Evaluation of specific T cell responses prior kidney transplantation: useful tool for rejection prediction?

Lucia Straňavová[1], Petra Hrubá[1], Jiří Froněk[3], Antonij Slavčev[4], Eva Girmanová[1], Irena Brabcová[1], Petra Reinke5[5,6], Hans-Dieter Volk[5,6], Ondřej Pelák[7], Tomáš Kalina[7], Ondřej Viklický[1,2]

[1]Transplant laboratory, Institute for Clinical and Experimental Medicine (IKEM); [2]Nephrology Department IKEM; [3]Transplant Surgery Department – IKEM; [4]Department of immunogenetics; [5]Department of Nephrology and Intensive Care Medicine, Berlin, Germany; [6]Institute of Medical Immunology, Charité University Medicine Berlin, Germany; [7]Department of Childhood Leukaemia Investigation Prague (CLIP), Motol University Hospital Prague

INTRODUCTION

Recently activated memory T cells are involved in acute rejection in kidney transplant recipients. These cells are not targeted by the depletive therapy and rapidly proliferate after depletion (Lakkis et al. 2003, Wu Z et al. 2004). Since memory T cells, after stimulation with the appropriate antigen, can produce a variety of cytokines, they can be identified by using the ELISPOT method (Fig. 1).

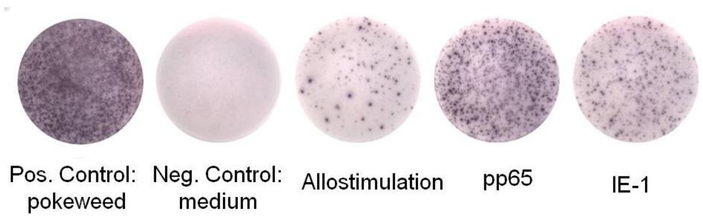

Fig 1: IFN- γ ELISPOT wells after stimulation with different antigens

METHODS

Using ELISPOT, the donor-specific and CMV-specific T cell responses prior- and 6 months after renal transplantation were tested in the 30 living donor kidney transplant recipients and results were correlated with posttransplant outcome.

To verify heterologous immunity we tested cross reactive T cells in healthy volunteers by flow cytometry analysis using *HLA*-A*02:01 NLVPMVATV HCMV pp65 pentamers staining.

RESULTS

In patients with positive IFN-γ ELISPOT (n = 12) before transplantation there was a statistically significant decrease in alloreactivity at month 6 ($p < 0.01$) (Fig. 2). Acute rejection occurred in 8/30 patients but just in 4 of those patients the IFN-γ allo-ELISPOT before transplantation was positive. ROC analysis showed that the IFN-γ allo-ELISPOT before transplantation cannot be used for prediction of rejection (AUC = 0.523, sensitivity 50%, specificity 64.6%) (Fig. 3).

	AUC	Cut-off	Sensitivity	Specificity
Allo pre Tx	0.523	14.25	50	64.6
Allo 6M	0.352	x	x	x
IE-1 pre Tx	0.736	113.25	75	72.7
IE-1 6M	0.659	313.75	50	91.9
pp65 pre Tx	0.67	264.5	75	72.8
pp65 6M	0.67	264.5	75	72.8

Tab. 1: The risk of rejection calculated with ROC analysis for allo and CMV specific T cells reactivity before an 6M after Tx. The best predictor of rejection is IE1 pre Tx.

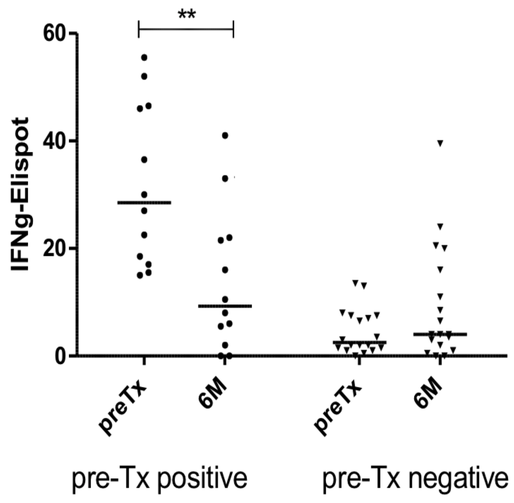

Fig. 2: In patients with positive IFN-γ allo ELISPOT before Tx is significant decrease of alloreactive T cells, probably to immunosuppression load

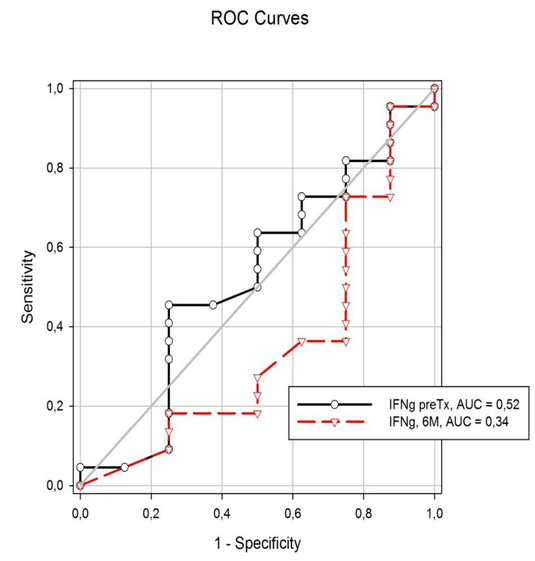

Fig. 3: IFN g does not predict rejection as shown by ROC analysis.

