generation (through e-NOS activation) and AMPK phosphorylation. Mitochondrial damage and oxidative stress were also prevented by BZ. In addition, BZ inhibited adiponectin, IL-1 and TNF alpha, only in steatotic livers. Conclusion: BZ at low and nontoxic doses is very efficient to protect fatty liver grafts against cold IRI, when added in UW preservation. BZ benefits are mediated by nitric oxide showing the relevance of UPS in fatty liver preservation.

Abstract# 1445 Poster Board #-Session: P7-IV

Liver Graft Washout Is Determinant for the Efficient Prevention of Reperfusion Injury: A Role for the Cytoskeleton and Glycocalyx Preservation. M. A Zaouali,[1] M. Bejaoui,[1] M. Calvo,[2] R. Adam,[3] E. Bonaccorsi-Riani,[4] A. Sánchez-Fueyo,[4] A. Rimola,[4] H. Ben Abdennebi,[5] J. Roselló-Catafau.[1] *[1]Institut d'Investigacions Biomèdiques de Barcelona (IIBB), CSIC-IDIBAPS, Barcelona, Spain; [2]Serveis Cientifico Tècnics, Universitat de Barcelona, Barcelona, Spain; [3]Liver Transplantation Unit, Paul Brousse Hospital, Villejuif, France; [4]Liver Transplant Unit, Hospital Clinic of Barcelona, (IDIBAPS-Ciberehd), Barcelona, Spain; [5]Human Physiology, Faculty of Pharmacy , University of Monastir, Monastir, Tunisia.*

Antecedents: Hepatic ischemia reperfusion injury (I/R) contributes to the initial poor function or primary non-function after transplantation. This is due to the cold preservation, rewarming and reperfusion, respectively. We evaluated the benefits of using a new rinse solution against I/R. Experimental: Sprague-Dawley rats (180-200 g; n= 6 for each group), were classified as follows: Group 1(controls)= Livers preserved in UW solution (24 hours; 4 ºC) were flushed with Ringer lactate solution(RLS) and subjected to 2 h-reperfusion at 37 ºC using an ex-vivo model; Group 2 = Same as 1 but the liver grafts were flushed with the new rinse solution composed by CaCl2.2H2O, KH2PO4, NaH2PO4, MgSO4.7H2O, lactobionate and rafinose at pH=7.4; Group 3: same as 2 but with polyethyleneglycol-35 (PEG-35) addition at 1g /L. Group 4 = same as 3 but with PEG-35 addition at 5g/L. Determinations: Liver injury (AST/ALT); mitochondrial damage (GLDH); function (bile output and vascular resistance), oxidative stress (MDA), nitric oxide; as well as glycocalyx (syndecan, hyaluronic acid) and cytoskeleton (filament and globular actin fraction) degradation were assessed. Also metalloproteinases (MMP2 and MMP9) were determined. In addition, actin confocal microscopy studies were carried out. Results: PEG-35 at 1g/L and 5g/L reduced AST/ALT levels, ameliorated liver function when compared to Groups 1 and 2. This was accompanied by decreases in GLDH, MDA and increases in e-NOS activation. Interestingly, the PEG-35 benefits were closely associated with the inhibition of MMP9 and the stabilization of actin filament fraction (western blot and confocal microscopy).Glycocalyx was also protected (low syndecan-1 and hyaluronic acid levels). Conclusions: The most suitable liver graft protection was observed for PEG35 (5 g/L) ("graft post-conditioning") that reinforced the stabilization of the liver graft cytoskeleton and glycocalyx.

Abstract# 1446 Poster Board #-Session: P8-IV

Arginase Inhibition Improves Acute Kidney Injury after Ischemia Reperfusion Injury and Renal Transplantation in Mice. N. Shushakova,[1,2] S. Rong,[1] P. Suzdak,[3] B. Tomczuk,[3] M. Mengel,[4] H. Haller,[1] F. Gueler,[1] J. Menne.[1,2] *[1]Dept. of Nephrology, Medical School Hanover, Hanover, Germany; [2]Phenos GmbH, Hanover, Germany; [3]Corridor Pharmaceuticals, Towson, MD; [4]Alberta Transplant Applied Genomics Centre, University of Alberta, Edmonton, Canada.*

Increased arginase expression and activity has been demonstrated in ischemic tissues and arginase inhibition has recently been shown to protect against myocardial ischemia and reperfusion (IR) injury.

In the present study we tested a novel arginase inhibitor, AX-01128, for its ability to improve survival and attenuate loss of kidney function in a clinically relevant model of renal IR injury and ischemia induced acute kidney transplant rejection in mice.

IR injury was induced in male C57Bl/6 mice by bilateral renal pedical clamping for 30 min. AX-01128 was administered iv at either 10 or 30 mg/kg) versus vehicle 15 min before placing the clamps and at 5 minutes prior to releasing the clamps. Survival and renal function were monitored.

For allogenic kidney transplantation, male C57Bl/6 mice ($H2^b$) were used as kidney donors and female BALB/c ($H2^d$) mice served as recipients. AX-01128 was administered iv at either 1, 3 or 10 mg/kg versus control to both donor and recipient 15 min before surgery and the second injection was given to the recipients at 5 minutes after transplantation. The allografts were flushed with AX-01128 solution diluted 1:10 with saline and kept in this solution during 60 min of cold ischemia. Renal function was monitored, and allograft pathology was evaluated according to the updated Banff classification at day 6 after transplantation.

