Effects of endothelial and inducible nitric oxide synthases inhibition on circulatory function in rats after myocardial infarction

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Abstract

Objectives: To examine the relative roles of eNOS and iNOS (endothelial and inducible nitric oxide synthases) on basal and β-adrenergic receptor (β-AR)-stimulated arterial hemodynamic responses after myocardial infarction (MI). Methods: Left ventricular (LV) pressures and steady-state and pulsatile arterial hemodynamics were measured at baseline, and after acute NOS inhibition with either Nω-nitro-L-arginine methyl ester (L-NAME, 100 mg/kg) or iNOS inhibition with aminoguanidine (AG, 75 mg/kg) in sham-operated and MI Sprague-Dawley rats. Results: In sham rats, L-NAME decreased (P<0.05) peak positive LV dp/dt and aortic blood velocity by 19% and 53%, respectively, and increased (P<0.05) mean arterial pressure (MAP), systemic vascular resistance, and LV end-diastolic pressure (EDP) by 20, 189 and 89%, respectively. The frequency-dependent components of hemodynamics including aortic input impedance modulus, characteristic impedance, and phase shift were increased (P<0.05) with L-NAME, while pulsatile power was decreased (P<0.05). AG increased (P<0.05) aortic input impedance modulus and characteristic impedance but had no effect on any other hemodynamic variable. In MI rats, L-NAME decreased (P<0.05) LV dp/dt and aortic blood velocity by 22 and 55%, respectively, and increased (P<0.05) SVR by 108%. There was no effect of L-NAME on MAP or LV EDP in MI rats. After MI, AG increased (P<0.05) heart rate and LV dp/dt but had no effect on other LV or pulsatile hemodynamic variables. Compared to sham rats, heart rate, LV dp/dt, and blood velocity–isoproterenol dose responses were shifted downward (P<0.05), while SVR–isoproterenol dose response was shifted upward (P<0.05) in MI rats. In sham rats, L-NAME potentiated (P<0.05, at >10^{-2} µg/kg) the isoproterenol-induced increase in LV dp/dt and aortic blood velocity, and potentiated (P<0.05) the isoproterenol-induced decline in SVR. As expected, AG had no effects on isoproterenol-stimulated hemodynamics in sham rats. After MI, there was no effect of L-NAME or AG on isoproterenol-stimulated hemodynamics. Conclusions: (1) Circulatory and cardiac responses to inhibition of NO by L-NAME suggest that eNOS, but not iNOS, is the principal regulator of integrated arterial hemodynamic function in rats. (2) Both basal and β-AR-stimulated NO regulation of hemodynamic are attenuated after MI. (3) The attenuation of arterial hemodynamic effects after isoproterenol is mediated, in part, by alterations in the β-AR-activation of eNOS system after MI. © 1999 Elsevier Science B.V. All rights reserved.

Keywords: Blood flow; Heart failure; β-Adrenergic receptors; Nitric oxide

1. Introduction

In heart failure, inappropriate vasoconstriction results from impaired endothelial-dependent vasodilatation. Although activation of a number of neurohumoral systems play a role in this inappropriate vasoconstriction, alterations in endothelial nitric oxide (NO) release and diminished vascular β-AR function [1] are important. Vascular endothelial release of NO, a potent vasodilator, and the arterial vasodilatory response to β-adrenergic receptor (β-AR) stimulation are attenuated in heart failure [2–4]. Since there is a cross-talk between the β-AR and the NO pathways, one potential mechanism that may contribute to β-AR hypo-responsiveness is altered vascular endothelial function.

