been regimens that induce hematopoietic chimerism via stem cell transplantation (HSCT)(1). The success of these strategies relies on the colonization of the recipient thymus by donor bone marrow derived dendritic cells (DC). In the thymus a population of stromal cells named thymic epithelial cells (TEC) works in concert with DC to select a T repertoire tolerant to self antigens but capable to defend the organism(2). However, TEC are not hematopoietic cells and donor TEC do not develop following HSCT-based tolerance protocols. Therefore, an ideal immunotherapy would promote the co-expression of donor and recipient MHC in the thymus – a condition we refer to as a "hybrid thymus". We have well validated capacity and technologies to impose in the thymus a stable expression of donor MHC by a LV platform for gene transfer (3). In this scenario we hypothesized that reengineer the thymic environment to achieve lasting expression of donor and recipient MHC molecules, in concert with DC, originating from the bone marrow niche of a VCA, will promote passive and active protection of transplanted tissues.

To demonstrate the therapeutic potential of a hybrid thymus generated via gene transfer, we will test whether LV-driven expression of donor-related MHC molecules will generate a hybrid thymus capable of reshaping the T cell repertoire. We will first determine whether MHC expression in mice defective of endogenous MHC expression restores positive and negative selection of developing thymocytes. Then we will define the capacity of LV-engineered thymus to contribute to the selection in a fully competent thymus and to synergize with donor bone marrow derived DC to promote VCA survival.

**Competenze che deve acquisire lo studente** (Max 600 caratteri spazi inclusi):

- capacity to plan and perform in vivo experiments to accomplish project's aims;

- capacity to plan and perform specific immunoassay (mixed leucocyte reaction (MLR), proliferation, elispot, cytokine determination) to answer to experimental questions;

- capacity to define proper controls for each experimental procedure;

- generate and analyze multiparametric cytofluorimetric data;

- capacity to interrogate the scientific literature to improve his knowledge and experimental creativity

- write scientific reports

- Present and discuss generated data in the context of internal lab-meetings and scientific meetings.

**Bibliografia** (max. 15)

1. Sachs, D. H., Sykes, M., Kawai, T. & Cosimi, A. B. Immuno-intervention for the induction of transplantation tolerance through mixed chimerism. Semin Immunol 23, 165-173, 2011

2. Klein, L., Hinterberger, M., Wirnsberger, G. & Kyewski, B. Antigen presentation in the thymus for positive selection and central tolerance induction. Nat Rev Immunol 9, 833-844, 2009

3. Russo F, Ruggiero E, Curto R, Passeri L, Sanvito F, Bortolomai I, Villa A, Gregori S, Annoni A. Editing T cell repertoire by thymic epithelial cell-directed gene transfer abrogates risk of type 1 diabetes development. Molecular Therapy Methods Clin Dev. 2022