AX-01128 was effective in reducing mortality and improving renal function in a dose dependent manner after acute IR induced kidney injury. Treatment with 10 or 30 mg/kg AX-01128 resulted in 20% and 30% long-term survival, respectively. All mice of the control group died within 5 days after surgery.

In the ischemia induced renal transplant rejection model serum creatinine and BUN elevation was significantly reduced in mice treated with AX-01128 compared to vehicle. Histological analysis revealed that AX-01128 significantly reduced acute tubular necrosis but had little effect on inflammation, suggesting that the arginase inhibition is suitable to prevent ischemia/reperfusion injury.

Conclusion: AX-01128 is a novel and promising therapeutic agent for treating AKI. Its beneficial effect may be due to reduced acute tubular necrosis after IR injury.

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Abstract# 1447 Poster Board #-Session: P9-IV

Renal Allograft Rejection and Delayed Graft Funciton Regulated by miRNAs. J. Wilflingseder,[1,2] H. Regele,[3] A. Kainz,[1,2] P. Perco,[4] A. Soleiman,[3] F. Mühlbacher,[5] B. Mayer,[4] R. Oberbauer.[1,2] *[1]Nephrology, KH der Elisabethinen, Linz, Austria; [2]Nephrology, Medical University of Vienna, Vienna, Austria; [3]Pathology, Medical University of Vienna, Vienna, Austria; [4]Emergentec Biodevelopment GmbH, Vienna, Austria; [5]Transplantation, Medical Universtiy of Vienna, Vienna, Austria.*

Increasing evidence accumulates on the central involvement of miRNAs in disease pathophysiology. In this study we identified deregulated miRNAs in renal allograft biopsies from patients who developed acute vascular rejection, acute interstitial rejection (AREJ), antibody mediated rejection (ABMR) and delayed graft function (DGF).

Sixty-five post-transplant kidney biopsy samples covering 30 cases with acute rejection, 11 cases showing ABMR, 14 DGF cases, and 10 protocol management biopsies (PBx) serving as controls were analyzed using the Affymetrix GeneChip® miRNA Array. Differentially regulated miRNAs were identified by Student's t-test and Bonferroni correction. Target genes were evaluated using prediction algorithms (>3 db), correlations to existing mRNA profiles and experimental evidence (mirTarBase) and were used as basis for functional annotation and molecular interpretation.

Patients with AREJ, ABMR and DGF discriminate from the control group (protocol biopsies) in the unsupervised cluster analysis and also in the principal component analysis suggesting miRNAs play an important role in rejection and DGF. Between six and twenty-one miRNAs were identified as significantly differentially regulated when comparing control samples and clinical biopsies. Cell cycle, apoptosis and metabolism were the main target processes of the differentially regulated miRNAs. Dicer protein expression as key protein of miRNA biogenesis was suppressed in tubule cells of AREJ and TGF-beta signalling was the most affected pathway in DGF. These data suggest that distinct miRNA signatures are associated with acute cellular and humoral rejection and delayed graft function. A detailed analysis of the miRNAs and target genes will be presented at the meeting.

Abstract# 1448 Poster Board #-Session: P10-IV

Protein Kinase C-delta (PKC-d) Contributes to Cardiac Allograft Rejection by Regulating IL-33 Expression. H. R. Cho,[1,2] S. J. Park,[2] J. S. Lee,[1,3] H. J. Kim,[4] H. C. Jung,[3] J. P. Jung,[5] B. Kwon.[1,4] *[1]Biomedical Research Center, Ulsan University Hospital, College of Medicine, University of Ulsan, Ulsan, Japan; [2]Dept. of Surgery, Ulsan University Hospital, College of Medicine, University of Ulsan, Ulsan, Korea; [3]Dept. of Internal Medicine, Ulsan University Hospital, College of Medicine, University of Ulsan, Ulsan, Korea; [4]School of Biological Sciences, University of Ulsan, Ulsan, Korea; [5]Dept. of Cardio-vascular Surgery, Ulsan University Hospital, College of Medicine, University of Ulsan, Ulsan, Korea.*

PKC-d plays a critical role in proliferation and apoptosis of various types of cells. In this study, we examined the role that PKC-d plays in cardiac allograft rejection in mice. In the C57BL/6 ($H\text{-}2K^b$)àC3H.SW ($H\text{-}2K^b$) minor histocompatibility antigen-mismatched cardiac allograft model, PKC-d-deficient cardiac allografts exhibited more severe vasculopathy, inflammation, and damage of cardiac muscles. Results from RT-PCR and cDNA microarray analysis were in agreement with histopathological data in that there was markedly increased expression of molecules involved in chemotaxis, costimulation, inflammation, tissue remodeling, and cell adhesion. IL-33 was rapidly upregulated in PKC-d KO syngeneic cardiac grafts or allografts. Further analysis showed that secretion of IL-33 was induced in PKC-d KO but not wild-type cardiac fibroblasts under a hypoxic condition. Finally, we demonstrated that there was milder leukocyte infiltration in IL-33 KO cardiac allografts. Taken together, our results clearly indicate that PKC-d regulates acute inflammation by modulating the expression and secretion of IL-33 during ischemia and reperfusion in cardiac allograft rejection.