In the cardiovascular system, NO is produced by two isoforms of nitric oxide synthase (NOS), eNOS and iNOS [5,6]. The eNOS species is constitutively expressed and can be stimulated with interventions that increase cytosolic Ca^{2+}. In contrast, iNOS is a cytokine-inducible isoenzyme
Heart failure is characterized by elevated eNOS and iNOS activity in cardiac muscle, while NO basal release in large arteries is diminished [2–4]. The eNOS isoform has been shown to play a major role in circulatory function in hypertension, but there are limited data in heart failure [7,8]. The iNOS isoform may have an important effect on hemodynamics early after myocardial infarction (MI) [9], when inflammatory cytokine levels are elevated, but the effects of iNOS in compensated ischemic heart failure are unknown. Although ischemic heart failure is characterized by increased vascular resistance, alterations in pulsatile hemodynamics [10], and impairment of endothelial function, the role of NO on in vivo circulatory function that includes steady-state and pulsatile hemodynamics has not been examined.

The objectives of the current study, therefore, are to (1) define the effects of NOS inhibition on complete arterial hemodynamics in ischemic heart failure, and to (2) define the effects of NOS inhibition on β-AR responsiveness. The underlying hypothesis is that NOSs play a major role in controlling circulatory function in heart failure. We also postulate that the effects of basal and β-AR stimulation on NO release are diminished in heart failure. This study is performed in rats with compensated heart failure, 6 weeks after coronary artery ligation [11,12].

2. Methods

The investigation conforms with the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health NIH Publication No. 85-23, revised 1996.

2.1. Myocardial infarction model

MI was created using techniques standard in our laboratory [10,11]. In brief, 3-month old Sprague–Dawley rats weighing 250–300 g were anesthetized with ketamine and acepromazine and a left thoracotomy was performed. The heart was expressed from the thorax and a ligature placed around the proximal left coronary artery. The heart was returned to the chest and the thorax closed. The rats were maintained on standard rat chow and water ad libitum. After 3 weeks, rats were anesthetized with methoxylflurane and a nine lead ECG performed. With criteria described previously [13], rats with evidence of large MI were selected for study. Briefly, the presence of Q waves (>1 mV) in the limb leads (I or aVL) and the sum of the R waves in the precordial leads (<10 mV) were used as criteria for large MI. Our laboratory has shown that rats selected in this fashion have large MI averaging 40% of the left ventricle [13]. MI was confirmed 3 weeks later by hemodynamics (LV end-diastolic pressure (EDP) >16 mmHg) and presence of large scar. Only rats with evidence of a large MI were used. Animals that underwent thoracotomy but did not have the coronary artery ligated were designated as sham-operated controls. Studies were performed 6 weeks post-infarction in sham and MI rats.

2.2. Hemodynamic measurements

In vivo hemodynamics were measured using methods previously reported by our laboratory [10]. In brief, rats were anesthetized with Inactin (100 mg/kg) and the trachea ventilated. A 2 F micromanometer catheter with two pressure sensors (Millar) was inserted via the right femoral artery such that one sensor was located in the left ventricle and the other in the ascending aorta. The zero pressure baseline was obtained by placing the pressure sensor in 37°C saline prior to measurements. A 3 F echo-Doppler catheter (Millar Instruments) introduced via the right carotid artery was used to measure the aortic blood velocity. After a period of stabilization, LV and aortic pressures and aortic blood velocity were recorded and digitized at a rate of 1000 Hz using a PC equipped with an analog–digital converter and customized software. From these data, LV dP/dt and the time constant of LV relaxation (τ) were calculated. A systemic vascular resistance (SVR) index was calculated as the quotient between mean arterial pressure (MAP) and blood velocity. The aortic pressures and flow waves were decomposed into harmonics using Fourier analysis. The impedance, as a function of frequency, was determined as the quotient of pressure and flow harmonics at that frequency. The characteristic impedance was computed as the averaged value of the moduli of impedance for high frequencies. Total peripheral resistance was taken as the impedance modules at zero frequency. The mean power was calculated by multiplying the mean pressure by the mean flow. Pulsatile power was calculated using the following equation:

\[ P_w = 0.5 \sum Q_n Z_n \cos \varphi_n \]

where \( n \) is the harmonic number, \( Q_n \) is the amplitude of the nth flow harmonic, \( Z_n \) is the amplitude of the nth impedance, and \( \varphi_n \) is the phase angle of the nth harmonics.

