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?

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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 ± 1*	98 ± 1	80 ± 5*	100 ± 2
isovol. dp/dt_{max}	81 ± 3*	73 ± 3*	94 ± 3	54 ± 5*	82 ± 4
cardiac output	90 ± 7	75 ± 7	93 ± 8	57 ± 6	68 ± 4

Means ± 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

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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

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To understand the cellular mechanism of action of azimilide (Azim), a novel class III antiarrhythmic agent, we studied its effegts on K currents in guinea pig and canine ventricular myocytes: slow (I_{Ks}) and rapid (I_{Kr}) delayed rectifier, inward rectifier (I_{KI}) 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 ± 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 ± 10% (n = 3). On the other hand, I_{to} (IC_{50} > 10 μM, n = 6) and I_{KI} (IC_{50} > 50 μM, 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 ± 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 ± 3	+5 ± 3	+4 ± 2	+4 ± 3
D-SOT 10^{-5} M	+11 ± 2*	+14 ± 3*	+10 ± 3*	+11 ± 3*
D-SOT 5 × 10^{-5} M	+18 ± 2*	+22 ± 3*	+24 ± 3*	+45 ± 10*
QUIN 10^{-6} M	+14 ± 5*	+22 ± 6*	+18 ± 4*	+28 ± 4*
QUIN 10^{-5} M	+22 ± 5*	+15 ± 5*	+29 ± 5*	+31 ± 6*
AMIO	+3 ± 2	−4 ± 2	−3 ± 2	+3 ± 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.

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

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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

used as a strong, non-baroreflex mediated stimulus to vasoconstriction.

Results: Phlebotomy decreased central venous pressure ($p < 0.001$), but did not change mean arterial pressure or heart rate in HF patients or controls. The major findings of the study are: 1) At rest, RCBF is markedly diminished in HF vs normals (2.4 ± 0.1 vs 4.3 ± 0.2 ml/min/g, $p < 0.001$). 2) In normal humans during phlebotomy, FBF decreased substantially (basal vs phlebotomy: 3.3 ± 0.4 vs 2.6 ± 0.3 ml/min/100 ml, $p = 0.02$), and RCBF decreased slightly, but significantly (basal vs phlebotomy: 4.3 ± 0.2 vs 4.0 ± 0.3 ml/min/g, $p = 0.01$). 3) The small magnitude of reflex renal vasoconstriction is not explained by the inability of the renal circulation to vasoconstrict since the cold pressor stimulus induced substantial decreases in RCBF in normals (basal vs cold pressor: 4.4 ± 0.1 vs 3.7 ± 0.1 ml/min/g, $p = 0.003$). 4) In humans with heart failure during phlebotomy, FBF did not change (basal vs phlebotomy: 2.6 ± 0.3 vs 2.7 ± 0.2 ml/min/100 ml, p = NS), but RCBF decreased slightly but significantly (basal vs phlebotomy: 2.4 ± 0.1 vs 2.1 ± 0.1 ml/min/g, $p = 0.01$). Thus, in patients with heart failure, there is an abnormality in cardiopulmonary baroreflex control of the forearm circulation, but not the renal circulation.

Conclusion: This study 1) shows the power of PET to study physiologic and pathophysiologic reflex control of the renal circulation in humans, and 2) describes the novel finding of selective dysfunction of cardiac baroreflex control of the forearm circulation, but its preservation of the renal circulation, in patients with heart failure.

948-47 Natriuretic Peptides and cGMP: Insights into Their Functional Role in Heart Failure

Gordon W. Moe, Etienne A. Grima, Norman L.M. Wong, Robert J. Howard. *U. of Toronto, Toronto, Ontario*

Plasma atrial and brain natriuretic peptide levels (ANP and BNP) are elevated in heart failure (HF) but their functional significance is unclear. Accordingly, we measured plasma ANP, BNP (pg/ml), and their second messenger cGMP (pmol/ml) together with PCWP (mmHg) in 8 dogs, before (BNP-0) and during infusion of synthetic BNP (25, 50 and 100 ng/kg/min; BNP-25, BNP-50 and BNP-100), at control (C) and HF after 3 wks of pacing. Mean data and pooled SD (pSD) from analysis of variance are shown.

	PCWP		BNP		ANP		cGMP	
	C	HF	C	HF	C	HF	C	HF
BNP-0	9	31†	12	57†	84	745†	14	16
BNP-25	8	29	273*	521*	99	910*	28*	19
BNP-50	6*	28*	611*	1053*	93	884*	31*	24*
BNP-100	6*	26*	769*	1230*	104*	871*	32*	24*
pSD	2	4	250	284	33	149	4	6

*$p < 0.05$ vs BNP-0, †$p < 0.05$ vs C

At HF, the elevated endogenous BNP and ANP (BNP-0) were unable to increase cGMP. Exogenous BNP induced a marked rise in plasma BNP, and also increased plasma ANP and cGMP at both C and HF. The observation that a higher plasma BNP level than that produced endogenously in HF is needed to elevate cGMP suggest that the increased circulating BNP may not play a functional role, although an important paracrine role cannot be ruled out. Furthermore, the fact that exogenous BNP increases plasma ANP despite falling PCWP suggests that BNP may compete with ANP for the clearance (NPR-C) receptors.

