946-108 **High Resolution Imaging of Human Arterial Walls Via Optical Coherence Tomography**

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Optical coherence tomography (OCT) is a recently developed technology which can perform micron scale, cross sectional tomographic imaging. OCT performs imaging using interferometric optical ranging of low coherence infrared light or ultrashort laser pulses which are backscattered from tissue. This technique is analogous to the reflected acoustical wave measurements of conventional B mode ultrasound. In this study, two dimensional cross sectional images were generated of *in vitro* human aorta and coronary artery sections obtained postmortem. The microstructure of normal and atherosclerotic tissue was displayed as a false color or grey scale image. The bandwidth of the photodiode source (1300 nm) allowed a resolution of 20 microns, almost 10 times greater than conventional intravascular ultrasound. Different morphologies including fatty, fibrous, and water based tissue, were well differentiated in the images and corresponded to histologic sections. Imaging was possible with up to 1 mm penetration into the tissue with little attenuation from heavy calcification. The typical image acquisition time was in the range of 2–3 seconds. A more detailed analysis of the optical properties of relatively uniform, structurally distinct tissues, such as adipose, skeletal muscle, and tendon, was performed to further confirm the contrast between fat, muscle, and connective tissue. The contrast ratios of muscle, tendon, and fat, were measured to be 1:78:22, respectively. The effective refractive indices and optical penetration depths in different tissue types were also measured. OCT is a promising new technology for high resolution "optical biopsy". It does not require direct contact with the vessel wall and could be performed via a catheter integrated with a relatively inexpensive fiberoptic bundle. Future studies utilizing currently available femtosecond lasers are expected to both increase the resolution of OCT, which is dependent on bandwidth or pulse duration, to ≈3–4 microns and allow visualization of structures deeper into tissues.

946-109 **Measurement of Right Ventricular Mass of Small and Large Hearts by Quantitative Volume-Rendered Three-dimensional Echocardiography Without the Use of Geometric Assumptions: Experimental Validation**

Lissa Sugeng, Qi-Ling Cao, Alain Delabays, Natesa Pandian. *Tufts-New England Medical Center, Boston, Massachusetts*

Despite a need for a method to measure right ventricular (RV) mass, the unusual geometry of the RV has made application of common imaging approaches unreliable. Recent developments in volume-rendered three-dimensional echocardiography (3DE) suggest that this technique could not only depict the anatomy but also aid in quantitation. Previously we have shown the utility of 3DE in obtaining RV cavity volumes. In this study we explored the accuracy of volume-rendered 3DE in estimating RV mass. Using a rotational imaging approach with a 5 MHz transducer, we obtained 2D images of 15 animal (dogs/pigs/cows) hearts of various sizes (simulating pediatric and adult hearts) in a watertank at every 2 degrees over a 180 degree span. Using various steps of 3D image processing (digital reformatting, thresholding and segmentation), 3DE images of the hearts were developed. Electronically, we sectioned each heart into 30 equidistant parallel slices that displayed all portions of the RV. The RV free wall volume was measured in each. Cumulative volume from these slices multiplied by myocardial density provided total RV wall mass. In addition, interobserver variability between 2 observers in RV mass estimation by 3DE was also assessed. These data were compared to the actual dry weight of the anatomic RV free wall measured by a third observer. *Results:* The actual RV mass of the anatomic hearts was (M + SD) 72 ± 44 grams (range 25–175 grams); 3DE measured RV mass was 84 ± 55 grams (range 25–218). 3DE (y) derived RV mass had an excellent correlation with actual mass (x): y = 1.22x − 5.4, r = 0.98, p < 0.0001. The interobserver concordance was also excellent (r = 0.97). In addition to such quantitative data, volume-rendered 3DE yielded tissue-depiction images of the RV interior, displaying the endocardial surface, papillary muscles and moderator band, and detailed visualization of the RV inflow and outflow regions in many cut-sections. 3DE projections also allowed visualization of the interventricular septum en face. We conclude that volume-rendered 3DE can be used to derive RV mass measurements accurately without unreliable geometric assumptions. This quantitative ability coupled to the capability of displaying RV anatomy enhances the clinical potential of 3-D echocardiography.

947 **Basic Pharmacology II**

Tuesday, March 21, 1995, 9:00 a.m.–11:00 a.m.
Ernest N. Morial Convention Center, Hall E
Presentation Hour: 9:00 a.m.–10:00 a.m.