After obtaining the hemodynamic measurements, a midsternal thoracotomy was performed and the ascending aorta was isolated to measure the aortic diameter. By hemorrhage or intravenous fluid infusion, the MAP was adjusted to the same pressure as that during acquisition of the impedance data. Then the aortic diameter was measured optically using a telemicroscope (M101, Gaenther Scientific, IL, USA). At the end of the experiment, the heart was arrested by KCl injection (2 mEq/v./ml) into the left ventricle, excised, rinsed, visually examined for infarct size, and the left and right ventricle separated and weighed.

2.3. Effects of NOS inhibition on hemodynamics

To determine the appropriate NOS inhibiting dose of L-NAME and AG, pilot studies were performed in four
separate groups of rats (20 rats) to measure the hemodynamic dose response for both L-NAME and AG. Two groups (sham and MI) received acute treatment with L-NAME (1–1000 mg/kg, i.v.). Two more groups (sham and MI) received acute treatment with (AG, 10–80 mg/kg, i.v.). The following variables were measured: MAP, left ventricular dP/dt (LV dP/dt), aortic blood velocity, and SVR were measured after the intravenous administration of different doses of L-NAME or AG.

For the remainder of the study, a total of 68 rats were divided into two groups: (1) sham, (2) MI. In order to determine the role of the basal release of NO on in vivo hemodynamics, measurements were made before and 20 min after the i.v. administration of either L-NAME (100 mg/kg) or AG (75 mg/kg) in sham and MI rats. The role of NO inhibition on β-AR hemodynamic response was determined from the dose–response data to isoproterenol (10⁻³–2 μg/kg) before and 20 min after the intravenous administration of either L-NAME (100 mg/kg) or AG (75 mg/kg). Measurements after L-NAME and AG were performed in separate groups of rats.

2.4. Statistical analysis

All results are expressed as mean±S.D. Statistical comparisons after the addition of L-NAME and AG were carried out using two-way analysis of variance (ANOVA). To examine the interaction between MI and NOS inhibition, a 2×2 matrix was employed in which sham/MI (yes/no) represents one dimension and L-NAME or AG administration (yes/no) the other. For both sham and MI rats, additional two 2×2 matrices were employed to examine the interaction between NOS inhibition and β-AR-mediated responses in which isoproterenol treatment (yes/no) represents one dimension and L-NAME or AG administration (yes/no) the other. Following the two-way ANOVA, intergroup comparisons using the Newman–Keuls procedure were performed. Analysis was performed using BMDP statistical software. Level of significance was taken at P<0.05.

3. Results

3.1. Animal characteristics

In rats after MI, body weight and LV weight decreased (P=0.005, 0.001, respectively) while right ventricular weight and LV EDP increased (P=0.001, Table 1). The elevation in both right ventricular weight and LVEDP confirm the presence of large MI.

3.2. Steady state and pulsatile hemodynamics (MI vs. sham)

MI resulted in a 34% decrease (P<0.01) in LV dP/dt, no change in MAP, a 40% decrease (P<0.01) in aortic blood velocity, with no change in heart rate (HR). The other hemodynamic changes after infarction were a four-fold increase (P<0.01) in LV end diastolic pressure, a doubling (P<0.01) of SVR, and a 48% increase (P<0.01) in the time constant of LV relaxation constant or τ (Fig. 1). MI was associated with an increase (P<0.05) in the characteristic impedance and decrease (P<0.05) in stroke volume and LV power (Table 2). These baseline measurements are consistent with previous data from our laboratory and others using the rat coronary artery ligation model of heart failure [10,11].