948-48 High-dose Lisinopril is More Effective than Low-dose in Suppressing Aldosterone in Patients with Chronic Heart Failure

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The optimal dose of ACE inhibitor for the treatment of heart failure is highly controversial and it is uncertain whether the neurohormonal suppression produced by these drugs is dose-dependent. We therefore compared the effects of low-dose and high-dose lisinopril therapy on plasma levels of aldosterone, which causes several important adverse effects including hypokalaemia, hypomagnesaemia and stimulation of myocardial fibrosis. 15 patients with LVEF < 45%, on chronic ACE inhibitor therapy, were given lisinopril 5 mg o.d. and 20 mg o.d. for two weeks each in a randomised order. At the end of each treatment period plasma aldosterone levels were measured, after 30 minutes bed rest, at 6 hours (peak effect) and 22 hours (trough effect) after the dose of lisinopril. The levels of aldosterone on each dose at these timepoints were compared using ANOVA.

Lisinopril Dose	Mean Plasma Aldosterone Concentration(pg/ml)	
	Peak (6 hours)	Trough (22 hours)
5 mg o.d.	163.0	186.7
20 mg o.d.	98.4	118.4

Plasma aldosterone levels were significantly lower during high-dose than during low-dose lisinopril treatment at both 6 hours ($p < 0.03$) and at 22 hours ($p < 0.04$).

Conclusion: The suppression of aldosterone activity during chronic ACE inhibitor therapy in patients with heart failure is dose-dependent. This provides further evidence that ACE inhibitors should be routinely prescribed in high doses.

948-49 Prognostic Importance of the Oxidation Products of Catecholamines (Adrenochromes) in Patients with Heart Failure

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In experimental models of heart failure (HF) free-radical formation has been shown to be increased and considered to be involved in pathophysiologic mechanisms leading to progressive ventricular dysfunction. Adrenochromes, the products of oxidation of norepinephrine (NE) are thought to reflect oxidative stress (amount of free-radical formation) and to be more cytotoxic than NE. To investigate the importance of adrenochromes in HF, we measured adrenolutins (AL), stable breakdown products of adrenochromes, along with plasma NE, and N-terminal atrial natriuretic factor (ANF) in 257 patients at baseline entering the PROFILE study of flosequinan. We then related these variables to mortality (21 — progressive HF, 27 — sudden cardiac deaths and 9 — other causes) over a 20-month period.

	ANF LVEF (pmol/l)	NE (pg/ml)	AL (ng/ml)	Age (yrs)	(%)
Alive (200)	2739	406	43	65	23
Dead (57)	3121	503*	80*	65	20*

*$P < 0.05$ Alive versus Dead

In a multivariant model including these variables, as well as NYHA class, gender, and treatment groups (flosequinan or placebo), AL was the most significant predictor of death ($P = 0.005$). In patients dying with progressive HF or of other causes, AL was a better predictor of death than NE, while in patients dying of a sudden cardiac death, NE was a better predictor. Thus, measurement of AL appears to be a new prognostic factor in patients with HF. These results suggest a role for free-radical formation in the pathophysiology of HF and provide a new opportunity for therapy.

948-50 Dissociation Between Changes of Plasma Volume and its Neurohumoral Determinants Post-HeartMate Implantation in Heart Failure Patients

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Congestive heart failure is associated with blood volume expansion which by itself increases the burden on the heart. High PV with congestive heart failure has been attributed to stimulation of the renin-aldersterone system and AVP. The use of left ventricular assist devices as bridges to heart transplantation has increased the survival of these patients during this critical period. We hypothesized that improvement of cardiac mechanical function by HeartMate (left ventricular assist device) is associated with a normalization of volume load secondary to normalization of neurohumoral determinants of plasma volume. To assess this hypothesis, we studied 15 patients (13 M: 2 F; age 50 ± 9 yrs) with end stage heart failure who were cardiac transplant candidates, before and after HeartMate 1000 (HM) implantation. We measured plasma volume (PV, RISA), and plasma levels of atrial natiuretic hormone (ANF), aldosterone (PA), renin (PRA), and arginine vasopressin (AVP), sequentially at pre HM, and post HM (weeks 2, 4 and 8). *Results:*

	Pre HM	W2	W4	W8
PV	117 ± 22	123 ± 17	119 ± 19	111 ± 12
ANF	276 ± 199	199 ± 62	180 ± 110	141 ± 66
PA	53 ± 52	16 ± 9	17 ± 18	14 ± 9*
PRA	46 ± 25	7 ± 3*	8 ± 7*	11 ± 18*
AVP	5.2 ± 5	0.8 ± 1	0.6 ± 0.7*	1.1 ± 1*

$\bar{x}$ ± SD, P (paired t): * < 0.05 vs pre HM

Conclusions: The reduction of PV, PRA, PA and AVP occurs earlier than the reduction of plasma volume and ANF after HeartMate, possibly due to decreased pulmonary congestion and improved renal perfusion. The reduction of ANF cannot be responsible for lack of adequate decrease of plasma volume; its reduction can be taken as a marker of improved cardiac pump function and decreased atrial stretch.