JACC Vol. 23, No. 1
January 1994:82–91

Systemic and Cardiac Neuroendocrine Activation and Severity of Myocardial Ischemia in Humans

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Objectives. **The purpose of this study was to assess the effect of different degrees of ischemia on circulating and cardiac neurohormones and vasotone.**

Background. **Neuroendocrine activation and subsequent systemic vasoconstriction may complicate ischemia. Whether this relates to severity of ischemia and subsequent cardiac dysfunction, and whether neurohormonal balance in the ischemic area changes, is unknown.**

Methods. **Fifty-six normotensive patients with coronary artery disease were evaluated during incremental atrial pacing. On the basis of ST segment changes, patients were classified in a nonischemic (n = 11) or ischemic group (n = 45), the latter patients were subsequently classified as lactate (n = 28) or nonlactate (n = 17) producing, to identify neurohormonal changes in the effluent of the ischemic myocardium.**

Results. **Angina occurred in 55%, 82% and 82% of patients in the nonischemic, lactate- and nonlactate-producing groups, respectively. Baseline hemodynamic variables and neurohormones were comparable in all groups, as were heart rate, rate-pressure product and coronary hemodynamic variables during pacing. In lactate producers, contractility did not improve, relaxation deteriorated, left ventricular filling pressure increased and cardiac output decreased during pacing, indicating more severe ischemia compared with that in nonlactate producers. Neurohormones did not change in the nonischemic group. In contrast, arterial and coronary venous catecholamines increased significantly more in lactate producers than in nonlactate producers (arterial norepinephrine by 68% vs. 36%, respectively). Moreover, arterial angiotensin II increased in lactate producers from a baseline mean ± SEM of 6.8 ± 0.9 to 9.7 ± 1.6 pmol/liter ($p < 0.05$), accompanied by a sustained 23% increase in systemic resistance and arterial pressures. In lactate producers, baseline net cardiac norepinephrine release changed to net uptake during pacing (−0.05 ± 0.02 vs. 0.06 ± 0.05 nmol/min, $p < 0.05$). Epinephrine uptake increased in all patients with ischemia, albeit more in lactate producers.**

Conclusions. **Circulating catecholamines and renin-angiotensin levels are activated, and systemic vasotone is increased in relation to the degree of ischemia. Cardiac epinephrine uptake increases, whereas net baseline norepinephrine release from the ischemic myocardium changes to net uptake. Modulation of this neurohormonal activation may provide an alternative mode to limit ischemia.**

(J Am Coll Cardiol 1994;23:82–91)

Systemic neuroendocrine activation is a consistent observation in heart failure (1) and in its preceding stage, asymptomatic left ventricular dysfunction (2). It is also well established in the acute phase of myocardial infarction, where circulating levels of catecholamines, renin-angiotensin and arginine-vasopressin appear to be related to the degree of left ventricular dysfunction (3–5). Recent evidence suggests that circulating catecholamines and renin-angiotensin levels also become activated during short periods of pacing-induced myocardial ischemia (6). Of potential clinical importance, ischemia-induced neurohormonal activation in humans is accompanied by systemic vasoconstriction and an increase in arterial pressures (6). It is unknown, however, whether this neurohormonal stimulation relates to the severity of ischemia and could result from ischemia-induced cardiac dysfunction.

In animal models of ischemia, cardiac norepinephrine efflux increases, indicative of enhanced cardiac sympathetic stimulation (7), but only when ischemia is severe and sustained. Whether, in humans temporary episodes of mild to moderate ischemia, such as occur during exercise- or pacing-induced stress, also result in enhanced cardiac sympathetic stimulation and norepinephrine release, is unclear (8–10). Neither is the effect of myocardial ischemia on cardiac angiotensin fluxes known.

In the present study, sequential changes in systemic and transcardiac catecholamines and angiotensin II levels were investigated during pacing-induced myocardial ischemia in normotensive patients with coronary artery disease without heart failure and were related to the severity of ischemia and subsequent cardiac dysfunction. Second, an attempt was made to study cardiac neurohormonal release in the effluent from ischemic myocardium.

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Manuscript received March 24, 1993; revised manuscript received August 11, 1993, accepted August 13, 1993.

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0735-1097/94/$6.00

Methods

Patients After approval by the institutional Ethical Review Committee and informed consent was obtained, 56 patients (4 women, 52 men; mean age 54 years [range 31 to 73]) were studied. Subjects were referred for coronary angiography for the evaluation of stable angina pectoris with documented exercise-induced ischemia or a previous myocardial infarction, or both, or anginalike chest pain without signs of myocardial ischemia. To be included, patients had to be normotensive without symptoms of heart failure, valvular disease or conduction disturbances. Previous myocardial infarctions had to be at least 1 month old. Patients with unstable angina were not included.

All cardiac therapy was withheld 36 to 72 h before study, depending on plasma half-lives. Only short-acting nitroglycerin was allowed up to 6 h before study. Oral anticoagulant agents and antiplatelet therapy (e.g., aspirin) were withheld 2 to 3 days and 10 days before the investigation, respectively.

Fifty-two patients had significant coronary artery disease, defined as ≥70% diameter narrowing of at least one epicardial coronary artery. Objective signs of ischemia during prestudy exercise testing were present in 32 patients and documented previous myocardial infarctions in 24. In the latter group, nine patients had no demonstrable signs of ischemia during exercise.

Catheterization procedures. All patients were studied in the fasting state at the same time in the morning, without premedication. Before the study, coronary angiography was performed with the Seldinger technique. Thereafter, a 7F Millar or 8F Sentron pigtail microtip manometer catheter was advanced to the left ventricle through an 8F or 9F arterial Desilet introducer system in the right femoral artery to record left ventricular pressures. The side arm of the introducer system was used to monitor arterial pressure. A 7F balloon-tipped triple-lumen thermodilution catheter (Edwards Laboratory) was positioned in a pulmonary artery through an 8F Desilet introducer system in the right femoral vein. A 7F coronary sinus thermodilution and pacing catheter (Wilton Webster Laboratories model CCS-7U-90B) was introduced through a right brachial vein in the coronary sinus. The position of this catheter was such that the proximal thermistor was at least 3 cm beyond the orifice of the coronary sinus, its position stable, enabling a rapid sequence of blood sampling. Absence of atrial reflux into the coronary sinus under the study conditions was confirmed by a bolus injection of 10 ml of saline solution into the right atrium. After instrumentation, the position of the catheters was recorded on videodisc to allow rechecking of their respective positions during the study.