3.3. L-NAME and AG dose-dependent changes in hemodynamics

In sham rats, L-NAME increased MAP in a dose-dependent manner (1–1000 mg/kg), but the animals could not tolerate doses higher than 100 mg/kg of L-NAME. Therefore, a dose of 100 mg/kg, which achieved a maximal increase in arterial pressure without distorting the rest of the hemodynamic variables, was chosen in the remainder of the study. In contrast, AG did not effect the hemodynamics at any dose studied (10–80 mg/kg). Doses of AG >80 mg/kg increased MAP due to nonspecific inhibition of eNOS [25]. Therefore, a dose of 75 mg/kg AG was chosen in the remainder studies to insure complete inhibition of the iNOS enzyme.

3.4. Effects of L-NAME and AG on baseline hemodynamics

The administration of L-NAME resulted in the following changes: decreases (P<0.01) in LV dP/dt of 19% in sham rats and 22% in MI rats, a 20% increase (P=0.01) in MAP in sham rats and no change in MI rats, decreases (P<0.01) of 53% in blood velocity in sham rats compared to 55% in MI rats, increases (P<0.01) of 189% in SVR in sham rats and a 108% in MI rats, a 89% increase (P=0.03) in LV end diastolic pressure in sham rats compared to no change in MI rats, similar decreases (P≤0.01) in HR (14% in sham rats and 18% in MI rats), and increases (P≤0.01) 105% in τ in sham rats compared to a 52% in MI rats (Fig. 1). AG (75 mg/kg) had no effect on any hemodynamic variable

Table 1

<table>
<thead>
<tr>
<th>BW (g)</th>
<th>RV (g)</th>
<th>LV (g)</th>
<th>LVEDP (mmHg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sham</td>
<td>403.7±44.7</td>
<td>0.211±0.03</td>
<td>0.86±0.10</td>
</tr>
<tr>
<td>MI</td>
<td>347.1±37.4</td>
<td>0.399±0.08</td>
<td>0.73±0.09</td>
</tr>
</tbody>
</table>

P value 0.005 0.001 0.007 0.001

Values are mean±S.D. n=8 for sham and n=10 for MI.

BW, body weight; RV, right ventricle; LV, left ventricle; LVEDP, LV end diastolic pressure.
Fig. 1. Effects of MI and L-NAME on baseline hemodynamics in sham and MI rats. Filled bars represent control data, unfilled bars represent data after L-NAME, and hatched bars represent data after AG. (a) Heart rate in beats per minute (bpm); (b) mean arterial pressure (MAP); (c) left ventricular dP/dt (LV dP/dt); (d) systemic vascular resistance (SVR); (e) relaxation constant (τ); (f) end-diastolic pressure (EDP) and (g) mean aortic blood velocity (Vmean); n=8–11 in each group. Data presented as mean±S.D.

except it increased (P≤0.01) both HR (13%) and LV dP/dt (29%) in rats after MI only (Fig. 1).

3.5. Effects of MI on β-AR-mediated circulatory response in intact animals

The cumulative isoproterenol dose–response data are presented as the change with each data set (sham and MI) normalized for their respective baselines, i.e., subtracting the baseline response in MI from sham (Fig. 2). In rats after MI, isoproterenol dose–response curves for LV dP/dt and aortic blood velocity were shifted downward, while dose–response curves for SVR and τ were shifted upward (Fig. 2). In both groups of rats, isoproterenol administration did not change LV dP/dt, aortic blood velocity, SVR, or τ at doses between 10⁻⁸ and 2 μg/kg, however, at doses higher than 10⁻² μg/kg, there were progressive decreases in SVR, τ, and aortic blood velocity, while LV dP/dt was progressively increased.

