waveform to accurately match the aortic waveform (Figure), and (ii) preload reduction achieved with phase I of Valsalva maneuver (VM). In 10 normal healthy male subjects aged 34.7 ± 5.8 (mean ± SD), we studied the reproducibility of this system during four consecutive preload reduction runs.

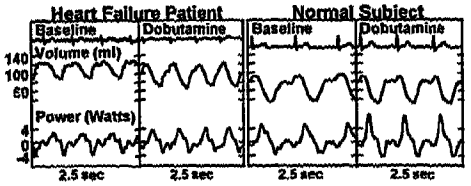
Results: VM reduces preload (end diastolic area: from 12.7 ± 1.8 to 10.3 ± 1.6 cm^2, $p < 0.001$, ANOVA), end systolic pressure (from 99.1 ± 14.8 to 89.7 ± 17.5 mmHg, $p < 0.001$) and LV work (stroke force: from 653.0 ± 165.3 to 454.6 ± 117.0 mmHg.cm^2, $p < 0.001$). Of the two measures of LV systolic function, the end systolic pressure-area relationship was highly nonlinear (r^2: 0.54 ± 0.28 vs 0.87 ± 0.17, $p < 0.001$) and exhibited higher variability (SD/mean) than the stroke force-end diastolic area relationship for both slope (0.31 vs 0.11) and intercept (0.46 vs 0.22).*Conclusion:* This totally non-invasive method should be useful in assessing the systolic LV function repeatedly in clinical settings.

11:45

708-6 Preload-Adjusted Maximal Power Using Echocardiographic Automated Border Detection to Assess Left Ventricular Function

Christine M. Mahler, William E. Katz, Srinivas Murali, Marc D. Feldman, John Gorcsan III. *University of Pittsburgh, Pittsburgh, PA*

Preload-adjusted maximal power (PAMP) has recently been validated as a load insensitive measure of LV function. The objective was to estimate PAMP in pts with severe congestive heart failure (CHF) and normal subjects using automated echocardiographic LV volume and simultaneous noninvasive arterial pressure. Ten CHF pts, aged 45 ± 10 yrs, (LVEF 21 ± 6%) and 10 normal subjects, aged 32 ± 3 yrs, (LVEF 56 ± 5%) were studied at baseline and with 10 μg/kg/min dobutamine infusion. LV volume by 4-chamber view Simpson's rule and arterial pressure by a finger cuff photoplethysmograph were acquired on-line. The first derivative of LV volume was multiplied by pressure to estimate maximal LV power, then adjusted by dividing by end-diastolic volume squared. Volume and power examples are shown.



Baseline PAMP was 0.84 ± 0.63 W/ml^2 in CHF pts vs. 4.18 ± 1.64 W/ml^2 in normals ($p < 0.001$). PAMP increased with dobutamine to 1.55 ± 1.56* in CHF pts and 8.39 plusmn; 3.73# W/ml^2 in normals (*$p < 0.05$ vs. baseline & normal, #$p < 0.001$ vs. baseline). PAMP using echo automated border detection has potential to assess LV function.

709 Molecular Analysis of Left Ventricular Hypertrophy and Remodeling

Monday, March 25, 1996, 10:30 a.m.–Noon
Orange County Convention Center, Room 222

10:30

709-1 Echocardiographic Assessment of Left Ventricular Systolic Function in Transgenic Mice With Cardiac Specific Over-Expression of Phospholamban

Brian D. Holt, Zia U. Khan, Nancy Ball, Vivek J. Kadambi, Evangelia G. Kranias, Richard A. Walsh. *University of Cincinnati, Cincinnati, OH*

