**924-76**

**A New Non-Invasive Method for Detection and Assessment of Aortic Regurgitation During Routine Blood Pressure Recordings**

Todd J. Brinton, Tsai-Lieh Hsu, Oi-Ling Kwan, Chun-Pang Liu, Mau Song Chang, Shiu-Shin Chio, Anthony N. DaMaria. Veterans Hospital, Taipei, Taiwan, University of California, San Diego, CA

Recently, we developed an oscillometric cuff technique (CUFF) to non-invasively derive arterial pressures and waveforms. Using this method we observed a unique pattern of pressure oscillations (PO) that occurred in patients with severe aortic regurgitation (AR). To further define the potential mechanism of this phenomenon, and its value in the detection and assessment of AR, we performed clinical and modeling studies. CUFF was performed in 10 normal (N) and 15 pts whom we documented and semi-quantitated by echo. In 10 N, and all 5 mild AR pts, a bell shaped distribution of PO was observed from supra-systolic to sub-diastolic cuff pressure (top figure). However, all 10 pts with grade III (severe) AR exhibited a phasic alteration of PO conforming to a resonance pattern (bottom figure). To test the hypothesis that this phenomenon represented a ventricular-fluid mechanics interaction produced by AR, we utilized a simple amplitude modulation model (W1 = incident, W2 = reflection) and found that the pattern could be reproduced at specific amplitudes and frequencies.

Thus, CUFF recordings of arterial pressure exhibit a marked resonance pattern in pts with severe AR, likely due to ventricular-fluid mechanics interaction. This phenomenon should be useful in detecting and assessing AR during routine blood pressure recordings.

**925**

**Transplant Rejection**

Monday, March 25, 1996, 3:00 p.m.-5:00 p.m.
Orange County Convention Center, Hall E
Presentation Hour: 4:00 p.m.-5:00 p.m.

**925-1**

**Deterioration in Cardiac Function is Associated With the Appearance of Specific HLA Antibodies After Transplantation**

Stephen H. Leech, Paul J. Mather, Howard J. Eisen, Valluvan Jeevanandam. Temple University Medical School, Philadelphia, PA

The purpose of this study was to investigate the effect of HLA antibodies specific for mismatched donor HLA antigens appearing in the circulation after human orthotopic heart transplantation. Although there is increasing evidence of a deleterious effect of mismatched donor HLA antigens on the outcome of human cardiac allografts, the role of HLA antibodies remains controversial. Thus, their presence prior to cardiac transplantation has been associated not only with poor outcome by some workers but also of no clinical significance by others. Furthermore, their appearance after cardiac transplantation has also been the subject of conflicting reports. HLA antibodies were identified by a standard microlymphocytotoxicity technique using panels of frozen lymphocytes from normal donors who had been tissue typed. Of 78 patients transplanted over a 12-month period, 4 developed HLA antibodies specific for mismatched donor HLA antigens on the outcome of human cardiac allografts, the role of HLA antibodies remains controversial. Thus, their presence prior to cardiac transplantation has been associated not only with poor outcome by some workers but also of no clinical significance by others. Furthermore, their appearance after cardiac transplantation has also been the subject of conflicting reports. HLA antibodies were identified by a standard microlymphocytotoxicity technique using panels of frozen lymphocytes from normal donors who had been tissue typed. Of 78 patients transplanted over a 12-month period, 4 developed HLA antibodies specific for mismatched donor HLA antigens on the outcome of human cardiac allografts.

**925-2**

**Myocardial Apoptosis in a Heterotopic Murine Heart Transplant Model of Chronic Rejection/Graft Vasculopathy**

Robert Shaddy, William White, Lynda Zhang, Michael Trautman, Sherrie Perkins, Elizabeth Hammond, Jane Shelley, University of Utah and Primary Children's Medical Center, Salt Lake City, UT

