Results: Assessment of remodelling deformation in the LAD distribution (n=2) revealed an increase in wall thickness (E33=0.14, 0.16) and the range of transverse shears (E13, E23) from –0.02 to +0.1, resulting in net increase in tissue volume (product of stretch ratios: 1.21, 1.04). Immunohistochemistry revealed evidence of uPA transfection in the LAD distribution (n=1), and an increase in type I collagen degradation in the LAD distribution, compared with the LCx distribution (n=2). No increase in type I collagen degradation was observed in both the LAD and LCx distribution of the pig in which β-galactosidase gene was delivered (n=1).
Conclusion: Results from this ongoing study indicate that uPA induces extracellular matrix degradation, yielding altered ventricular remodelling.

4:30 p.m.

864-3 Reduction of Secondary Vasospasm Limits Cardiomyopathy Progression

Matthew T. Wheeler, Keith A. Collins, Claudia E. Korcarz, Andrew A. Hack, Sara Zarnegar, Matthew Lyons, Elizabeth M. McNally, University of Chicago, Chicago, IL

Background: In humans and mice, sarcoglycan gene mutations cause cardiomyopathy and muscular dystrophy characterized by focal cardiac necrosis, cardiomyopathy and vasospasm. Mice engineered to lack γ-sarcoglycan show evidence of cardiomyopathy, but the contribution of vasospasm to this pathology is unknown.
Methods: Smooth muscle tissues of γ-sarcoglycan mutant mice were studied by immunofluorescence and immunoblotting. Microvascular filling was used to evaluate vasospasm. γ-sarcoglycan mutant mice (n=10) were treated with oral verapamil at 1 mg/kg/day for 5 months. Treated and untreated γ-sarcoglycan mutant mice were compared by transthoracic echocardiography, ambulatory telemetry, histopathology, and microvascular filling analysis.
Results: Microvascular filling of γ-sarcoglycan mutant mice showed focal stenoses consistent with vasospasm, yet the sarcoglycan complex was not perturbed in vascular smooth muscle in γ-sarcoglycan mutant mice. These data indicate that vasospasm develops as a secondary consequence of intrinsic cardiomyocyte degeneration. Verapamil treatment to reduce secondary vasospasm in γ-sarcoglycan mutant mice attenuated myocardial fibrosis. Membrane permeability defects were present but reduced in verapamil-treated hearts. Verapamil-treated γ-sarcoglycan mice had improved left ventricular fractional shortening (44.3% treated vs. 37.4% untreated), improved maximal velocity at the aortic outflow tract (114.9 vs. 92.8 cm/s, p<0.05) and improved cardiac index (1.06 vs. 0.67 ml/min/g, p<0.001) compared to untreated mice.
Conclusions: These data indicate that secondary, not primary, vasospasm contributes to cardiomyopathy progression in sarcoglycan gene mutations. Moreover, verapamil can reduce secondary vasospasm and limit the progression of cardiomyopathy.

4:45 p.m.

864-4 External Pharmacological Control of Vector-Based Phospholamban-Antisense-RNA Expression for the Flexible Modulation of Cardiac Calcium Homeostasis

Wolfgang C. Poller, Henry Fechner, Xiaomin Wang, Dick Dekkers, Jos Lamers, University Hospital Benjamin Franklin, Berlin, Germany, Erasmus Universiteit Rotterdam, Rotterdam, The Netherlands

Background: Impaired sarcoplasmic reticulum Ca2+ pump (SERCA2) activity plays an important role in the development of diastolic dysfunction of the failing heart. Genetic complementation experiments in MLP-/- x PL-/- knock out mice, and other data, suggest that the expression ratio of PL to SERCA2 may be a target for the improvement of diastolic dysfunction in the failing heart. Previously we have developed PL-antisense-RNA strategies using either a constitutive CMV or an endothelin-1-inducible ANF promoter. External control of applied vectors in vivo can not be achieved using these systems. We report here on the application of a tetracycline (Tet)-inducible promotor to create vectors allowing "exogenous activation" by a drug. Methods and Results: In order to reduce background activation of the transgenes, PL-antisense-RNA (Plas) and PL-sense-mRNA (PLs) used as control, respectively, were cloned downstream of a Tetracyclin Response Element (TRE) in a first vector (Ad5TRE-PLas). A second vector (Ad5-rtTA) expresses a reverse tetracyclin Transactivator (rtTA). Analogous two-vector systems were developed for luciferase (Luc). The Tet-systems were compared to standard CMV vectors in neonatal rat cardiomyocytes (CMCs). PL in CMCs, Ca2+ uptake activity in CMC homogenates, and Ca2+ transients in intact CMCs, were measured using standard methods, Luc activity by luminometry. More than 85 % of CMCs were routinely expressing both TRE-transgene and rtTA vector. In the TRE-Luc system, basal transgene activity in the absence of Tet was not significantly above the background, whereas at 400 ng/ml Tet, Luc activity was 46 % above that of the CMV vector. Under maximal stimulation, the TRE-PLas vector had similar effects as the CMV-Plas vector upon dose-and time-dependent reduction of PL-mRNA and protein, Ca2+ uptake activity, and Ca2+ transients. Both TRE-PLs vector and TRE-Plas without Tet had no significant effects. Conclusions: Pharmacological control of vector-based PL-antisense-RNA expression may become a more flexible gene therapy approach to the improvement of diastolic dysfunction than the previously used CMV and ANF promotor vectors, allowing rapid adaptation to the hemodynamic situation.