In vitro studies have demonstrated that dephosphorylated phospholamban (PLB) is an inhibitor of the cardiac sarcoplasmic reticulum (SR) calcium ATPase (Ca^{2+} ATPase), and that phosphorylation of this protein by catecholamine-mediated stimulation of cyclic AMP-dependent protein kinase relieves this inhibition and facilitates reuptake of calcium by the SR. To determine the effects of PLB on *in vivo* LV systolic function and assess the stoichiometry between PLB and the SR Ca^{2+} ATPase, 15 transgenic mice with cardiac specific overexpression of phospholamban (PLBOE) driven by the α-myosin heavy chain promoter, and 16 wildtype age-matched controls (CON) were studied under light anesthesia with 2D-directed M-mode and Doppler using a 9 MHz imaging and 5–7.5 MHz Doppler transducer (Interspec-ATL CX 200). LV shortening fraction (SF), heart rate-corrected velocity of circumferential shortening (Vcf_c), peak aortic velocity (AoV) and mean aortic acceleration (Acc) were compared at baseline (BASE) and after intraperitoneal isoproterenol (ISO) injection (2 μg/gm):

	CON		PLBOE	
	BASE	ISO	BASE	ISO
HR (bpm)	292 ± 87	452 ± 69*	280 ± 98	431 ± 95*
SF (%)	44 ± 5	66 ± 5*	35 ± 6†	64 ± 4*
Vcf_c (circ/s)	6.2 ± 1.9	11.5 ± 2.1*	4.2 ± 1.0†	10.4 ± 2.1*
AoV (cm/s)	71.3 ± 12.9	88.5 ± 17.7*	66.5 ± 8.4	92.5 ± 13.0*
Acc (m/s^2)	3.7 ± 1.5	6.0 ± 2.2*	2.9 ± 0.9	6.5 ± 1.6*

Data are mean ± SD; *$p < 0.05$ vs. BASE, †$p < 0.05$ vs. CON

With ISO, the percent increase in SF, Vcf_c and mean acceleration were significantly greater in PLBOE than CON. We conclude that overexpression of phospholamban: 1) decreases basal LV systolic function, indicating that a fraction of the sarcoplasmic reticulum Ca^{2+} ATPase in wildtype mice is not under regulation by phospholamban, and 2) enhances inotropic, but not chronotropic sensitivity to β adrenergic stimulation.

10:45

709-2 Rapid Coordinate Upregulation of Nuclear and Mitochondrial Gene Expression in Response to Cardiac Load

Gregg T. Schuyler, Terrence X. O'Brien, John D. Rozich, Robert L. Kent, Diane E. McDermott, Paul J. McDermott, George Cooper IV, Donald R. Menick. *Gazes Cardiac Research Institute and Med. Univ. of SC, Charleston, SC 29425*

To identify important transcripts upregulated during acute right ventricular (RV) pressure overload in the adult cat, differential hybridization was utilized. A modified Swan-Ganz catheter was used to partially obstruct the RV outflow, under flouroscopy, creating a RV pressure overload. The balloon was positioned such that the systemic pressure was unchanged resulting in a same animal control normally loaded left ventricle (LV). cDNA libraries construced from RV and LV were differentialy hybridized to select for clones present in the RV but absent in the LV. One upregulated clone, confirmed by Northern blot hybridization, corresponded to the mitochondrial F_1 ATPase subunit Fo_8 whose sequence is encoded in the mitochondrion (Mt). This upregulation was also demonstrated in related Mt encoded transcripts, cytochrome *b* and cytochrome oxidase subunit II. To demonstrate a coordinate upregulation of mitochondrial respiratory proteins which are encoded in the nucleus, the levels of transcripts of cytochrome C, cytochrome oxidase subunit IV, and mitochondrial RNA processing RNA were analyzed. Again a rapid upregulation in response to acute hemodynamic load was observed. This *in vivo* effect was then tested *in vitro* in isolated neonatal rat ventricular cardiocytes treated with phenylephrine (PE) stimulation. Similar changes in the mRNA levels were seen after 1–2 hr. At the protein level, Western analysis of cytochrome *c* showed a significant increase within 3 days of PE stimulation. Additionally, to correlate the neonatal cell studies with adult cells, feline adult cardiocytes were stretched for 1 hour on a deformable membrane, with similar results. These studies suggest both *in vivo* and *in vitro*, a cardiac regulatory mechanism that responds to hypertrophic stimuli with a rapid *coordinate* upregulation of nuclear and mitochondrial genes encoding the oxidative phosphorylation components.

