Name: GREGORI SILVIA

Unit: MECCANISMI DI TOLLERANZA PERIFERICA

Position:

☐ Professor at Vita-Salute San Raffaele University

☐ Researcher at Vita-Salute San Raffaele University

X Group Leader at IRCCS San Raffaele Hospital

☐ Project Leader at the IRCCS San Raffaele Hospital

☐ Other _____

I am already a DoS X I am applying as a new DoS (CV in attachment) ☐

PROJECT 1

Title: **Tolerogenic approaches to prevent unwanted immune responses in protein replacement and gene therapies for Lysosomal Storage Disorders.**

PhD Curriculum:

☐ Basic and Applied Immunology and Oncology

☐ Cellular and Molecular Biology

☐ Experimental and Clinical Medicine

☐ Neuroscience and Experimental Neurology

X Gene and Cell Therapy

The Study will be based on:

X Basic research activity

☐ Clinical research activity

☐ Clinical research activity that includes interaction with the patients

The Study will involve:

☐ basic oncological research

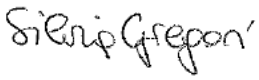
☐ clinical oncological research

PROJECTS IN ATTACHMENT (please include your link to the properly updated OSR website)

I declare that:

X the PhD student will be first author on the main publication originating from his/her project and on all publications (including after graduation) that are mostly based on his/her experimental work.

X in the case a student is not an awardee of a UniSR fellowship (i.e. he/she has won a position without fellowship: "posizione senza borsa"), I am prepared to cover the fellowship of the student. I am aware that the amount of the fellowship cannot be less than the minimum required by Law (Ministerial Decree 40, January 25th 2018): 15.343,28 € gross per year, per three years.

Signature  **Date** 15/04/2021

When it applies:

Group Leader Dr./Prof _____

Signature _____ **Date**_____

PROJECT 1

DoS: GREGORI SILVIA

Title: **Tolerogenic approaches to prevent unwanted immune responses in protein replacement and gene therapies for Lysosomal Storage Disorders.**

Curriculum: GCT

Link to OSR/UniSR personal page:
https://research.hsr.it/en/institutes/san-raffaele-telethon-institute-for-gene-therapy/mechanisms-of-peripheral-tolerance/silvia-gregori.html

Project description (*Number of characters*, *including spaces*: 2.000 - 3.000):
Lysosomal storage diseases (LSDs) are a group of inherited metabolic disorders caused by enzyme deficiencies within the lysosome resulting in accumulation of undegraded substrate. This storage process leads to a broad spectrum of clinical manifestations depending on the specific substrate and site of accumulation. These disorders are progressive, although the rate of progression is variable, and the advent of novel therapies has changed the natural history of the disease in some cases. Enzyme replacement and gene therapy (ERT and GT) for LSDs have been shown to be safe and effective in pre-clinical and clinical studies. Although these approaches represent major advancements in the treatment of LSDs, their application have revealed several challenges. Due to individual patient genetics, ERT or viral vectors encoding the enzyme can lead to detrimental immune responses that attenuate treatment benefits and can impact patients' safety. Up to date, the mechanisms underlying immune response towards infused recombinant enzymes and or vector-derived transgenic enzymes have been characterized and T and B cell modulation has been shown to be effective in preventing infusion associated reactions in patients under ERT and in pre-clinical models of GT.

Approaches designed to selectively control unwanted antigen(Ag)-specific immune responses and promote tolerance such as *in vivo* induction of Ag-specific regulatory cells or CAR regulatory cell therapy represent innovative tools for the management of several pathologic conditions, including detrimental responses to ERT and transgenes. We aim at applying these strategies to control immune responses towards the missing enzyme in the context of ERT and GT for LSDs, including Pompe and Mucopolysaccharidosis. To this end we will: i) characterize the immune responses to the missing enzyme evocated by ERT or GT in murine models and, possibly, in patients focusing on identification of the immunodominant regions of the target enzyme, ii) design enzyme specific CAR molecules, iii) generate genetically engineered tolerogenic cells (Tol-DC or CAR-Tregs) to control immune responses to the replaced enzyme *in vitro* and *in vivo* in preclinical models of the disease.

Results from these studies will provide new insights into the mechanisms underlying immune response to the missing enzyme in patients with LSDs and will be instrumental to improve current therapies and to the shaping of innovative approaches for the cure of Pompe disease and other LSDs, such assert and *in vivo* gene therapy, in which pre-existing immunity to the transgene could hamper stable enzymatic activity.

Skills to be acquired by the student:

- Generation of lentiviral vectors
- Protocol for induction of tolerogenic DC and T cells in vitro
- Generation of engineered murine BMDC
- Multiparametric flow cytometric analysis
- *In vitro* proliferative and suppressive assays and cytokine production profiles

References (max. 3)