Apoptosis (programmed cell death) has been implicated in myocardial reperfusion injury and in experimental transplant rejection. One mechanism of apoptosis is through the interaction of the cell-surface receptor on target cells and the Fas ligand which is expressed on cytotoxic T cells. To determine whether apoptosis occurs in the myocardium of transplanted hearts, we examined a murine heterotopic heart transplant model of chronic rejection/graft vasculopathy (strain B10.A to B10.RH). Hearts harvested after 30 days showed an initial index of the allograft (0.5 ± 0.1) (mean ± SE) that was 15 to 50 times more than syngeneic grafts (0.03 ± 0.01) and native (nontransplanted) hearts (0.01 ± 0.01) (p < 0.001). In situ end-labelling of partially degraded DNA with terminal deoxynucleotidyl transferase showed an increase in apoptotic cells/20 hpf in allografts (2.0 ± 0.3) and syngeneic grafts (3.0 ± 0.4) compared to native hearts (0.0 ± 0.0) (p < 0.001, ANOVA). Both myocytes and nonmyocytes appeared to be undergoing apoptosis. RT-PCR detected equal myocardial RNA signal intensity of Fas in allografts, syngeneic grafts, and native hearts (n = 4). In contrast, allografts showed a strong signal for the Fas ligand mRNA, a signal not seen in syngeneic grafts or in the native hearts. We conclude that apoptosis is occurring in both murine cardiac allografts and syngeneic grafts and that Fas ligand is strongly expressed in murine allografts. Since both allografts and syngeneic grafts show in-situ evidence of apoptosis, but only allografts express Fas ligand, the mechanisms of apoptosis in this model may not be exclusively related to Fas/Fas ligand interactions.

**925-3**

**Immunologic Characterization of Allograft-Infiltrating Cells in Human-Severe Combined Immunodeficiency Mouse Chimeras: Evidence for Human Effector Cell Mediation of Rejection**

Jin Kang, Sheni E. Beiland, Howard J. Eisen. Temple University, Philadelphia, PA

Novel immunosuppressive agents are initially tested in experimental animals, usually rodents. This approach often fails as there are substantial differences between human and rodent immune systems. We have developed a potential in vivo model of human cardiac transplant rejection by reconstituting severe combined immunodeficiency (SCID) mice with human peripheral blood lymphocytes (PBL's) and 28 days later implanting segments of human atrial tissue from patients undergoing CABG surgery spontaneously (SQ) in the human-SCID chimera. 7 days after lymphocytic infiltration and myocyte necrosis is seen in the cardiac allografts in these chimeras by hematoxylin and eosin staining, resembling human cardiac transplant rejection. The origin and type of cells infiltrating the grafts are unknown. To determine the phenotype and origin of these cells, we administered 5 x 10^6 PBL's intraperitoneally to 4 SCID mice and confirmed engraftment of human PBL's by flow cytometry: 3 control SCID mice were not reconstituted. Segments of human atrial tissue were implanted SQ in the human-SCID chimera and control SCID mice. 7 days later, cardiac tissue was removed and stained with human-specific anti-CD4, CD8, IL-2 receptor (R), CD3 and ICAM-1 antibodies. Infiltrating cells were seen in the reconstituted SCID mice and were primarily CD4+ and CD3+ with over 25% IL-2+ receptors. Myocardial lymphocytic infiltration or myocyte necrosis was seen in the control SCID mice. Endothelial ICAM-1 expression was seen only in allografts in reconstituted SCID mice. We conclude that cells infiltrating cardiac allografts in human-SCID chimera are human-derived, activated CD4+ similar to those seen in human cardiac allograft rejection and that they induce allograft endothelial activation.

**925-4**

**Detection of Heart Transplant Rejection by Doppler Tissue Imaging**

John A. Pullo, Mark W. Weston, Michael F. French, Guillermo Cintron, Mitchell G. Davis, Hector L. Fontaine. Division of Cardiology, University of South Florida and Tampa General Hospital, Tampa, FL

Heart transplant rejection (HTR) commonly occurs in heart transplant recipients and requires invasive endomyocardial biopsy (EB) for diagnosis. Thus far, non-invasive studies have had inadequate sensitivity to detect HTR. Pulsed wave Doppler tissue imaging (PWDTI) is a new non-invasive imaging modality capable of quantifying myocardial tissue velocities. Since HTR may cause disturbances in myocardial relaxation, we performed this study to determine whether moderate HTR results in reduced myocardial peak early relaxation velocities (PEV). Methods: 40 orthotopic heart transplant recipients underwent serial PWDTI at the time of routine surveillance EB.
patients had moderate rejection (grade 3a or 3b), the remainder were grade 0 to 1b. All HTR patients had myocardial relaxation velocities determined by PWTDI prior to, during, and following successful treatment from HTR at the time of EB. PEV were measured from the short axis view of the inter-

posterior wall by PWTDI (Acuson XP20). Results: There was a significant reduction in PEV during HTR from 0.196 ± 0.021 to 0.120 ± 0.029 m/sec; p < 0.0001 with subsequent normalization following successful treatment to 0.184 ± 0.036 m/sec. p < 0.0001 (see graph). Furthermore, a PEV of <0.15 m/s has a negative predictive value of 95% to exclude rejection, a sensitivity of 82% and specificity of 60%.

**Conclusion:** HTR results in a reduction in myocardial peak early relaxation velocities. Hence, PWTDI may be a clinically useful noninvasive tool for the diagnosis of HTR and for following responses to therapy.

