



MÁSTER EN INVESTIGACIÓN BIOMÉDICA
Research Project Proposal
Academic year 2026-2027

Project Nº 23

Title: Genome-wide CRISPR screening to identify neutrophil host factors required for defense against Acinetobacter baumannii

Department/ Laboratory Department of Microbiology and Parasitology-Edificio de Investigación

Director 1 Nahuel Agustin Riviere

Contact: nriviere@unav.es

Codirector: Tomás Maira Litrán

Contact: tmairalitra@unav.es

Summary

Goals: This project aims to establish a genome-wide CRISPR screening platform in neutrophil-like HL-60 cells to identify host genes involved in the interaction between neutrophils and Acinetobacter baumannii, a major multidrug-resistant pathogen responsible for severe hospital-acquired infections. The main objective is to use CRISPR technology to systematically disrupt human genes and identify host factors that regulate bacterial phagocytosis, intracellular killing, and potential bacterial survival within neutrophils.

Methodology: The student will first generate stable Cas9-expressing HL-60 cells through lentiviral transduction and validate Cas9 activity. These cells will then be transduced with the Brunello human genome-wide CRISPR knockout library, enabling genome-wide loss-of-function screening across thousands of human genes.

Following antibiotic selection, HL-60 cells will be differentiated into neutrophil-like cells and infected with fluorescently labeled A. baumannii. Flow cytometry will be used to isolate cell populations showing altered bacterial uptake or intracellular survival phenotypes. Additional validation experiments will include gentamicin protection assays, intracellular survival assays, and fluorescence microscopy to confirm bacterial persistence and host-cell interactions.

Finally, enriched sgRNAs will be identified by next-generation sequencing to uncover host genes and pathways involved in neutrophil defense against A. baumannii. This project offers students hands-on experience in CRISPR functional genomics, host-pathogen interactions, and antimicrobial research.

Table with 2 columns and 2 rows: yes, no, X

Does the project include the possibility of supervised animal manipulation to complete the training for animal manipulator?