11:00

709-3 Determinants of the Variability of Left Ventricular Hypertrophy in Patients With Hypertrophic Cardiomyopathy

Winifred Kelsey, Antoine Abchee, Marcel Lechin, Robert Roberts, Ali J. Marian. *Baylor College of Medicine, Houston, TX*

The hallmark of phenotypic expression of Hypertrophic cardiomyopathy (HCM) is left ventricular hypertrophy (LVH). However, the magnitude and extent of LVH in patients with HCM, even in those with the same underlying mutation, are variable. A number of genetic and environmental factors are likely to influence the phenotypic expression of LVH in patients with HCM. We have previously shown that ACE genotypes account for 5–10% of the variability of the extent and magnitude of LVH in patients with HCM. In this study we determined the influence of gender, age, height, weight, body mass index, angiotensinogen (AGT) genotypes T174M, and M235T, angiotensin II receptor Ia (ATR1a) genotypes on left ventricular mass index (LVMI) calculated by area-lenghth method, and extent of hypertrophy determined using a semiquantitative point score (Score 1–10). Multiple regression analysis showed that only gender and ACE genotypes were correlated with LVMI and extent of LVH. LVMI was greater in male (n = 61) than in female (n-47) patients with HCM (145.96 ± 34 vs. 129.42 ± 33, $p = 0.013$). Similarly, male patients had more extensive hypertrophy than female patients (socre 6.4 ± 2.2 vs. 5.3 ± 1.9, $p = 0.009$). Gender accounted for 4.8% and 5.4% of the

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variability of the LVMI and extent of LVH in patients with HCM. Together, gender and ACE genotypes accounted for 5.4, and 10.1% of the variability of LVMI and extent of hypertrophy. Neither AGT genotypes (T174M and M235T) nor AT IIRa genotypes had any influence on LVMI and score of hypertrophy. Thus, gender and ACE genotypes account for a fraction of the variability of the phenotypic expression of LVH in patients with HCM. The greater magnitude of hypertrophy in male patients has prognostic implications. Previous reports have shown than male patients with HCM are more likely to be victim of sudden cardiac death than females. Our results suggest that the reported higher incidence of sudden cardiac death in male patients is due in part to two genetic factors: male gender and ACE genotype. Mutations causing HCM are associated with more extensive ventricular hypertrophy when it occurs in the male as opposed to female gender. The combination of male gender and the ACE genotype DD may predispose to a higher incidence of sudden cardiac death in HCM.

11:15

709-4 Analysis of SERCA2 Promoter Activity by Direct Gene Transfer Into Rat Hearts

Teruhiko Aoyagi, Yasunobu Hirata, Shin-ichi Momomura, Muthu Periasamy. *University of Tokyo, Tokyo, Japan; University of Cincinnati, Cincinnati, Ohio*

The cardiac sarcoplasmic reticulum Ca^{2+}ATPase (SERCA2) protein and mRNA were known to be depressed in many pathological situations such as pressure-overload hypertrophy. Molecular mechanisms controlling transcription of SERCA2 are yet to be investigated, especially in in vivo setting. Here, SERCA2 promoter/luciferase (Luc) reporter constructs were injected into a left ventricular apex of adult rat hearts. Transcription level of SERCA2 was evaluated by Luc assay in myocardial tissue homogenate. Transfer efficiency was examined by the expression of co-injected constitutively active β-gal gene. First, we injected the construct, -1110Luc consisting of -1110bp upstream region of SERCA2 promoter and Luc reporter. Myocardial Luc activity was 256 ± 119*, 44 ± 26*, 40 ± 8*, 31 ± 15*, 13 ± 1*; times higher than background at 1, 2, 3, 4 and 5 weeks after the injection, respectively. The expression of constructs with deletions in SERCA2 promoter; -658Luc, -284Luc -267Luc, -72Luc were examined one week after the injection. Luc activity were 61 ± 11*, 24 ± 5§, 20 ± 2 and 0.05 ± 0.01§ folds of that in -1110Luc-injected rats, respectively. In compensated pressure-overload hypertrophy induced by the DOCA-salt method, myocardial expression of the -1110Luc construct was similar (75.2%) to that in Sham controls at one week. In contrast, a severe ascending aortic banding (mean pressure gradient: 38 mmHg under anesthesia, mortality: 40%) resulted in depressed expression (35.7%* of Sham controls) at one week. Thus, the expression of SERCA2 promoter injected in vivo was maintained in compensated hypertrophy, but depressed in severe decompensated pressure-overload. (§ $p < 0.05$, *$p < 0.01$)