Hemodynamic and electrocardiographic measurements. The hemodynamic variables in this study included left ventricular peak systolic, mean and end-diastolic pressures; left ventricular pressure-derived contractility (first derivative of left ventricular pressure [dP/dt], peak positive dP/dt, at 40 mm Hg and V_{max} [dP/dt/pressure (P) extrapolated to P = 0 mm Hg total pressure]) and relaxation indexes (peak negative dP/dt, isovolumetric relaxation indexes tau_1 and tau_2), mean and phasic arterial pressures, right atrial pressure, cardiac output and coronary sinus blood flow. All fluid-filled catheters were calibrated, using Bentley transducers, with a zero reference level set at midchest. The micromanometer pressure was balanced to zero and superimposed on the conventional pressure tracings. After appropriate calibration, all pressures, coronary flow, cardiac output and dP/dt were recorded on paper, using a CGR 1000 catheterization laboratory system. They were also determined on-line by a Mennen catheterization laboratory computer system. Through this system, pressures and pressure-derived contractility indexes were calculated in 15 to 20 consecutive beats to average out respiratory variations. Contractility indexes and peak negative dP/dt were calculated and displayed on-line. In contrast, tau_1 and tau_2 were measured off-line (11). Coronary sinus blood flow was determined by the thermodilution technique, as previously described (12). The Mennen catheterization laboratory system displays 1-s measurements of mean coronary flow, which allows instantaneous estimation of the stability of coronary flow recordings.

At the end of the study, the pressure curves from the femoral artery were compared with a simultaneous recording from the aortic root by the micromanometer catheter to compensate for any difference between proximal and distal arterial pressures. Heart rate and ST segment changes were determined from 100-mm/s recordings of electrocardiographic (ECG) leads I, II and V_5. The ST segment was measured in 3 to 5 consecutive beats 80 ms after the J point, using a calibrated magnifying glass.

Blood sample collection and assay of metabolites. Simultaneous blood samples from the left ventricle and coronary sinus were used to determine lactate levels and oxygen saturation values on an OSM-80 oximeter (Waters Association). The sampling procedure and analysis of lactate have been published in detail elsewhere (13). The standard deviation in our laboratory is 0.012 mmol/liter.

Neurohumoral measurements. Neurohormones were assessed in a minimum of 6 ml of blood simultaneously collected from the left ventricle and coronary sinus in precooled syringes. For the assay of angiotensin II, 3 ml was immediately transferred to ice-cold tubes containing 4 mg of ethylenediaminetetraacetic acid and 0.06 mg of *o*-phenanthroline. The remaining 3 ml was transferred to ice-cold tubes containing 500 liters/U of heparin and 3 mg/ml of glutathione for determination of norepinephrine, epinephrine and dopamine. Samples were centrifuged at 3,000*g* under cooled conditions and the supernatants frozen at −50°C. Catecholamines were determined by radioenzymatic assay and high pressure liquid chromatography to separate the radioactive products (14), whereas angiotensin II was assessed by radioimmunoassay (15).

Calculations. From the measured hemodynamic variables, the following variables were derived using standard formulas (16): coronary vascular resistance (mm Hg/ml per

min), systemic vascular resistance (dynes·s·cm^{-5}), stroke index (ml/beat per m^2), stroke work index (g·m/m^2). Myocardial oxygen extraction (ml oxygen/ml) was calculated as arterial oxygen content (ml oxygen/ml) minus coronary venous oxygen content (ml oxygen/ml), and myocardial oxygen consumption (ml/min) was calculated as the product of myocardial oxygen extraction (ml oxygen/ml) and coronary blood flow (ml/min). Percent myocardial lactate extraction was calculated as 100 × (arterial lactate content [mmol/liter] − coronary venous lactate content [mmol/liter])/arterial lactate content (mmol/liter). Myocardial lactate, catecholamine and angiotensin II uptake (mmol/min, nmol/min and pmol/min, respectively) were calculated by multiplying the respective differences in arterial and coronary venous levels with the instantaneous coronary sinus blood flow (ml/min).

Study protocol. Patients were studied 45 to 60 min after the last coronary angiogram and 25 to 30 min after instrumentation. Hence, all studies were performed between 10:30 and 11:30 AM, with the patient resting and supine for approximately 2 h. First, multiple control measurements of all hemodynamic variables were performed to ensure stable baseline values. Next, duplicate arterial and coronary venous blood samples for metabolic and neurohumoral variables were collected. Then, an atrial pacing stress test was carried out, during which heart rate was elevated by 10 beats/2 min until anginal pain or atrioventricular block occurred, or a maximal heart rate of 170 beats/min was reached. The patient always determined which level of anginal pain he or she would endure and, hence, the duration of the atrial pacing stress test. However, pacing was typically continued until the level of angina was at least comparable to that at which the patient would normally discontinue exercise or take sublingual nitroglycerin.

During maximal heart rates, just before cessation of pacing, all variables, including neurohormone levels, were reassessed, followed by repeated measurements of all variables at 1, 2 and 30 min after pacing. The sequence of blood sampling after pacing was such that exactly at 15 s after pacing, blood was collected for lactate levels, followed immediately by the collection of blood for neurohormone levels. This sequence was repeated during the 2- and 30-min interval after pacing. Coronary flow and cardiac output were not determined during the first 2 min after pacing. In a limited number of patients, neurohormones and lactate levels and hemodynamic variables were also evaluated at 5 min after pacing.

