Oral Presentations

OP-01

The Use of Donor-Derived Cell Free DNA in Post-Kidney Transplant Monitoring

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Kidney transplantation is the optimal treatment for end stage kidney disease. However, rejection episodes remain a main cause of reduced graft survival and graft biopsy is the gold standard of the diagnosis.

Consequently, the need of a non-invasive and early detectable biomarker of rejection has recently led to the study of donor-derived cell free DNA (dd-cfDNA).

Thirty patients who underwent kidney transplantation (16 living and 14 deceased) from October 2023 until July 2024, were randomly selected for the dd-cfDNA quantification with the AlloSure test. Measurements of dd-cfDNA from patients' plasma took place 1 month, 3 months and 6 months post transplantation and in some cases before a "for cause" graft biopsy.

The preliminary results of dd-cfDNA measurements were combined with established laboratory markers used to diagnose rejection (serum creatinine, proteinuria, DSAs) and the tacrolimus level at the time of the measurement. Elevated values (equal or above the threshold of 0.5%) were noticed in 16/30 samples at 1 month after transplantation and correlated with T-cell-mediated rejection, acute cell-mediated rejection, mixed rejection and graft injury of other causes. Twenty-two patients of the sample underwent a second measurement at 3 months and 4 of them had a third measurement at 6 months. Also, in cases of "for cause" graft biopsy the results were associated with the dd-cfDNA value.

Dd-cfDNA is a new promising biomarker that contributes in the early detection of rejection. Therefore, threshold values and surveillance strategies in kidney transplant patients need to be established by further studies.

OP-02

Time-Dependent Change of $CD4^+$ and $CD8^+$ T-Cell Ratio in the Presence of Donor-Specific Antibody

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Abstract: In the context of cell, organ, and tissue transplants, the functionality of the graft tissue is a critical factor in determining survival outcomes. The cellular and secretory components of both the innate and adaptive immune systems initiate a rejection response against graft tissue that is recognized as foreign. $CD4^+$ T-cells have been shown to participate in the inflammatory response by secreting cytokines. In contrast, $CD8^+$ T-cells become effectors and kill target cells by exerting a cytolytic effect. B lymphocytes activate the complement system with donor-specific antibodies against human leukocyte antigen (HLA) structures of the donor that they recognize as foreign.

Introduction: Anti-donor HLA antibodies can be detected de novo after cell, tissue and organ transplantation and play an important role in long-term survival. *De novo* donor-specific antibodies (dnDSAs) are the cause of antibody-mediated allograft rejection (1,2). Rejection reactions, especially after heart transplantation, have been associated with death. In the context of antibody-mediated rejection, graft tissue damage is typically characterized by complement activation (3,4). This activation of the complement system subsequently leads to the migration of inflammatory cells, which plays a pivotal role in the rejection mechanism (5). $CD8^+$ T-cells have been observed to exert a cytolytic effect on target cells by secreting proteins such as perforin and granzyme. $CD4^+$ T-cells, on the other hand, have been shown to participate in the rejection response by secreting cytokines that promote inflammation (6). The aim of this study was to investigate the time-dependent changes in $CD4^+$ and $CD8^+$ T-cell ratios in the presence of donor-specific antibody. The aim of this study was to investigate the time-dependent changes in $CD4^+$ and $CD8^+$ T-cell ratios in the presence of donor-specific antibody.

Material and Methods: Lymphocytes were isolated from a healthy volunteer donor's peripheral blood sample. The presence of DSA was detected using LABScreen Single Antigen Assay (One Lambda). Mean fluorescence intensity (MFI) values greater than 1000 for DSAs against HLA-A, -B, -C, -DR and -DQ at four-digit levels were considered positive. The study protochol was planned as fallows; lymphocytes were divided into three major groups; first lymphocyte and DSA + A*24:02 (MFI: 10.173), DRB1*11:04 (MFI: 14.469), lymphocyte with autologous serum group and lymphocyte with medium groups. Each group incubated and cultured in CO_2 incubator at 37°C for 6 and 24 hours. Cell distributions from each group were evaluated by flow cytometry method using CD45, CD3, CD4, CD8 monoclonal antibodies. Each experimental setup was repeated 3 times and the results were evaluated as compatible with each other.

Results: As a result, it was clear that $CD45^+$, $CD3^+$, and $CD3^+CD4^+$ cell ratios were increased in all of the three groups in a time-dependent manner and the increase was higher in the presence of DSA. For the $CD3^+CD8^+$ cell group, a time-dependent decrease was observed and the decrease was not found to be correlated with the presence of DSA. Each experimental setups' results were aligned with each other (Figure 1).