However, positive IE-1 (antigen CMV) IFN-γ ELISPOT before transplantation was found to predict acute rejection (AUC = 0.736, sensitivity 75%, specificity 72.7%, cut-off = 113.25, $p = 0.0112$) (Fig. 4, 5). Therefore heterologous immunity is suggested to influence this results. To prove this concept we tested cross reactivity in healthy volunteers by FACS analysis (Fig. 6). On fig. 6C cross reactive T cells are displayed. We successfully detected cross reactive cells in healthy volunteer and their proportion from CMV specific CD8+ T cells was 0.2%.

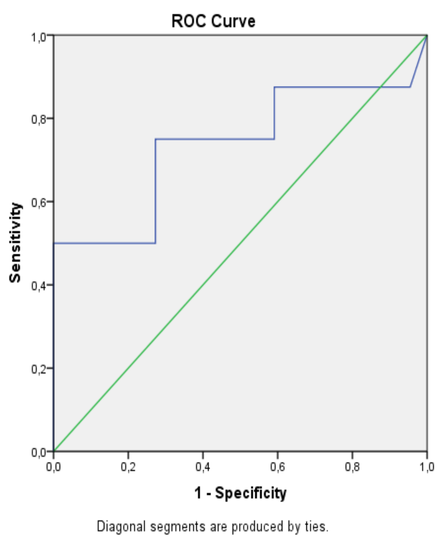

Fig. 4: Prediction of rejection risk on the basis of pre Tx IE1 ELISPOT

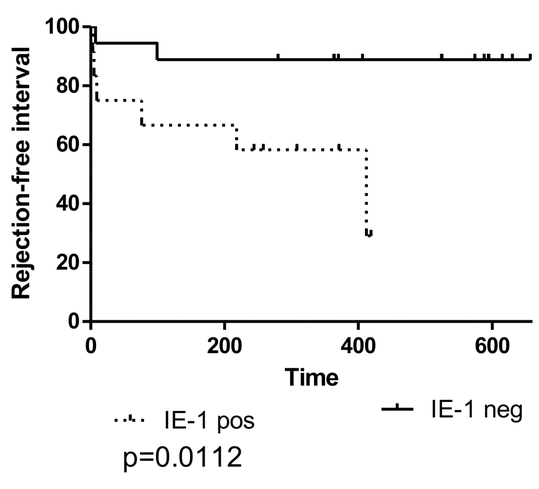

Fig. 5: Rejection-free interval for patients with IFN-γ IE-1 positive and negative ELISPOT

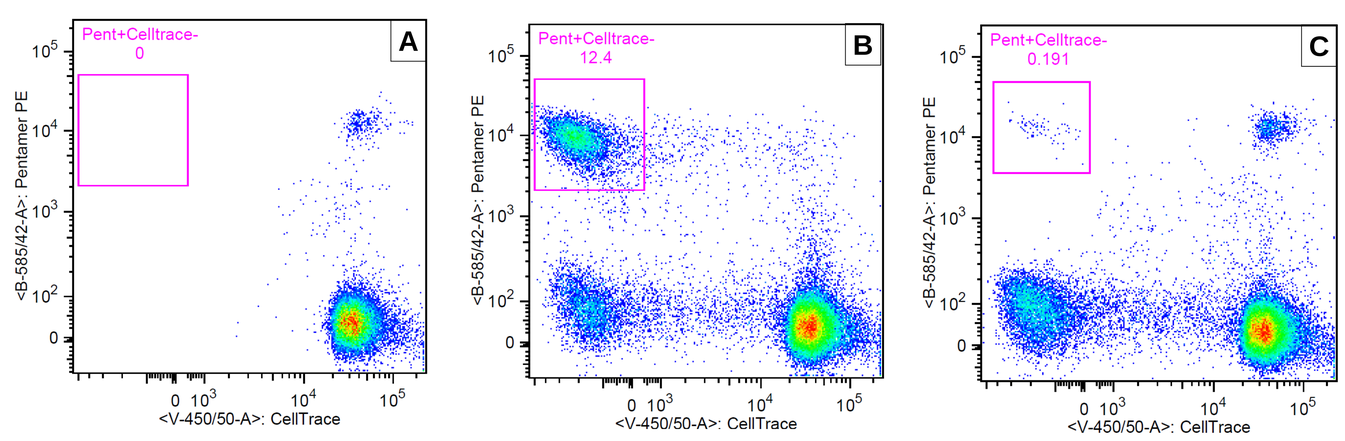

Fig. 6: Pentamer analysis of cross reactivity. A: Negative control, recipient cells with medium, B: recipient cells stimulated with CMV pp65, C: recipient cells stimulated with donor cells

CONCLUSIONS

It is likely that the detection of CMV-specific T cell reactivity in kidney transplant recipient could be used as a tool to predict risk of acute rejection. These results support the hypothesis about crucial role of heterologous immunity in posttransplant immune response.

REFERENCES

Lakkis FG, Sayegh MH. Memory T cells: a hurdle to immunologic tolerance. J Am Soc Nephrol. 2003 Sep;14(9):2402-10

Wu Z, Bensinger S, Zhang J, Chen C, Yuan X, Huang X, Markmann J, Kassaee A, Rosengard BR, Hancock WW, et al. Homeostatic proliferation is a barrier to transplantation tolerance. Nature Medicine. 2004;10:21–23

ACKNOWLEDGEMENT

Project supported by 7FP programs of EU BioDrim and MABSOT and by the Internal Grant Agency of the Ministry of Health of the Czech Republic No: 15-26865A.

53 ERA
692--MP
Renal transplantation. Clinical.
Lucia Stranavova
DOI: 10.3252/pso.eu.53era.2016
ePosters supported by F. Hoffmann- La Roche Ltd.
53rd CONGRESS VIENNA AUSTRIA ERA-EDTA MAY 21st-24th, 2016
Held jointly with Österreichische Gesellschaft für Nephrologie
PosterSessionOnline.com