3.6. Effects of β-AR-stimulated NO on steady circulatory response in intact animals

The cumulative isoproterenol dose–response data after L-NAME are presented as the change with each data set (sham and MI) normalized for their respective baselines, i.e., subtracting the baseline response to L-NAME such that the change represents differences from the sham data (Fig. 2). This was done in order to define the effects of NOS inhibition on isoproterenol induced responses independent of baseline effects, which are shown in Fig. 1. In sham rats, at doses >10⁻² μg/kg of isoproterenol, L-NAME potentiated the isoproterenol mediated increases in LV dP/dt and aortic blood velocity and potentiated the isoproterenol-induced reduction in SVR and τ in sham rats (Fig. 2). No such effects were found in MI rats. In contrast, L-NAME did not alter the isoproterenol stimulated MAP or LV end diastolic pressure in either sham or MI rats (data not shown). In addition, no effects of L-
NAME were found on the isoproterenol stimulated HR responses in either group of rats (Fig. 2). In sham rats, interaction between isoproterenol-mediated response and L-NAME is significant \((P<0.05)\) for SVR, aortic blood velocity, \(\tau\), and LV \(dP/dt\), which means that the effects of isoproterenol on these parameters is dependent on the presence of L-NAME, i.e., there is coupling between \(\beta\)-AR and NO pathways. However, this interaction is significant

### Table 2

Effects of L-NAME and AG on pulsatile hemodynamics in sham and MI rats

<table>
<thead>
<tr>
<th></th>
<th>(Z_1)</th>
<th>(Z_2)</th>
<th>(Z_0)</th>
<th>SV</th>
<th>(\Phi)</th>
<th>(P_s)</th>
<th>(P_v)</th>
<th>(P_t)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sham</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>3.729±1.881</td>
<td>2.390±0.952</td>
<td>2.661±0.844</td>
<td>1.54±0.46</td>
<td>-33±11</td>
<td>-119±47</td>
<td>11±5</td>
<td>136±56</td>
</tr>
<tr>
<td>L-NAME</td>
<td>7.286±2.403</td>
<td>4.732±1.107</td>
<td>4.861±1.259</td>
<td>0.72±0.50</td>
<td>-45±7</td>
<td>-62±46</td>
<td>6±7</td>
<td>69±33</td>
</tr>
<tr>
<td>AG</td>
<td>9.441±3.760</td>
<td>5.854±1.434</td>
<td>5.153±2.354</td>
<td>0.72±0.06</td>
<td>-34±35</td>
<td>52±7</td>
<td>3±2</td>
<td>56±9</td>
</tr>
<tr>
<td><strong>MI</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>5.051±1.971</td>
<td>3.656±1.884</td>
<td>4.647±0.868</td>
<td>0.74±0.17</td>
<td>-45±14</td>
<td>53±13</td>
<td>3±2</td>
<td>56±14</td>
</tr>
<tr>
<td>L-NAME</td>
<td>6.199±2.559</td>
<td>4.129±2.066</td>
<td>4.911±2.203</td>
<td>0.44±0.10</td>
<td>-56±8</td>
<td>28±10</td>
<td>1.5±1</td>
<td>30±11</td>
</tr>
<tr>
<td>AG</td>
<td>4.897±2.386</td>
<td>3.703±1.554</td>
<td>4.391±1.758</td>
<td>0.97±0.46</td>
<td>-36±8</td>
<td>83±41</td>
<td>5±2</td>
<td>88±43</td>
</tr>
</tbody>
</table>

Values are mean±S.D. \(Z_1, Z_2, Z_0\), impedance modulus at the 1st and 2nd harmonics, respectively. \(\Phi\) is phase shift; SV, stroke volume (cm³/s); \(P_s, P_v, P_t\), steady state, pulsatile and total cardiac output power.

\(* P<0.05\) between sham and MI; \(\dagger P<0.05\) within group between baseline and treatment with either L-NAME or AG \((n=10\) for baseline and \(n=4\) per treatment each group).