During the interval between pacing and the 30-min evaluation, patients rested without further instrumentation or investigational procedures. During the study, approximately 60 ml of blood was collected. Before each sample, catheters were emptied to obviate the collection of residual saline solution and blood, which were returned to the patient after sampling.

Statistical analysis. Data are presented as mean value ± SEM. The change in values between measurements during and after pacing and baseline were calculated. Group differences at baseline and for calculated changes were examined using a one-way analysis of variance. Within-group comparisons with baseline values were evaluated using an analysis of variance for repeated measurements, followed by a multiple comparison test according to Dunnett. Comparisons between baseline values and noncontinuous data assessed at 5 and 30 min after pacing were carried out with a two-tailed paired t test. Coefficients of correlation were calculated where appropriate. A p value < 0.05 was regarded as significant.

Results

On the basis of objective signs of myocardial ischemia during pacing (e.g., >0.1-mV ST segment depression), patients were classified in an ischemic (n = 45) or nonischemic group (n = 11). Patients with ischemia were subsequently classified into those with (n = 28) and without (n = 17) lactate production. The reason for this subgrouping of patients with ischemia was twofold. First, it was hypothesized that patients who produce lactate have more severe ischemia than those who do not produce lactate. Hence, a correlation between neurohormonal stimulation and degree of ischemia might be made. Second, lactate production at least ensured that (part of) the coronary venous effluent, collected during the study, represented the ischemic area and might thus allow identification of changes in neurohormonal release from that area. Angina during pacing-induced stress was not considered a true representation of myocardial ischemia (17). Anginalike symptoms occurred in 6 (55%), 14 (82%) and 23 (82%) patients in the nonischemic and lactate- and nonlactate-producing groups, respectively.

Clinical and angiographic characteristics of the three groups (Table 1). Groups were comparable with regard to gender, age, number of old infarcts, left ventricular volume

Table 1. Baseline Clinical and Angiographic Data

	Group		
	NI (n = 11)	LP (n = 28)	NLP (n = 17)
M/F	9/2	28/0	15/2
Age (yr)	49 ± 3.1	55 ± 1.7	54 ± 2.4
Range	31–66	37–73	36–71
Previous MI (ant/inf)	4/1	8/4	2/5
Positive exercise test result		20	10
Coronary angiography (≥70% diameter stenosis)			
0 vessel	4	0	0
1 vessel (L/R)	3/1	7/1	4/1
2 vessels	1	12	6
3 vessels	2	9	5
LVEF (%)	54 ± 3.5	52 ± 2.6	53 ± 3.1
LVEDV (ml/m^2)	74 ± 6.7	69 ± 4.8	67 ± 3.7

Unless otherwise indicated, values presented are mean value ± SEM or number of patients. ant = anterior; F = female; Inf = inferior, LP = lactate producing; L/R = left/right; LVEDV = left ventricular end-diastolic volume; LVEF = left ventricular ejection fraction; M = male; MI = myocardial infarction; NI = nonischemic; NLP = nonlactate producing.

Table 2. Systemic Hemodynamic Variables During and After Pacing

		After Pacing			
	Baseline	Maximal Pacing Rate	1 min	5 min	30 min
LVSP (mm Hg)					
LP	153 ± 5.3	149 ± 6.0	162 ± 6.6*†	153 ± 5.3	144 ± 7.1
NLP	143 ± 6.5	139 ± 7.1	143 ± 6.9	141 ± 7.3	137 ± 5.5
NI	138 ± 6.6	130 ± 4.0†	136 ± 6.5	134 ± 5.2	135 ± 5.5
DP (HR × LVSP × 10^{-3})					
LP	12 ± 0.5	22 ± 0.9*	13 ± 0.8	12 ± 0.5	9 ± 0.6
NLP	11 ± 0.7	20 ± 1.2*	11 ± 1.0	11 ± 0.9	12 ± 0.6
NI	10 ± 0.7	17 ± 1.2*†	10 ± 0.7	10 ± 0.7	10 ± 0.5
dP/dt pos (mm Hg·s^{-1})					
LP	1,779 ± 80	2,241 ± 126*	1,840 ± 117	1,796 ± 65	1,686 ± 108
NLP	1,715 ± 102	2,363 ± 125*	1,740 ± 130	1,753 ± 103	1,720 ± 106
NI	1,511 ± 134	1,897 ± 138*	1,567 ± 124	1,547 ± 92	1,326 ± 69
Vmax (s^{-1})					
LP	50 ± 2.6	56 ± 2.7*‡	51 ± 2.9	49 ± 2.5	49 ± 3.3
NLP	50 ± 2.4	68 ± 3.5*	52 ± 2.7	50 ± 2.4	51 ± 2.5
NI	45 ± 3.5	59 ± 3.8*	44 ± 3.8	45 ± 3.6	41 ± 2.7
dP/dt neg (mm Hg·s^{-1})					
LP	1,914 ± 86	1,971 ± 118	1,960 ± 103	1,980 ± 100	1,875 ± 136
NLP	1,883 ± 89	2,111 ± 115*	1,963 ± 125	1,858 ± 104	1,788 ± 74
NI	1,769 ± 127	1,999 ± 121*	1,776 ± 137	1,958 ± 147	1,852 ± 150
Tau_1 (ms)					
LP	50 ± 45	47 ± 4.7‡	51 ± 4.9	51 ± 4.8	50 ± 7.0
NLP	46 ± 1.7	39 ± 2.2*	46 ± 2.3	48 ± 2.1	50 ± 1.4
NI	53 ± 4.7	43 ± 3.9*	52 ± 4.9	56 ± 7.2	56 ± 5.9
Tau_2 (ms)					
LP	44 ± 4.7	55 ± 5.5*‡	48 ± 5.1	44 ± 5.2	38 ± 4.0
NLP	37 ± 1.2	39 ± 3.1	38 ± 2.5	38 ± 1.7	38 ± 1.5
NI	42 ± 3.9	42 ± 4.7	43 ± 4.1	41 ± 4.2	43 ± 4.2
CO (liter/m)					
LP	5.9 ± 0.3	—	—	5.3 ± 0.3*	6.0 ± 0.4
NLP	5.4 ± 0.3	—	—	4.9 ± 0.2	5.7 ± 0.5
NI	5.0 ± 0.4	—	—	4.8 ± 0.4	4.7 ± 0.5