947-110 **Measurement of Adenylyl Cyclase Activity in the AV Nodal Region of the Canine Heart: Evidence for Inhibition by Adenosine and Acetylcholine**

Atsushi Sugiyama, Scott McKnite, Phi Wiegn, Wayne Adkisson, Jennifer Pennington, Keith G. Lurie. *University of Minnesota, Minneapolis, MN*

Although essential to cardiac conduction, little is known about the biochemistry underlying post-receptor adrenergic, cholinergic, and purinergic processes in the AV node. To study these mechanisms, we developed a highly sensitive fluorometric assay for cAMP and used it to characterize regional adenylyl cyclase activity (ACA) (pmol/min/mg of protein) in membrane preparations made from freeze-dried 20 micron thick micro-dissected pieces of canine (n = 6) right atrium (RA), AV nodal region (AVN), and left ventricle (LV). Adjacent sections were stained for acetylcholinesterase activity to identify the AVN. Basal and NaF-stimulated ACAs (mean ± SEM) were 7.2 ± 0.4 and 72.4 ± 7.5 in RA, 15.6 ± 1.3 and 58.8 ± 4.7 in AVN, and 6.4 ± 0.9 and 66.7 ± 5.0 in LV, respectively. Isoproterenol (10^{-7} – 10^{-4} M) increased ACA in a dose-dependent fashion and the increment of cAMP production rate was similar in three different regions. Adenosine (10^{-3} M) and carbachol (10^{-5} M) inhibited isoproterenol (10^{-6} M)-stimulated ACA as follows:

☑ Isoproterenol
☐ Isoproterenol+Adenosine
■ Isoproterenol+Carbachol
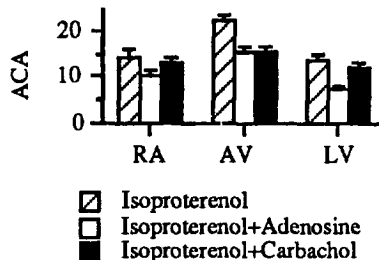
Results of this study demonstrate for the first time that (1) there are significant regional differences in ACA under basal conditions and after adrenergic, purinergic, and cholinergic stimulation in the heart, and (2) adenosine inhibits ACA in the RA, AVN and LV, whereas the inhibitory effects of cholinergic stimulation are more specific for the AVN.

947-111 **I_k Blockade Selectively Eliminates Slow Antegrade Atrioventricular Nodal Conduction in the Rabbit**

Kenneth W. Hewett, Francis K. Le, Christine A. McKay, Paul C. Gillette, David S. Buckles. *South Carolina Children's Heart Center, Medical University of South Carolina, Charleston, SC*

We studied the effects of an I_k blocker, risotilide (R) on the dynamic properties of perfused rabbit AV nodes using intermittent right atrial premature pacing and bipolar surface electrograms. R was serially perfused with 1, 3 and 6 × 10^{-6} M R for 15 minutes following pre-drug measurements.

Recovery Interval, H_1–A_2 (ms)
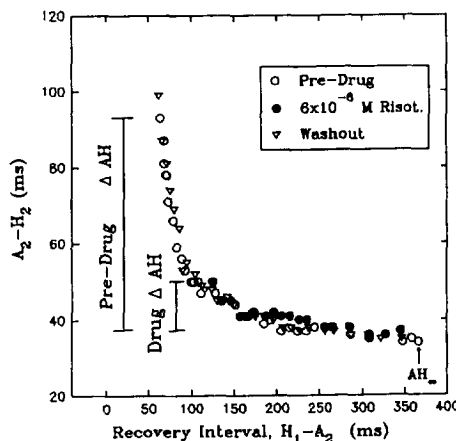
Results: Typical changes in the antegrade recovery curve produced by R are shown in the figure. There was a concentration dependent decrease in ΔAH from the pre-drug value of 56 ± 15 ms to 29.2 ± 16.9 ms and 13.7 ± 5.9 ms by 3 and 6×10^{-6} M respectively ($n = 6$). A reciprocal increase in the AV node effective refractory period was observed from 91 ± 15 ms (pre-drug) to 139 ± 16 ms (3×10^{-6} M) and 170 ± 16 ms (6×10^{-6} M). AH_∞ was unchanged at any concentration of R.

Conclusions: These results suggest that class III antiarrhythmic agents may increase action potential duration and the refractory period of the structure(s) limiting AV conduction and thereby eliminate a majority of the "slow" AV node conduction. These findings may have important implications for a pharmacologic approach to the treatment of AV node reentry tachycardias.

947-112 Has the Inotropic Effect of the Class-III Antiarrhythmic Drug Amiodarone a Frequence-dependency In Vivo?

