Pump-Priming Grant

Report

Oliver McCallion

Title

Novel gene-edited universal regulatory T cells for transplant tolerance

Objective

To overcome allorecognition of regulatory T cells from an unrelated third-party donor through modulation of HLA expression in order to evaluate their feasibility and efficacy for induction of transplant tolerance.

Method

Using a novel adenine base editor and CRISPR/Cas9 we generated hypoimmune Treg without expression of HLA class 1 and HLA class 2. In order to overcome 'missing self' NK recognition we knocked in the gene encoding the monomorphic non-classical HLA class 1 related protein HLA-E.

Results

We undertook extensive *in vitro* characterisation and evaluation of the edited Treg to demonstrate retained phenotypic stability, suppressive function, and capacity to avoid NK-mediated cell lysis. Using two animal models of transplant rejection—one short term and one long term—we demonstrated that edited third party alloTreg afforded comparable levels of graft protection to autologous Treg. We evaluated the transcriptional effect that edited alloTreg induced within the skin grafts demonstrating that whilst unedited third party Treg grafts exhibited clear cytotoxic inflammatory rejection signatures, grafts from animals treated with edited alloTreg expressed signatures associated with tissue maintenance, lipid presentation, and homeostasis.

Outputs (publications/presentations)

These results are available at the preprint server bioRxiv (https://doi.org/10.1101/2023.08.06.551956) and have been provisionally accepted for publication in *Nature Communications*. The results are planned to be presented at upcoming conferences.

Next Steps

This work has generated important proof-of-concept data supporting the feasibility and efficacy of the approach. Next steps include further optimising the editing process to remove the need for two editing steps and further efficacy testing. The data described above, generated with the generous support of the Oxford Transplant Foundation pump priming grant, will be integral to applications for project funding.