\(Z_c\), the characteristic impedance modulus.
Fig. 2. Effects of L-NAME on isoproterenol-mediated hemodynamic response in sham and MI rats. Data are presented as the change with each data set (sham and MI) normalized for their respective baselines. (a) Change in heart rate in beats per minute (ΔHR, bpm); (b) change in left ventricular dP/dt (ΔLV dP/dt); (c) Change in systemic vascular resistance (ΔSVR); (d) change in relaxation constant (Δτ) and (e) change in mean aortic blood velocity (ΔVmean); n=8–11 in each group. Each data point presented as mean±S.D. * P<0.05 L-NAME compared to untreated.
only for SVR in MI rats. AG (75 mg/kg) had no effect on isoproterenol dose–responses in either group of rats (data not shown).

3.7. Effects of L-NAME and AG on pulsatile hemodynamics

In sham rats, L-NAME increased \((P<0.05)\) the impedance moduli \(Z_1\) and \(Z_2\), the characteristic impedance \(Z_0\), and the phase shifts \(\Phi_1\) and \(\Phi_2\) (Table 2). In contrast, stroke volume, pulsatile and total ventricular power were decreased \((P<0.05)\) with L-NAME. However, AG increased \((P<0.05)\) the impedance moduli \(Z_1\) and \(Z_2\), the characteristic impedance \(Z_0\), and decreased \((P<0.05)\) stroke volume and total ventricular power. In MI rats, the only changes due to L-NAME were increases \((P<0.05)\) in \(Z_1\) and \(\Phi_1\), and a decrease \((P<0.05)\) in LV power. AG administration did not affect any of the pulsatile hemodynamic parameters in MI rats.

4. Discussion

The current study examines the hemodynamic effects of NOS inhibition in intact animals after MI. The study design allowed the separation of the effects of basal and \(\beta\)-AR-stimulated NO release on pulsatile and steady-state arterial function. Our major findings are that, in this compensated model of ischemic heart failure, (1) circulatory and cardiac responses to inhibition of NO by L-NAME suggest that eNOS, but not iNOS, is the principal regulator of integrated arterial hemodynamic function in rats. (2) Both basal and \(\beta\)-AR-stimulated NO regulation of hemodynamic are attenuated after MI. (3) The attenuation of arterial hemodynamic effects after isoproterenol is mediated, in part, by alterations in the \(\beta\)-AR-activation of eNOS system after MI.

4.1. Effects of NO inhibition on in vivo steady-state hemodynamics

L-NAME had a hypertensive effect in sham rats, with no measurable effect in MI rats. Previous work suggests that the increase in arterial blood pressure in sham rats probably results from blocking the vasodilatory action of NO, rather than from any direct effect of NOS inhibition on the sympathetic nervous system [14–17]. In contrast to the effect of L-NAME in sham rats, NOS inhibition had negligible effects on arterial pressure in rats after MI. Since arterial blood pressure is the product of cardiac output and SVR, the lack of effect on blood pressure was the result of opposite but balanced effects of L-NAME on these two variables in MI rats.

The differential effect of NOS inhibition via L-NAME on arterial pressure in sham versus MI is the result of either more intense vasoconstriction in sham rats, or a greater decrease in cardiac output (aortic flow velocity), relative to vascular constriction, in MI rats. While the former mechanism could only be proven by direct measurement of peripheral arteriolar vasomotion in response to L-NAME, the latter mechanism is the plausible result of greater afterload sensitivity in the setting of LV dysfunction. A decrease in cardiac output due to any direct influence of NO inhibition is not likely, since most investigators believe NO has a negative contractile effect. By similar reasoning, the decrease in LV \(dP/dt\) is probably the result of increased LV afterload rather than due to decreased cardiac contractility.

