Molecular Pathways of Cardiac Allograft Dysfunction
Independent of Acute Cellular Rejection

Methods: Cardiac allograft recipients were prospectively enrolled as part of The Cardiac Allograft Rejection Gene Expression Observational (CARGO) multi-center study. Subjects were followed at each post-transplant visit with biopsy (read by 3 pathologists blinded to clinical data), whole blood BNP, echocardiography and hemodynamics. GE profiles of circulating cells were evaluated using Luminex microarrays with > 8,000 genes and validated with real-time PCR (RT-PCR).

Results: 42 subjects were followed for two years. For 342 encounters the median BNP level was 190 pg/ml. Levels differed significantly as a function of gender and ethnicity (higher in women and black Americans, p < 0.05). BNP levels were elevated in those with Grade 3A rejection (n=9) compared to Grade 0 (n=35, p < 0.003) but lacked specificity for acute rejection. GE profiles of patients with elevated BNP levels (≥295 pg/ml) compared to those with lower levels (≤182 pg/ml, n=27) identified 25 genes correlated to BNP (p<0.035). The genes were associated with granulocyte and monocyte lineages and included elastases, adherence receptors, metalloproteinases and cytokine receptors. They were distinct from genes correlated to acute cellular rejection using microarrays and RT-PCR in the multi-center study. For 35 patients, BNP levels were compared to quantitative results of a clinically validated 14 gene RT-PCR test for acute cellular rejection. No correlation was found.

Conclusions: Peripheral immune cell molecular pathways indicative of acute allograft dysfunction are associated with elements of innate immune distinct from cellular immunity pathways. GE assays for acute rejection and assessment of graft function by BNP may be complementary for detection of the quiescent state in cardiac allograft recipients.

Inhibition of p38 Mitogen Activated Protein Kinase Mediates Endothelial Cell Survival Following Cardiac Transplantation

Background: The hypothesis of ischemic preservation required for cardiac transplantation exposes the donor heart to myocardial ischemia/reperfusion (IR) injury upon implantation. p38 mitogen-activated protein kinase (MAPK) has been directly linked to increased apoptosis in models of myocardial IR injury and its inhibition has significantly improved post-ischemic myocardial function in in vivo models. However, the intracellular signaling pathways responsible for these changes are not well determined. Additionally, the inhibition of p38 MAPK into myocardial preservation solutions has yet to be examined. Here we hypothesize that the incorporation of the p38 MAPK inhibitor, SB239063, into University of Wisconsin (UW) preservation solution results in effective inhibition of TNF-α-induced p38 MAPK activation. The inhibition of p38 MAPK may play a key role in mediating endothelial cell survival through the activation of the pro-survival signals, AKT and ERK.

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Results: 1) UW solution with SB239063 (0.5M, 4°C, 12hr) is completely ineffective in inhibiting TNF-α-induced p38 MAPK activation (n=3). 2) Inhibition of p38 MAPK produced an average upregulation of AKT activity of 42% and an average upregulation of ERK1/2 activity of 148% (n=3).

Conclusion: The p38 inhibitor, SB239063, has been effectively incorporated into UW solution to inhibit TNF-α-induced p38 MAPK activation. Inhibition of p38 MAPK upregulates the activities of the anti-apoptotic signals AKT and ERK1/2. These data suggest that p38 MAPK is a pro-apoptotic signal whose inhibition may represent a novel method to mitigate apoptosis and improve myocardial performance following cardiac transplantation.