*$p < 0.05$ versus baseline. †$p < 0.05$ versus other groups. ‡$p < 0.05$ ΔAPST (change from baseline during maximal pacing) versus other ΔAPST. Values presented are mean value ± SEM. CO = cardiac output; DP = double (rate-pressure) product; dP/dt = first derivative of left ventricular pressure; dP/dt neg (pos) = peak negative (positive) dP/dt; HR = heart rate; LVSP = left ventricular systolic pressure; Tau_1, Tau_2 = isovolumetric relaxation indexes; Vmax = dP/dt/pressure (P) extrapolated to P = 0 mm Hg total pressure. Other abbreviations as in Table 1.

and ejection fraction. In contrast, objective signs of ischemia during exercise occurred more often in both groups with ischemia than in the nonischemic group. Similarly, coronary artery disease was less severe in the nonischemic group. The two ischemic groups were similar with respect to the number of vessels diseased. Although a >70% stenosis in a dominant right coronary artery was the only significant lesion in one patient in each group, each of these two patients had additional 50% lesions in the left coronary artery.

Hemodynamic, metabolic and neurohumoral values at baseline. At baseline, no patient had clinical signs or symptoms of myocardial ischemia. Although baseline values of hemodynamic variables were comparable in the three groups (Tables 2 and 3), coronary resistance was significantly higher in lactate producers. In this group, baseline coronary venous norepinephrine and epinephrine levels were also significantly higher. Neurohormone levels otherwise did not differ in the three groups, except for arterial and coronary venous dopamine values, which were lower in nonlactate producers. Moreover, median values and range of individual arterial epinephrine and norepinephrine levels were similar in the three groups, as were individual arterial angiotensin II values, although the latter varied widely, from <0.2 to 24.2 pmol/liter.

Metabolic and ECG changes during and after pacing. By design, only the lactate-producing group had abnormal myocardial lactate metabolism (lactate extraction −76 ± 15%, 15 s after pacing, [mean ± SEM]). ST segment changes, however, were similar in both ischemic groups (−0.21 ± 0.03 and −0.17 ± 0.02 mV in lactate and nonlactate producers, respectively) but were absent in the nonischemic group.

Coronary and systemic hemodynamic variables during and after pacing. Changes in heart rate and the rate-pressure product between baseline and maximal pacing were compa-

Table 3. Coronary Hemodynamic Variables During and After Pacing

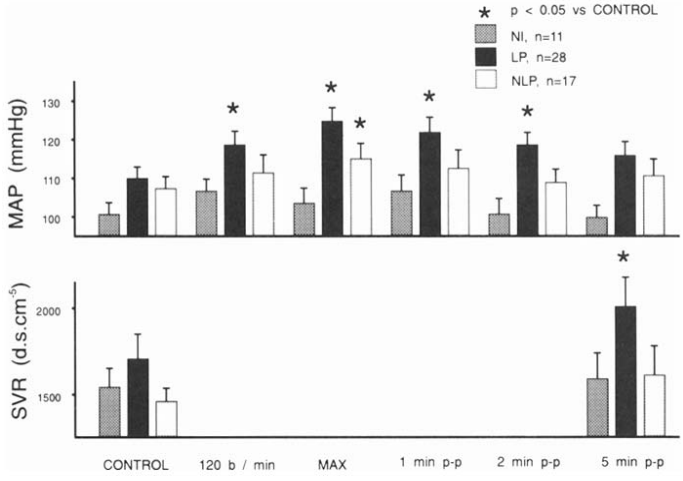
	Baseline	Maximal Pacing Rate	5 min After Pacing	30 min After Pacing
CSBF (ml/min)				
LP	117 ± 12	205 ± 19*	126 ± 14	112 ± 19
NLP	122 ± 6	175 ± 24*	116 ± 9	110 ± 12
NI	137 ± 15	202 ± 32	149 ± 15	144 ± 25
CVR (mm Hg/ml per m)				
LP	1.19 ± 0.12†	0.73 ± 0.06*	1.19 ± 0.13	1.32 ± 0.14
NLP	0.90 ± 0.10	0.75 ± 0.09	0.95 ± 0.10	1.01 ± 0.11
NI	0.71 ± 0.08	0.58 ± 0.13	0.74 ± 0.13	0.87 ± 0.18
$\Delta A\text{-}CSO_2$ (ml/min)				
LP	12 ± 0.5	12 ± 0.6	11 ± 0.6	11 ± 0.6
NLP	12 ± 0.7	12 ± 0.7	12 ± 0.7	13 ± 0.8
NI	14 ± 0.6	12 ± 0.9	13 ± 0.8	13 ± 0.9
MVO_2 (ml/min)				
LP	12 ± 1.1	22 ± 2.0*	12 ± 1.0	11 ± 1.5
NLP	14 ± 1.6	21 ± 2.5*	13 ± 1.3	15 ± 2.0
NI	19 ± 2.8†	23 ± 4.9	20 ± 2.7†	20 ± 5.5

*$p < 0.05$ vs. baseline. †$p < 0.05$ vs. other groups. Values presented are mean value ± SEM. CSBF = coronary sinus blood flow; CVR = coronary vascular resistance; $\Delta A\text{-}CSO_2$ = myocardial oxygen extraction; MVO_2 = myocardial oxygen consumption; other abbreviations as in Table 1.

rable in the three groups, although as a result of a more profound reduction in left ventricular systolic pressure during pacing in the nonischemic group, the absolute value for rate-pressure product was significantly lower than in the other groups (Table 2). Also, changes in coronary hemodynamic variables during pacing were similar in the three patient groups (Table 3). In contrast, isovolumetric contractility and relaxation variables improved markedly in the nonischemic and non–lactate-producing groups but did not improve, or were significantly lower, in the lactate-producing group (Table 2). Moreover, tau_2 lengthened in lactate producers but not in the other groups. Also, left ventricular end-diastolic pressure increased significantly from 13 ± 1.5 (baseline) to 24 ± 2.5 mm Hg at 10 s after pacing. In contrast, in the other groups left ventricular end-diastolic pressure decreased significantly during pacing and returned to baseline levels immediately after pacing. Furthermore, cardiac output decreased from 5.9 ± 0.35 (baseline) to 5.3 ± 0.37 liters/min (5 min after pacing, $p < 0.05$) in the lactate-producing group, accompanied by a significant 23% increase in systemic vascular resistance (Fig. 1). No such changes were observed in the other groups. Moreover, whereas arterial pressures increased in both ischemic groups, these changes were more extensive and longer lasting in lactate producers than in nonlactate producers.