Martin E. Beyer, Hans Martin Hoffmeister, Ludger Seipel. *Medical Hospital, Dept. III, University of Tübingen, Germany*

The prolongation of the action potential duration (APD) by class-III antiarrhythmic drugs causes in vitro a positive inotropic effect. On the other side a frequence-dependency of the APD-prolongation ("reverse use-dependence") was described. This would mean, that a heart rate reduction should have a positive effect on myocardial contractility after administration of class-III drugs.

We examined the hemodynamic effects of amiodarone (10 mg/kg, 20 mg/kg i.v.) in thoracotomized rats vs. saline controls (NaCl) without and with bradycardia. Heart rate reduction was produced by vagal stimulation (reduction about 50%). Besides measurements in the intact circulation isovolumic maximum registrations (isovol. LVSP, isovol. dp/dt$_{max}$) were performed to determine myocardial contractility.

	spontaneous heart rate			vagal stimulation	
	10 mg/kg	20 mg/kg	NaCl	20 mg/kg	NaCl
isovol. LVSP	93 ± 2	$88 \pm 1^*$	98 ± 1	$80 \pm 5^*$	100 ± 2
isovol. dp/dt$_{max}$	$81 \pm 3^*$	$73 \pm 3^*$	94 ± 3	$54 \pm 5^*$	82 ± 4
cardiac output	90 ± 7	75 ± 7	93 ± 8	57 ± 6	68 ± 4

Means $\pm$ SEM in % of preinfusion values, *p < 0.01

Conclusion: The prolongation of the action potential duration by the class-III antiarrhythmic drug amiodarone does not cause in vivo a positive inotropic effect since the cardiodepressive effects of the drug (e.g. sodium- and calcium-channel blockade) are stronger. A reverse use-dependence of the inotropism of this drug is not detectable in vivo.

947-113 Aprotinin Produces Ion Channels in Lipid Bilayers

Steven Hanish, Slava Titov, Richard Kovacs. *Department of Medical Research, Methodist Hospital, Indianapolis, IN*

Aprotinin, a 58 Amino Acid peptide protease inhibitor, is used clinically to prevent postoperative blood loss and reduce transfusion requirements in those procedures which employ extracorporeal circulation. Its mechanism of action is unknown. We have observed that Aprotinin has produced channel-like artifacts in other experimental systems. We tested the hypothesis that Aprotinin could form ion channels in lipid bilayers. Lipid bilayers were created on the tips of patch clamp pipets using phosphatidylethanolamine, phosphatidylserine and cholesterol in a 5:3:2 ratio respectively. Single channel currents were recorded in symmetric solutions of KCl + Tris/HEPES, pH = 7.2, as well as asymmetric solutions. Aprotinin was added to the bath solution. Data were digitized and recorded on video tape for off-line analysis using custom software. 8 μM Aprotinin produced typical ion channel currents in symmetric solutions of KCl at concentrations of 50 mM, 100 mM, 150 mM, and 250 mM, with resultant peak currents of 7.33 pA. The conductance of the Aprotinin channel was 31.8 pS in 250 mM KCl. Aprotinin exhibited multiple conductance levels and complex kinetics. In 250 mM potassium, the channel had a steady state Popen of 0.5. Popen was not voltage dependent. The channels were saturable, selective, and highly specific for potassium ions over sodium ions. No definite blockers of the channel have yet been identified. Our experiments show that Aprotinin produces ion channels in lipid bilayers, and this in turn may lead to better understanding this drug's mechanism of action.

947-114 Effects of Azimilide (NE-10064) on Cardiac K Channels

J.-A. Yao, G.-N. Tseng. *Columbia University, New York, NY*

To understand the cellular mechanism of action of azimilide (Azim), a novel class III antiarrhythmic agent, we studied its effects on K currents in guinea

pig and canine ventricular myocytes: slow (I_{Ks}) and rapid (I_{Kr}) delayed rectifier, inward rectifier (I_{K1}) and transient outward (I_{to}) currents. To facilitate the quantification and comparison of drug potencies, the recording conditions were designed to "isolate" these K currents from other ionic currents and from each other. In particular, since it is difficult to separate I_{Kr} and I_{Ks} under normal physiological conditions, a Na- and Ca-free external solution was used to dissect the two. This was confirmed by the selective action of dofetilide. Azim blocked I_{Ks} concentration dependently (0.2–10 μM) and reversibly. Block was potentiated by more positive V_t. At $+30$ mV, 2 μM Azim blocked I_{Ks} by $58 \pm 13\%$ ($n = 6$). Azim caused a time-dependent reduction of I_{Ks} during depolarization and slowed I_{Ks} deactivation, suggesting that block and unblock occurred mainly in the open state. Azim also blocked I_{Kr} concentration dependently (0.1–2 μM) and reversibly. At -20 mV, 1 μM Azim blocked I_{Kr} by $86 \pm 10\%$ ($n = 3$). On the other hand, I_{to} ($IC_{50} > 10 \mu$M, $n = 6$) and I_{K1} ($IC_{50} > 50 \mu$M, $n = 2$) were much less sensitive to Azim than I_{Kr} or I_{Ks}. In conclusion, blockade of both delayed rectifiers makes important contributions to the class III action of Azim.