11:30

709-5 Serial Echocardiographic Assessment of Left Ventricular Function and Gene Analysis of Contractile Protein and Collagen After Myocardial Infarction

Akihisa Hanatani, Minoru Yoshiyama, Iku Toda, Kaname Akioka, Masakazu Teragaki, Kazuhide Takeuchi, Jyunichi Yoshikawa. *First Department of Internal Medicine, Osaka City University Medical School, Osaka, Japan*

The purposes of this study were to assess the effect of angiotensin II type 1 receptor antagonist (TCV-116; 10 mg/kg/day) on left ventricular function by using echocardiography and to analyze gene expression of contractile protein and collagen turnover after myocardial infarction. Myocardial infarction was made by ligation of coronary artery in Wistar rats and TCV-116 was administered after surgery. On 1, 2 and 3 weeks after myocardial infarction, left ventricular geometry was measured and mRNAs in cardiac tissue were analyzed. LV end-diastolic dimension (LVDd) and fractional shortening (FS) on 1 week were 9.1 ± 0.6 mm and 16 ± 4%, respectively, and LVDd on 3 weeks increased to 11.1 ± 0.8 ($p < 0.05$) and FS decreased to 11.5 ± 5% ($p < 0.01$). TCV-116 prevented an increase of LVEDV (9.2 ± 0.5 mm; $p < 0.01$) and improved FS (17 ± 5%; $p < 0.01$) on 3 weeks. The left ventricular weight significantly increased at 3 weeks after myocardial infarction (2.41 ± 0.07 g/kg), which was prevented by TCV-116 treatments (2.09 ± 0.11 g/kg; $p < 0.01$). The gene expressions of β-myosin heavy chain (MHC), atrial natriuretic peptide (ANP), transforming growth factor (TGF)-β1, collagen types I, III, IV, and matrix metalloproteinase (MMP)-2 in the non-ischemic left ventricular myocardium increased by 3.0-, 6.7-, 1.6-, 7.9-, 4.1-, 2.1-, and 1.4-fold on 1 week, respectively ($p < 0.01$). α-skeletal actin mRNA increased on 3 weeks by 2.9-fold ($p < 0.01$). TCV-116 significantly suppressed the increased gene expressions of β-MHC, α-skeletal actin and ANP. On the other hand, TGF-β1, fibronectin, collagen I, III, IV, and MMP-2 mRNAs in the ischemic region significantly increased by 6.6, 12.0, 26.2-, 12.7, 5.6-, and 2.2-fold, respectively. TCV-116 did not affect TGF-β1, fibronectin, and collagen I, III, IV mRNAs in ischemic and nonischemic regions However, it increased MMP-2 mRNAs on 1 week in both regions. In conclusion, TCV-116 prevented progressive cardiac dysfunction by left ventricular remodeling after myocardial infarction and the change of the properties of contractile protein. TCV-116 accelerated the gene expression of MMP-2, which may be contributed to prevent cardiac fibrosis.