Neurohumoral changes during and after pacing. Neither arterial nor coronary venous neurohumoral levels changed during or after pacing in the nonischemic group (Fig. 2), whereas in the ischemic group dopamine levels remained unaltered. In contrast, arterial norepinephrine increased significantly from 1.91 ± 0.18 (baseline) to 2.8 ± 0.26 nmol/liter (after pacing), with a further increase to 3.21 ± 0.34 nmol/liter (1 min after pacing) in lactate producers, accompanied by significant, albeit less pronounced, increases in coronary venous norepinephrine levels. In nonlactate producers, these changes were not as profound and consistent. However, arterial norepinephrine increased by 36% in nonlactate versus 68% in lactate producers ($p < 0.05$). Elevated arterial and coronary venous epinephrine levels were observed in

Figure 1. Effect of pacing-induced ischemia on arterial pressures and systemic vascular resistance (SVR). In patients producing lactate (LP) a marked increase is observed in mean arterial pressure (MAP), starting at 120 beats/min (b/min) and persisting until 2 min after pacing (p-p). In contrast, in nonlactate producers (NLP) a moderate increase occurs at maximal pacing (MAX) only, whereas arterial pressures do not change in patients in the nonischemic (NI) group. Also, systemic resistance is still elevated at 5 min after pacing in lactate producers but not in the other groups. Values are mean value ± SEM. d = dynes.



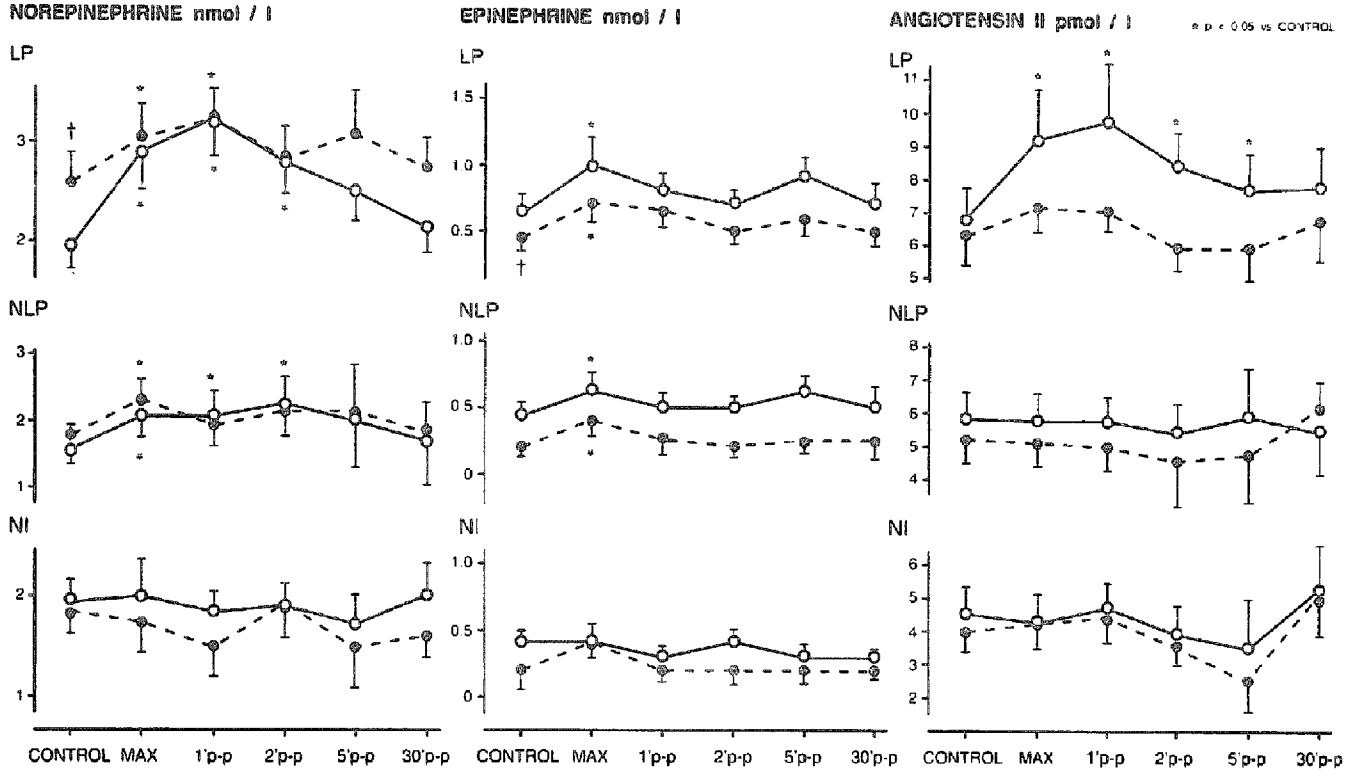


Figure 2. Sequential changes in neurohormone levels during maximal pacing and for 30 min (') after pacing. At baseline, neurohormone values are comparable, although coronary venous (open circles) norepinephrine and epinephrine values in patients producing lactate (LP) are significantly higher than in the other groups. During pacing, no changes occur in patients in the nonischemic group (NI). In contrast, a significant increase in arterial (closed circles) and coronary venous norepinephrine and epinephrine occurs in lactate and nonlactate producers. Changes are more pronounced and sustained in lactate producers. Also, arterial angiotensin II increases significantly in lactate producers but not in nonlactate producers. *$p < 0.05$ vs. baseline; †$p < 0.05$ vs. other groups. Abbreviations as in Figure 1.

both ischemic groups during maximal pacing. Again, changes were more profound in lactate producers. Arterial angiotensin II levels also increased but only in lactate producers, from 6.8 ± 0.9 (baseline) to 9.7 ± 1.6 pmol/liter (1 min after pacing, $p < 0.05$). Values were still significantly elevated at 5 min after pacing. Coronary venous levels did not change in either group.

