Universidad de Navarra
Facultad de Ciencias

MASTER'S DEGREE IN BIOMEDICAL RESEARCH
Research Project Proposal
Academic year 2023-2024

Project Nº 25 ASIGNADO

Title: Validation of relevant factors influencing CAR T cells activity using CRISPR/Cas9-based genome editing tools.

Department/ Laboratory
Laboratory 104, Adoptive Cell Therapy, Hemato-Oncology Program, CIMA Universidad de Navarra.

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Summary
In the last decade, adoptive cell therapy strategies based on T lymphocytes engineered with chimeric antigen receptors (CAR T cells) have evolved to become a real therapeutic option for patients with certain B cell malignancies, like acute lymphoblastic leukemia and some types of lymphoma. However, CAR T therapies still present several limitations for other haematological malignancies, such as multiple myeloma (MM), where the lack of long-term efficacy and treatment associated toxicities compromise the therapeutic efficacy. Moreover, the molecular mechanisms promoting CAR T cell expansion and persistence are not yet fully understood.

Recent gene editing technologies, such as CRISPR systems, have transformed the biological approach to understanding the molecular mechanisms associated with different biological processes. Combining this technology with novel sequencing methods, our group has already identified several factors influencing CAR T cell function. Thus, the **main objective** of this project is to **modulate relevant factors** in CAR T cells, by using **CRISPR technologies,** and **evaluate** the effect of their modulation in the **functionality of** CRA T therapies.

In this project we will generate primary human CAR T cells deficient in each of the selected factors using CRISPR systems, that will be characterized both *in vitro* and *in vivo* against tumoral cells. *In vitro* lytic capacity of CAR T cells will be measured by standard cytotoxic assays using a luminescence-based method. Cytokine production will be measured by ELISA and phenotypic analysis will be performed by flow cytometry. Finally, the *in vivo* antitumor efficacy of CAR T cells will be evaluated in MM xenograft models generated in immunodeficient NSG mice inoculated with MM cell lines expressing GFP-Luc.

yes	X	Does the project include the possibility of supervised animal manipulation to complete the training for animal manipulator?
no		