PCR-BASED DETECTION FOR MICROCHIMERISM AND GRAFT OUTCOME IN KIDNEY TRANSPLANT RECIPIENTS

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OBJECTIVES

Microchimerism (MC) is the presence of a small amount of foreign cells or DNA within a person's circulation or tissues. It has been identified in recipients of solid-organ transplants where it seems to be critical for the development and maintenance of immunological tolerance. Nevertheless, natural and/or iatrogenic MC can be also acquired prior to transplantation., deriving from pregnancy and/or blood transfusion. Aim of the present study was to analyse the possible influence of donor MC after kidney transplantation for possible tolerance mechanism purposes

METHODS

This study included 12 female renal transplant recipients (RTR), mean age 47±8.5 years, undergoing their first cadaveric kidney transplantation. All patients were on prophylactic immunosuppressive therapy based on triple drug association (cyclosporine, steroids and Mycophenolate mofetil). They were prospectively studied by using a quantitative real time PCR method (qPCR) for male MC detection in plasma DNA based on the detection of the DYS14 gene sequence on the Y-chromosome. The presence of Y-related DNA sequence can be considered as a cell-death marker released from necrotic or apoptotic cells in the transplanted organ or donor-derived haemopoietic cells in the recipient's blood or other organs. Persistence of donor DNA in recipient plasma samples was assessed at 15 days and 12 months after transplantation. A pre-transplant blood sample was collected from each patient to serve as an individual negative control.

Graphs and tables

Parameter	Value
Age at transplantation (years)	47±8.5
Number of pregnancies (Median and quartiles)	1 (0,25-2)
Time on dyalisis in months (Median and quartiles)	18,5 (2-40)
Immunosuppressive therapy	Cyclosporin, Mycophenolic acid and steroid
Serum creatinine at 1 year (mg/dl)	1.36±0.35
eGFR at 1 year (ml/min)	74±15.5
Median of Mismatches	3
Blood transfusions (median and quartiles)	2 (0-4,5)
Mean DNA at 15 days	0.80±0.69 ng/ml 121.8±104,8 genome equivalents/ml
Mean DNA at 1 year	0.80±0.69 ng/ml 23.1±40.0 genome equivalents/ml

RESULTS

Mean serum creatinine levels were 1.36±0.35 mg/dl and mean GFR was 74±15.5 ml/min after one year follow up period of the 12 female RTR.

-No acute rejection episode was documented.
-The median of HLA mismatches were 3
-No Y-related DNA was detected in pre-transplant samples.
-Mean DNA quantity after 15 days resulted 0.80±0.69 ng/ml plasma corresponding to 121.8±104,8 genome equivalents/ml plasma.
-A 5-fold decrease was recorded in mean plasma Y-related DNA quantity after 12 months from the transplant, resulting 0.80±0.69 ng/ml plasma (23.1±40.0 genome equivalents/ml plasma). It is worth to note that most of the patients under study (80%) had levels of donor DNA below 10 genome equivalents/ml plasma after one year from the intervention.

CONCLUSIONS

Donor-specific DNA sequences are present in the plasma of all patients after 15 days from kidney transplant. A marked decrease in plasma DNA donor concentration was recorded after one year from transplantation.
None of the patients experienced acute rejection.
Association with clinical and immunological variables remains to be elucidated for those patients experiencing acute rejection.

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