Neurohumoral changes did not correlate with lactate production values. However, a significant correlation was observed between changes in arterial norepinephrine and left ventricular end-diastolic pressure in the lactate-producing group ($p < 0.05$).

Cardiac neurohormonal balance during pacing and ischemia. Data for transcardiac neurohormone levels and coronary flow were available in 19 lactate and 15 nonlactate producers and 9 patients in the nonischemic group. Baseline values for the transcardiac balance of catecholamines and angiotensin II did not differ in the three groups (Fig. 3). During and after pacing, no changes were found in cardiac dopamine balance. Similarly, myocardial fluxes of the other catecholamines remained unaltered in the nonischemic group. In contrast, in lactate producers, net myocardial norepinephrine release, present at baseline, changed significantly to net uptake during maximal pacing rates (−0.05 ± 0.02 vs. 0.06 ± 0.05 nmol/min, respectively, (Fig. 3). No such changes occurred in nonlactate producers. Cardiac epinephrine uptake increased significantly in both ischemic groups, although it was more pronounced in lactate producers. Ischemia-induced changes in catecholamine fluxes were back to baseline values at 5 min after pacing (Fig. 3). In the lactate-producing group, individual values for angiotensin II were variable at baseline, with uptake in the majority of patients and release in seven (Fig. 4). Pacing did not alter this pattern significantly, although lactate producers tended to have greater cardiac angiotensin II uptake during pacing-induced ischemia. Changes in arterial angiotensin II levels during pacing correlated significantly with simultaneous cardiac uptake values ($p < 0.01$).

Hemodynamic, metabolic and neurohumoral variables after a 30-min rest period. After 30 min of rest, all variables had returned to baseline values in the three patient groups.

Discussion

The present study provides evidence that systemic catecholamines are stimulated during short periods of stress-induced myocardial ischemia and that the level of activation

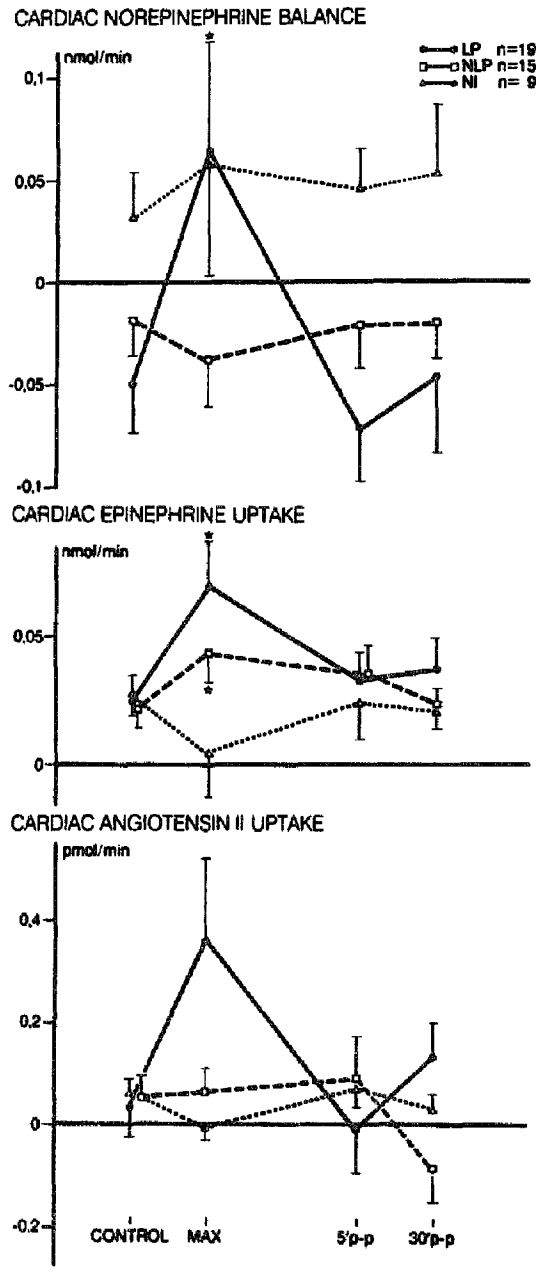


Figure 3. Sequential alterations in cardiac neurohumoral balance. Changes in catecholamine balance in ischemic patients with and without lactate production are present during maximal pacing rates and return to baseline values at 5 min after pacing. Values are mean value ± SEM. *$p < 0.05$ vs. baseline. Abbreviations as in Figure 1.

depends on the degree of ischemia. Second, it shows that, in humans, ischemia activates the circulating renin-angiotensin system, again as a function of the severity of ischemia, and that more severe ischemia and, hence, greater neurohormonal stimulation lead to a more profound increase in systemic resistance and arterial pressures. Finally, it indicates that acute, short-term ischemia results in a change from net cardiac norepinephrine release to net uptake, but only in patients who produce lactate, most likely reflecting an alteration in the ischemic area. In contrast, epinephrine uptake is enhanced in all patients with ischemia.

Ischemia-induced changes in systemic catecholamine levels: secondary to angina? In this study, arterial norepinephrine and epinephrine levels increased significantly in both ischemic groups during pacing-induced ischemia. By contrast, this did not occur in the nonischemic group despite equivalent increments in heart rate and the presence of anginal symptoms in the majority of these patients. The latter finding does not support a role for angina in sympathetic activation as a result of ischemia.