ORAL CONTRIBUTIONS

874 Heart Failure Insights From Ejection Fraction

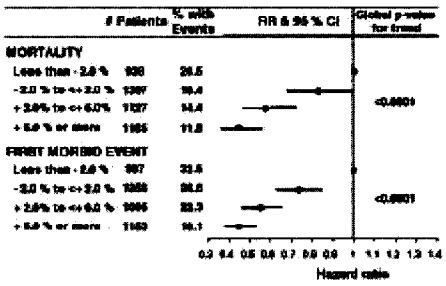
Wednesday, April 02, 2003, 8:30 a.m.-10:00 a.m.
McCormick Place, Room S403

8:30 a.m.

874-1 Relation Between Changes in Ejection Fraction Over Time and Subsequent Mortality and Morbidity in Val-HeFT

Inderjit S. Anand, Maylene Wong, Riberto Latini, Lloyd Fisher, Yann Tong Chiang, Jay N. Cohn, on behalf of the Val-HeFT Investigators, VA Medical Center and University of Minnesota, Minneapolis, MN, Istituto di Ricerche Farmacologiche "Mario Negri", Milano, Italy

Background: Left ventricular remodeling is considered an important determinant of progression of heart failure (HF), and ejection fraction (EF) has consistently been shown to predict mortality and morbidity (M&M). Whether changes in EF over time correspond to subsequent changes in M&M has not been studied. Such a relationship would further strengthen the role of LV remodeling as a surrogate for events. Val-HeFT evaluated the efficacy of the ARB valsartan in patients with moderate and severe HF and measured EF in over 5000 patients at baseline and over 4500 patients during follow-up, and provided data for this study. Methods: Change in echocardiographically derived EF from baseline (BL) to 4, 12, and 24 months were analyzed in subgroups by quartiles for subsequent M&M. Risk ratio (RR) and 95% CI were calculated for M&M in all patients, irrespective of treatment using Cox regression, adjusting for baseline EF. Results: BL EF in quartiles showed a significant quartile-dependent increase in M&M risk (data not shown). Changes in EF from BL to 4 months also showed a significant and corresponding change in M&M. As compared with patients who had no or little change in EF over time (quartile 2), patients whose EF decreased (quartile 1) showed a significant increase in RR for M&M, whereas patients with an increase in EF (quartiles 3 & 4) showed a decrease in RR for M&M. Similar findings were seen for changes in EF at 12 and 24 months. Conclusions: These data further reinforce the role of EF as a significant surrogate marker in HF.



8:45 a.m.

874-2 Hemodynamic Mechanisms of Ejection Fraction Improvement With Chronic Carvedilol Treatment in Heart Failure: Insights From Three-Dimensional Echocardiography

Mathew S. Maurer, Daniel Burkhoff, Lyna El-Khoury Coffin, Donald L. King, Norma Medina, Madeline Yushak, Jonathan D. Sackner-Bernstein, Columbia University, New York, NY, St. Luke's-Roosevelt Hospital, New York, NY

Background. Carvedilol is known to improve ejection fraction in patients with CHF but the relative contributions of heart rate reduction, afterload reduction and positive inotropism to this beneficial effect are unknown.
Methods. 32 patients (64±12 years old, 20 with idiopathic and 12 with ischemic cardiomyopathy) with NYHA II-III underwent serial three dimensional echocardiogram (3DE) at baseline and 4 months after initiation of carvedilol. 3DE derived measures of LV volumes and mass were coupled with sphygmomanometic measurements of arterial pressures yielding estimates of chamber contractility (end-systolic pressure-volume ratio, Res, where Pes˜SBP*0.9), effective arterial elastance (Ea, ~TPR*60/HR) and ventricular vascular coupling (Ea/Res).
Results. EF increased by 6 ± 8 percentage points (from 26±8% to 32±9%, p<0.0001). The change in EF was correlated with changes in stroke volume (r=0.86, p<0.0001) but not with changes in end diastolic volume (r=.09, p=NS). Pressure-volume based analysis show that Res increased from 0.72±0.39 to 0.80±0.36 mmHg/ml (p=0.03). Ea decreased from 2.1±0.6 to 1.7±0.5 mmHg/ml (p<0.001), which was due to a significant decrease in HR (from 83±11 to 65±8 bpm, p<0.001) with no significant change in TPR (1.3±0.4 vs 1.3±0.4 ml/mmHg/min). Ea/Res improved from 3.4±1.5 to 2.5±1.3 (p<0.001).
Conclusions: Improved chamber contractility (indexed by increased Res) and decreased