CIRCULATING ENDOTHELIAL PROGENITOR CELLS EFFECTIVELY RECONSTITUTE ENDOTHELIUM AND REDUCE ALLOREJECTION OF MOUSE AORTIC ALLOGRAFT

J. Li[1], M. Mueller[2], K. Paul[3], N. Werner[3], B. Metzler[1], J. Kovarik[1], M. Luyten[1], G. Weckbecker[1], H.-G. Zerwes[1], A. Katopodis[1],C. Bruns[1] and **G. Wieczorek**[1]

[1]Autoimmunity and Transplantation Research, Novartis Institutes of Biomedical Reserach, Basel, Switzerland; [2]Developmental and Molecular Pathways/Models of Disease Center, Novartis Institutes of Biomedical Research, Basel, Switzerland and [3]Molekulare Kadiologie, Universitaetsklinikum Bonn, Bonn, Germany.

OBJECTIVES: Allograft endothelial cells are the first biological interface between the transplanted donor organ and circulating immunocompetent cells of the recipient that play a key role in rejection processes. Endothelial cell apoptosis induced by an inflammatory environment results in exposure of the subendothelial matrix and enhancement of the inflammatory reaction. Circulating endothelial progenitor cells (EPCs) may allow endothelial regeneration and lead to a decrease of alloreactivity. The main objective of this study was to demonstrate whether increased number of circulating EPCs lead to accelerated reconstitution of endothelium and diminish allogeneic rejection of mouse aortic allograft.

METHODS: $1.5x10^6$ EPCs expressing sca-1^+VEGFR2^+c-kit^+CD34^+ were isolated from spleens of C57Bl/6-actb-EGFP knock-in mice and transfused intravenously into C57Bl/6 recipients of Balb/c mice aorta (n=22) or non-transplanted controls (n=3) on day 0, 3 and 7 after transplantation. Aorta allografts were harvested on day 3, 14 or 35 post-transplantation (post-Tx) for histological analysis of endothelial and intimal layers. In addition, allo-antibody levels and hematology as well as homing of circulating EPCs were evaluated in the recipients.

RESULTS: EPC transfusions caused accelerated re-endothelialization of the allografts on day 14 post-Tx (up to 75% of the graft surface versus 10% in controls). This phenomenon was associated with a 30% reduction of neointima formation in comparison to control animals on day 35 post-Tx. Newly generated endothelial cells were confirmed to derive from transfused EGFP-positive EPCs. Allo-antibody levels (IgG and IgM) in serum were reduced significantly early after EPC transfusion up to 14 days post-Tx ($p<0.05$). WBC and lymphocyte counts in blood were also reduced. Transfused EPC distribution in tissues was determined as blood > LN > BM. Circulating EPC count in transplanted mice was higher than that in non-transplanted controls.

CONCLUSION: For the first time it was possible to demonstrate that EPCs significantly reconstitute endothelium of vessel allografts and reduce the progression of the rejection process. Further studies are needed to elucidate the mechanism of EPC mobilization, migration, differentiation, and homing to the target areas. A better understanding of the biology of EPCs will help to increase their therapeutic potential in clinic medicine.