Previous investigations have focused on neurohormonal changes during stress-induced angina. Because these studies have not always stipulated the presence of objective signs of ischemia (9,18,19) or included patients without myocardial ischemia (20), it is difficult to dissociate the effect of stress caused by angina from that of ischemia as such. Taken together, in most studies in which pacing-induced stress was applied, no significant changes in circulating norepinephrine or epinephrine levels have been observed during angina (8–10,21). Only Emmanuelson et al. (18), in a study design comparable to that of ours, reported a rise in circulating catecholamine levels. Thus, other mechanisms than angina may be responsible for the effect of ischemia on neurohormone levels observed in our study.

Baroreceptor-induced sympathetic stimulation and severity of ischemia. As hypothesized at the outset of the study, patients who produced lactate had clearly more severe ischemia, as illustrated by the abnormalities in contractility, relaxation and left ventricular filling pressure in these patients compared with the nonlactate producers, despite similar baseline characteristics and pacing conditions. Moreover, cardiac output was significantly reduced at 5 min after pacing and probably more so during and immediately after pacing. A reduction in cardiac output during the development of ischemia may reset arterial baroreceptors, stimulating the vasomotor center in the medulla oblongata. Whether this results in a secondary overshoot or mainly helps to amplify the stimulus for systemic vasoconstriction derived from sympathetic afferent nerves is unknown. Also, it does not explain why moderate, short-term neuroendocrine activation was observed in nonlactate producers who were in the ischemic group but had myocardial muscle and cardiac pump function that remained unimpaired during the test.

Is the cardiac sympathosympathetic reflex involved in catecholamine activation during ischemia? An intriguing hypothesis is that a cardiac sympathosympathetic reflex may be involved in sympathetic activation during ischemia in the absence of cardiac dysfunction (22,23). Very little is known of this reflex in humans. Although animal studies suggest that ischemia needs to be transmural (i.e., relatively severe) to induce a sympathetic reflex response (24), our observations in nonlactate producers would support a reflex that initiates in the heart, secondary to local ischemic changes, and leads to increased circulating catecholamines and a subsequent increase in arterial tone, even in conditions of

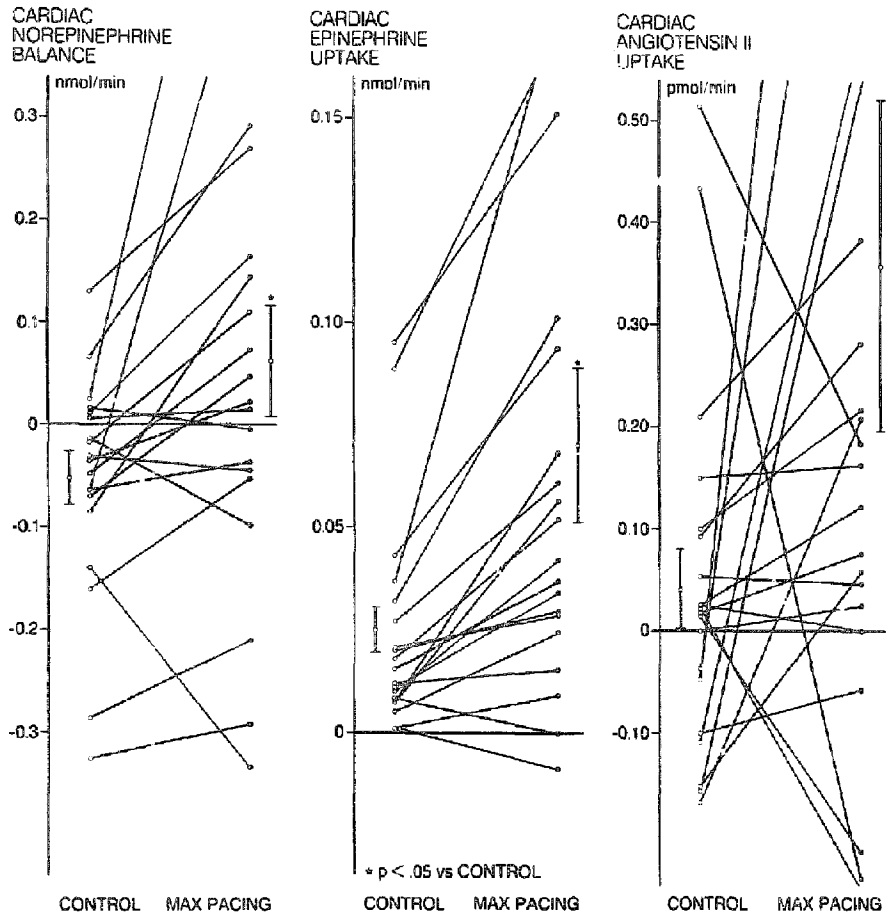


Figure 4. Individual changes in cardiac norepinephrine balance and in epinephrine and angiotensin II uptake by the heart in ischemic patients with myocardial lactate production. During maximal pacing (MAX) rates, norepinephrine release present at baseline reverses to uptake. Also, epinephrine uptake increases significantly. In contrast, angiotensin II fluxes are too variable, although there is a tendency toward enhanced uptake. *$p < 0.05$ vs. baseline.

mild ischemia. The mechanisms that initiate this reflex are not clear from our data.

Mechanical stimuli or lactate may activate mechanoreceptors with afferent nerves to the tractus solitarius in the medulla oblongata during anterior wall ischemia. Alternatively, receptors with vagal C fiber afferents located in the inferoposterior part of the heart depress the vasomotor center after increments in preload. The (overall) hemodynamic and metabolic profile during mild pacing-induced ischemia in nonlactate producers does not favor a central modulation of sympathetic tone through these mechanisms in these patients. In contrast, in patients who produce lactate and have more pronounced ischemia, these factors, together with baroreceptor-dependent effects, may have contributed to the profound neurohormonal stimulation and subsequent systemic vasoconstriction observed in this group. Do they also explain the increase in circulating angiotensin II levels?