**926-6** Non-Invasive Detection of Cardiac Allograft Rejection by Multivariate Analysis of Computer Generated M-Mode Data in Pediatric Patients

Andrew J. Maxwell, Daniel Bernschein. Stanford University, Stanford, CA

The aim of this study was to determine if a computer program, designated to analyze echocardiographic M-Mode, could predict allograft rejection. Serial echocardiograms (n=7) were performed in 5 patients, each having 24 hours of RV and LV myocardial biopsy. M-Mode were digitalized and retrospectively analyzed by a computer program designed to curve-fit the inner and outer posterior wall and both edges of the septum averaged over 4 to 7 cardiac cycles. Manipulation of the equations describing these curves produced 47 data values of distances, volumes, rates of change and time periods from each M-Mode analysis. The collective data were analyzed for correlation with biopsy results. The positive predictive value for grade III and IV (6) after conversion to individual Z scores. Data was subjected to Hierarchical Discriminate Function Analysis (HDFA) designated to minimize Wilks' Lambda. HDFA produces a set of classification equations which assigns subjects to rejection groups and additionally produces a Centroid Map giving visual information on the reliability of any one assignment. HDFA resulted in several combinations of variables and coefficients producing significance (p < 0.001) between the three rejection groups. Cross-validation resulted in a sensitivity to detect any rejection of 0.89, to detect M of 0.74, and to detect S of 0.78 (specificity = 0.86). The positive predictive value for M = 41% and S = 64%. Misassigned cases plotted on the Centroid Map in locations suspicious for reliability.

**Conclusion:** HDFA and HDFA with Wilks' Lambda produces a set of classification equations which assigns subjects to rejection groups and additionally produces a Centroid Map giving visual information on the reliability of any one assignment. HDFA resulted in several combinations of variables and coefficients producing significance (p < 0.001) between the three rejection groups. Cross-validation resulted in a sensitivity to detect any rejection of 0.89, to detect M of 0.74, and to detect S of 0.78 (specificity = 0.86). The positive predictive value for M = 41% and S = 64%. Misassigned cases plotted on the Centroid Map in locations suspicious for reliability.

**925-7** Protective Effects of Complement Blockade In an Isograft Model of Lung Preservation and Transplantation


Agents which inhibit complement activation protect against myocardial reper-
sition injury and may be beneficial in xenotransplantation, but it is not known whether complement activation is deleterious following lung preservation and transplantation in an isograft model. Lungs harvested from Lewis rats subjected to 4°C 6 hour preservation in Euro-Collins solution followed by transplantation into Lewis recipient rats demonstrated increased immunostaining for C5b-9 compared with nontransplanted controls, confirming local complement activation in this model. To investigate the physiologic relevance of complement activation in the transplanted lung, the native pulmonary artery (PA) was ligated following transplantation, and pulmonary vascular resist-

ance (PVR, mm Hg/min/l), PA flow (PAl=, ml/min), arterial oxygenation (PaO2, mm Hg), graft neutrophil infiltration (myeloperoxidase activity, MPO, units/mg), and recipient survival (Surv, %) were measured at 30 minutes. Animals received either saline (control; n = 12) or soluble comple-
ment receptor type-I (sCR1, 15 mg/kg, serum level range 127-355 μg/ml by ELISA; n = 9) 2 minutes prior to reperfusion. sCR1 treated animals showed a marked reduction in serum complement activity (CH50 90% lower than control animals, p < 0.0001).

**Conclusion:** These findings suggest that moderate rejection ISHT 2 progresses to higher grade of rejection in a considerable number of patients and close monitoring is necessary even late after tx.

**926 Pediatric Interventional Catheterization**

Monday, March 25, 1996, 3:00 p.m.–5:00 p.m.
Orange County Convention Center, Hall E
Presentation Hour: 3:00 p.m.–4:00 p.m.

Robert J. Sommer, Frank F. Ing. Mount Sinai Medical Center, New York, NY; Schneider Children's Hospital, New York, NY; Texas Children's Hospital, Houston, TX

Transarterial and transvenous techniques for transcatheter occlusion of small patent ductus arteriosus (PDA) using a Gianturco coil (GC) have been compi-

licated by GC embolization to the pulmonary and systemic circulation, by

**Conclusion:** In parallel with the reduction in CH50 in sCR1 treated animals, immunohis-
tology revealed decreased C5b-9 deposition compared with controls. Taken together, these data indicate that local complement activation occurs follow-
ing lung preservation and transplantation in an isograft model, and that inhibiting complement improves outcomes following transplantation.