947-115 Comparative Effects of D-Sotalol, Quinidine and Amiodarone on Dispersion of Ventricular Repolarization in the Isolated Intact Rabbit Heart

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It has been hypothesized that antiarrhythmic as well as proarrhythmic effects of antiarrhythmic drugs (AA) can be linked to changes in dispersion of ventricular repolarization (DISP). The influence of d-sotalol (D-SOT), quinidine (QUIN) and amiodarone (AMIO) was studied in isolated Langendorff-perfused rabbit hearts. Between 5–7 monophasic action potentials were recorded simultaneously from both ventricles at steady-state cycle lengths (CL) between 300 and 1200 msec and measured at 90% repolarization (APD$_{90}$). DISP was defined as APD$_{90}$max $-$ APD$_{90}$min. The protocol was repeated after infusion of D-SOT ($n = 12$, 10^{-6} M, 10^{-5} M and 5×10^{-5} M) and QUIN ($n = 8$, 10^{-6} M and 10^{-5} M). AMIO was given chronically po. for 4 weeks ($n = 9$) and compared to $n = 18$ normal hearts. DISP change compared to the respective baseline (values ranged between 20–27 msec) is shown in the table at selected CLs (all values mean $\pm$ SEM in msec, * p < 0.05). AMIO tissue levels correlated with APD duration but not with DISP.

CL	300	600	900	1200
D-SOT 10^{-6} M	$+2 \pm 3$	$+5 \pm 3$	$+4 \pm 2$	$+4 \pm 3$
D-SOT 10^{-5} M	$+11 \pm 2^*$	$+14 \pm 3^*$	$+10 \pm 3^*$	$+11 \pm 3^*$
D-SOT 5×10^{-5} M	$+18 \pm 2^*$	$+22 \pm 3^*$	$+24 \pm 3^*$	$+45 \pm 10^*$
QUIN 10^{-6} M	$+14 \pm 5^*$	$+22 \pm 6^*$	$+18 \pm 4^*$	$+28 \pm 4^*$
QUIN 10^{-5} M	$+22 \pm 5^*$	$+15 \pm 5^*$	$+29 \pm 5^*$	$+31 \pm 6^*$
AMIO	$+3 \pm 2$	-4 ± 2	-3 ± 2	$+3 \pm 3$

Conclusions: Neither of the three drugs with class III-action shows a reduction of DISP. While QUIN and D-SOT show dose-dependent increases in DISP, especially at long CLs, AMIO tissue concentrations are not related to DISP and no increase is seen at long CLs. These effects on DISP may explain the different clinical incidence of torsade de pointes between the three drugs.

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948 Heart Failure: Renal and Adrenal Characteristics

Tuesday, March 21, 1995, 9:00 a.m.–11:00 a.m.
Ernest N. Morial Convention Center, Hall E
Presentation Hour: 9:00 a.m.–10:00 a.m.

948-46 Preserved Cardiac Baroreflex Control of Renal Cortical Blood Flow in Advanced Heart Failure Patients: A Positron Emission Tomography Study

Holly R. Middlekauff, Egbert U. Nitzsche, Michele A. Hamilton, Heinrich R. Schelbert, Gregg C. Fonarow, Jaime D. Moriguchi, Antoine Hage, Saleh Saleh, G. Gary Gibbs. *UCLA Medical Center, Los Angeles, CA*

Cardiac baroreflex (CBR) control of forearm blood flow (FBF) is blunted or reversed in humans with heart failure (HF), but little is known about CBR control of renal cortical blood flow (RCBF) in HF due to technical limitations. Positron emission tomography (PET) 0-15 water is a new, precise method to measure RCBF quantitatively. We compared CBR control of RCBF and FBF (venous plethysmography) in 8 patients with HF (mean age, 47 ± 3 y, ejection fraction 0.25 ± 0.02) and 10 normal humans (mean age 35 ± 5 y) during CBR unloading with phlebotomy (450 ml). In 5 normals, cold pressor test was