11:45

709-6 Dilated Cardiomyopathy Associated With Deficiency of the Cytoskeletal Protein Metavinculin

Masato Maeda, Emma Holder, Brian Lowes, Roger D. Bies. *Division of Cardiology, VA Medical Center and University of Colorado Health Sciences Center, Denver, CO.*

Metavinculin is a cardiac and smooth muscle specific isoform of the cytoskeletal protein vinculin, which connects actin filaments to the sarcolemma in the cell. Metavinculin contains a 3' alternating spliced exon encoding an additional 68 amino acids near the C terminus of vinculin, and is localized to the intercalated discs and costameres of cardiac muscle and membrane associated plaques in smooth muscle. In this report we describe a patient with idiopathic dilated cardiomyopathy (IDC) and absence of cardiac metavinculin. Polymerase chain reaction (PCR) analysis demonstrated absence of the metavinculin mRNA transcript isoform in cardiac tissue from a subject with IDC compared to controls. In contrast, the vinculin transcript was present in both our IDC subject and controls. PCR of genomic DNA from the IDC subject demonstrated the presence of an intact metavinculin exon. Western blot analysis demonstrated absence of metavinculin but not vinculin in tissue homogenate from the IDC heart. Immunostaining of cardiac vinculin demonstrated disorganized intercalated disc structures in our subject compared to those of cardiomyopathic controls. The metavinculin deficiency in this subject does not appear to be secondary to a generalized process in diseased heart tissue. Twenty eight human heart specimens were analyzed including three normal controls, five ischemic cardiomyopathy, one X-linked cardiomyopathy, and nineteen subjects with idiopathic dilated cardiomyopathy. Metavinculin deficiency was only detected in the one subject described in this report. Cardiac expression of vinculin and other cytoskeletal proteins associated with both the Z disc (α-actinin), and the membrane (dystrophin) were normal. These results demonstrated a deficiency of metavinculin mRNA and protein in a subject with dilated cardiomyopathy, presumably due to a defect in mRNA alternative splicing.

710 Hypertension: Basic and Clinical

Monday, March 25, 1996, 10:30 a.m.–Noon
Orange County Convention Center, Room 230C

10:30

710-1 Does ACE Genotype Influence the Development of Left Ventricular Hypertrophy in Untreated Hypertension

Jamil Mayet, Kevin P. J. O'Kane, Manjit Shahi, Mick A. Ozkor, Heather A. Johnstone, Neil R. Poulter, Peter S. Sever, Rodney A. Foale, David J. Webb, Simon A. McG. Thom. *St. Mary's Hospital, London and Western General Hospital, Edinburgh, UK*

The presence of LVH predicts an increased morbidity and mortality in patients with hypertension. The ACE gene has been found to have 3 genotypes and the D allele has been found to be associated with a number of cardiac diseases. Our hypothesis was that the increased plasma ACE levels present in the DD genotype group may result in an increased tendency to develop LVH either directly or indirectly via the effects of BP. We have addressed this issue in a group of 69 previously untreated hypertensives. Each underwent 2D guided M-mode and Doppler echocardiography as well as ACE genotyping.

Each genotype group had similar male/female ratios (II 10/6, ID 22/10, DD 14/7), ages (II 47 ± 3, ID 48 ± 3, DD 47 ± 3 years), BP (II 163 ± 4 / 96 ± 3, ID 167 ± 4 / 98 ± 2, DD 161 ± 4 / 96 ± 3 mmHg) and cardiac structural and functional parameters [left ventricular mass index (LVMI), II 136 ± 9, ID 132 ± 7, DD 131 ± 8 g/m^2; E/A ratio, II 1.07 ± 0.1, ID 1.07 ± 0.09, DD 1.03 ± 0.07; isovolumic relaxation time, II 105 ± 7, ID 103 ± 4, DD 97 ± 6 ms]. In each group there was a significant relationship between systolic BP and LVMI (II $r = 0.56$, $p = 0.02$, ID $r = 0.40$, $p = 0.02$, DD $r = 0.49$, $p = 0.02$. The difference in these correlations between the 3 genotype groups was not significant.

This would suggest that ACE genotype does not exert a large influence on