## **OP-04**

## Effect of Golgi Stress on CXCR5+ HLA- G+ Expression in T Follicular Helper Cells

<u>Miray Kavuzlu</u>1, Bilkay Baştürk<sup>2</sup>, Özgür Kütük<sup>3</sup>, Belgin Ataç<sup>1</sup>, Kenan Çalışkan<sup>4</sup>

<sup>1</sup>Başkent University Faculty of Medicine, Department of Medical Biology Ankara, Turkey

<sup>2</sup>Başkent University Faculty of Medicine, Department of Immunology Ankara, Turkey

<sup>3</sup>Sabancı University Faculty of Engineering and Natural Sciences, İstanbul, Turkey

<sup>4</sup>Başkent University Faculty of Medicine, Department of General Surgery Ankara, Turkey

**Objectives:** The aim of study was to investigate the effect of Golgi stress induced in follicular B-cells and steroid treatment on human leukocyte antigen (HLA)- G expression in CXCR5- expressing T follicular helper (Tfh) cells.

**Materials and Methods:** T- and B-cells isolated by negative selection from spleen samples of 3 different volunteers were used in the study. Golgi stress was induced by treatment of B-cells with brefeldin A (BFA) and steroids were administered by T:B co-culture. For the effect of BFA and steroids, the time-dependent (0/6/12 hours) change in CXCR5+ HLA- G+ expression in Tfh cells was evaluated by flowcytometry method.

**Results:** Golgi stress induced in follicular B-cells significantly increased CXCR5+ HLA- G+ expression on Tfh cell surface in a time-dependent manner. HLA- G expression on the CXCR5+ Tfh cell surface was found to increase in a time-dependent manner with steroid treatment. In the presence of BFA, HLA- G expression on the CXCR5+ Tfh cell surface was found to increase in a time-dependent manner with steroid effect.

**Conclusion:** HLA- G is a non-classical class-I major histocompatibility complex molecule that plays a role in modulating the immune system. Expression of the HLA- G molecule on the CXCR5+ Tfh cell surface, which shows the chemotactic effect of T follicular helper cells, is increased. The increased expression of HLA-G on the surface of CXCR5+ Tfh cells indicates that the immunomodulatory effects of these cells increase depending on the relationship of these cells with B-cells under the effect of BFA and time.

## **OP-05**

## Using Machine Learning to Examine Pre-Transplant Factors Influencing *De Novo* HLA-Specific Antibody Development Post-Kidney Transplant

Alex Rothwell<sup>1</sup>, <u>George Nita<sup>2,3</sup></u>, Matthew Howes<sup>3</sup>, Dan Ridgway<sup>3</sup>, Abdul Hammad<sup>3</sup>, Sanjay Mehra<sup>3</sup>, Andrew R. Jones<sup>2</sup>, Petra Goldsmith<sup>3</sup>

<sup>1</sup>University of Liverpool Computational Biology Facility, Liverpool Shared Research Facilities, Liverpool, United Kingdom

<sup>2</sup>University of Liverpool Institute of Systems, Molecular and Integrative Biology, Liverpool, United Kingdom

<sup>3</sup>Liverpool University Hospitals NHS Foundation Trust, Royal Liverpool University Hospital, Department of Renal Transplantation, Liverpool, United Kingdom

**Objective:** The development of *de novo* donor-specific antibodies (dnDSA) against human leukocyte antigens (HLA) is linked to premature graft failure in kidney transplantation. However, rates and influencing factors of *de novo* DSA formation vary widely in existing literature. This study aims to identify pre-transplant factors affecting *de novo* HLA-specific antibody development post-transplantation using machine learning models.

**Materials and Methods:** Data from 460 kidney transplant recipients at a single center between 2009-2014 was analysed. Pre-transplant variables were collected, and post-transplant sera were screened for HLA antibodies. Positive samples underwent further investigation with single antigen bead testing. Machine learning models, including CART, Random Forest, XGBoost, and CatBoost, were trained on pre-transplant data to predict dnDSA formation. Models were evaluated with and without SMOTE oversampling, using F1 scores for performance and SHAP for feature importance.

**Results:** In the full cohort analysis, XGBoost models performed the best, achieving the highest F1 scores (0.54-0.59 without SMOTE; 0.72-0.79 with SMOTE). The strongest predictors were pre-transplant HLA antibodies, number of previous kidney transplants, cold ischemia time (CIT), recipient age, and female gender. Pre-existing HLA antibodies and past transplants increase the risk of dnDSA development. Notably, extreme CIT durations and older age (over 65) link to a lower predicted probability of dnDSA. In the unsensitised cohort, models had poor predictive power.

**Conclusion:** Machine learning models can identify pre-transplant risk factors for *de novo* DSA development in kidney transplantation. Monitoring and stratifying patients based on these factors may guide preventive immunological strategies and recipient selection, potentially improving long-term allograft outcomes.