Circulating angiotensin II levels and degree of myocardial ischemia. Our data indicate that stimulation of the circulating renin-angiotensin system only occurs when ischemia is severe enough to reduce cardiac and stroke output. Even slight decreases in stroke volume are sensed by arterial baroreceptors and may activate renal sympathetic nerves (25). Baroreflex activation of renal nerves stimulates renin release, possibly through a direct beta-adrenergic influence on juxtaglomerular cells (26). Alternatively, the significant increase in circulating catecholamines may increase the threshold pressure for renin release (27). Several animal studies have indicated that components of the circulating renin-angiotensin system change during ischemia (28,29). A significant elevation of arterial renin and angiotensin II levels has been observed 30 s after coronary occlusion in the dog (30). Of importance, these changes do not occur in nephrectomized animals. In contrast, pretreatment with propranolol has no effect on ischemia-induced renin activation (30). Although beta-adrenergic receptor blockade does not effect the change in threshold pressure for renin release induced by reflex activation of renal sympathetic nerves (31), alpha-adrenergic receptor blockade does (32). In view of the significant increase in circulating catecholamines in our study, such a resetting may have influenced angiotensin II levels during ischemia despite the concomitant increase in arterial pressure.

Cardiac neurohumoral balance at rest and during myocardial ischemia. Although, during ischemia, a positive correlation was present between changes in arterial levels of angiotensin II and cardiac uptake, cardiac angiotensin II balance was not altered by ischemia. By contrast, ischemia significantly changed net cardiac norepinephrine release, present at baseline, to net uptake in patients with lactate production. Because no such changes were found in patients in the ischemic group without lactate production or in the

nonischemic group, this suggests an alteration in norepinephrine kinetics in the ischemic area.

Whether myocardial ischemia resulted in diminished regional norepinephrine spillover or enhanced clearance cannot be deduced from the present study. This determination would necessitate tracer techniques with long equilibration times under control conditions not well suited for invasive investigations, in which the overall study duration is an important restrictive factor (10,33,34). In animal experiments, relatively long periods of severe myocardial ischemia result in norepinephrine overflow (7), which is generally believed to reflect enhanced sympathetic activity secondary to stress or enhanced autonomic reflexes (35). However, during short periods of ischemia, comparable to the model of pacing-induced stress in humans, norepinephrine overflow is either minor (7) or absent (36,37). Thus far, patient studies in which mild to moderate ischemia was induced over short intervals were unable to show enhanced cardiac norepinephrine release (8–10). The present study is the first to indicate that during myocardial ischemia, not only is enhanced net norepinephrine release absent, but also, in contrast, net release observed at baseline may be reversed to net uptake in the ischemic area. The underlying mechanisms for this change in norepinephrine kinetics can only be hypothesized in the absence of clearance measurements. A reduction in efflux from the ischemic area as a result of diminished venous outflow is unlikely in view of the increase in coronary venous lactate levels. Enhanced neuronal reuptake of norepinephrine during ischemia has been described, but only in experiments that are unlikely to reflect our study conditions (38). Local norepinephrine release from stimulated cardiac sympathetic nerves (exocytotic release) depends on neuronal high energy phosphate levels (39) and may decrease as a result of reduced availability of the latter during ischemia. However, this process is time dependent and probably cannot account for the early changes observed in our studies. An alternative, attractive hypothesis is presynaptic inhibition of exocytotic norepinephrine release by adenosine (40), which accumulates in the ischemic area (41) and is produced during pacing-induced ischemia in humans (42,43).

In contrast to norepinephrine, epinephrine uptake was enhanced in all patients with ischemia irrespective of the degree of ischemia. This finding suggests that epinephrine uptake by the heart is not specific. Enhanced cardiac extraction of epinephrine has been described in association with increased arterial epinephrine levels during relatively short periods of ischemia in a canine model (37).

Neurohumoral stimulation and vasoconstriction. Our study indicates that magnitude and duration of systemic vasoconstriction depend on the degree of ischemia and subsequent catecholamine activation, and, in severe ischemia, on stimulation of the circulating renin-angiotensin system. Consequently, arterial pressures and systolic wall stress are elevated, particularly in severe ischemia, and result in an increase in myocardial oxygen demand. A vicious circle may then ensue, whereby neurohormonal activation and systemic vasoconstriction lead to further aggravation of the initial ischemic event.

Our study does not provide evidence that ischemia-induced neuroendocrine stimulation affects coronary vasotone. If anything, coronary vascular resistance decreased more in lactate producers than in other groups. This observation may reflect metabolic vasodilation in ischemic areas, or, alternatively, because the thermodilution method measures overall rather than regional flow, pronounced coronary vasodilation in nonischemic areas. During progressive reductions in coronary flow, cardiac sympathetic stimulation may convert metabolic coronary vasodilation to alpha-adrenergic vasoconstriction (44–46). Also, sympathetic stimulation may lead to constriction of stenotic coronary segments without an apparent effect on overall coronary flow (47). Therefore, the possibility that a local decrease in coronary flow did indeed occur during ischemia cannot be excluded in the present study.

Clinical implications. In this study, systemic neuroendocrine activation was associated with a sustained increase in systemic vascular resistance, at least in more severe ischemia. Because this increase may significantly influence the severity of the ischemic attack that initially provoked this neurohumoral response, the clinical usefulness of therapy that may reduce myocardial ischemia through neurohumoral modulation should be further evaluated. In accordance with our model and the results of the present study, anti-ischemic efficacy may be anticipated during periods of relatively severe ischemia in the resting, supine patient (e.g., during unstable angina). Preliminary observations that enalaprilat and perindoprilat reduce stress-induced ischemia at rest by modulating neurohormonal activation (48,49) emphasize the potential importance of converting enzyme inhibitors as (additional) anti-ischemic therapy.

We gratefully acknowledge the technical assistance of the nurses and technicians of the Catheterization Laboratory and the Laboratory for Circulation Research.

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