In contrast to the effects of L-NAME, AG did not affect steady-state hemodynamics in sham rats. This suggests that iNOS does not significantly regulate normal cardiovascular function. However, in MI rats, LV \(dP/dt\) was increased, in the absence of any effects of AG on SVR or LV EDP (LV pre-load). Thus, it is possible that LV \(dP/dt\) increased after AG because the potentially positive contractile effects of iNOS inhibition were unopposed by any peripheral vasoconstrictive effects. Since AG did not increase either MAP or SVR, we concluded that iNOS inhibition had no effect in the resistance vessels. Further, since LV \(dP/dt\) is afterload-dependent and this afterload did not change with AG and since NO is known to depress cardiac contractility, the increase in LV \(dP/dt\) with AG may imply a role for iNOS in the heart.

4.2. Effects of NO inhibition on in vivo pulsatile hemodynamics

While resistance arteries modulate steady-state hemodynamics, large arteries primarily determine pulsatile hemodynamics (aortic input and characteristic impedance). The findings that basal aortic input impedance and phase shift are increased, and that ventricular power is decreased in rats after MI are in agreement with a previous study from our laboratory [10]. Aortic input impedance is a function of loading and arterial radius, while the characteristic impedance is proportional to aortic stiffness [18]. Therefore, while the increases in impedance moduli and the decrease in stroke volume with L-NAME is explained by the vasoconstrictive effects of NO inhibition, the increase in characteristic impedance in sham rats with L-NAME is the first demonstration that NO also modulates vascular stiffness. This effect is associated with decreased ventricular power. In contrast, L-NAME does not further increase either aortic input or characteristic impedance after MI. This lack of response to NOS inhibition after MI is probably explained by the well established observation that endothelial-dependent NO release is diminished after MI [4].

Despite the absence of effects of iNOS inhibition on peripheral resistance, it increased aortic input impedance and decreased stroke volume in sham rats to the same degree as non-specific NOS inhibition with L-NAME.
These data suggest, therefore, that iNOS controls vasomotion in large, but not resistance, vessels in sham rats. However, the lack of effect of AG on pulsatile hemodynamics in MI rats suggests that iNOS may be downregulated in the large arteries in the setting of heart failure. Combined with the observations on steady-state hemodynamics, these data suggest that the iNOS isoform plays a major role in modulating vasomotion in large conduit arteries while eNOS controls primarily resistance vessels in sham rats. In addition, aortic input impedance is a function of the mechanical properties of large arteries as well as the contractility of the heart. Since elevation of LV dP/dt after AG may indicate a role of iNOS in the myocardium, as stated above, iNOS effects on cardiac muscle contractility may provide another possible explanation for the effects of AG on aortic input impedance.

4.3. Effects of NO inhibition on β-AR hemodynamic responses

Our data show that NOS inhibition in intact animals results in potentiation of LV dP/dt, SVR, and aortic blood velocity responses to β-AR stimulation in sham rats. In contrast, NOS inhibition with L-NAME had no effect on the responses to β-AR stimulation in MI rats. The data in the sham group support the hypothesis that NO inhibition modulates arterial hemodynamic responses to β-AR stimulation [19] and are consistent with previous work in normal conscious dogs and in humans with LV dysfunction [20,21]. The apparent absence of NO modulation of adrenergic-stimulated responses in MI rats could be the result of reduced endothelial NO production, a partial decrease in β-AR responsiveness, and/or uncoupling of the NO and AR systems in the peripheral vasculature after MI [1,3,4,14,16,22]. While the exact mechanism(s) remain unclear, this is the first study to demonstrate an abnormal linkage in the NO–β-AR systems in the presence of heart failure due to MI.

The NO-mediated circulatory responses, reported here, are due to blockade of the NOS in the whole animal. The vasoconstrictive effect of NO inhibition may be due to blockade of NO release not only from the endothelium, vascular smooth muscle cells, cardiac cells, but also perivascular nitrooxidergic nerves. Previous studies in dogs and monkeys have shown the presence of NOS containing nerve fibers in the arterial wall [23,24], that can be activated by ganglionic stimulant to produce NO and vasorelaxation. However, neurogenic NO plays a minor role in vasorelaxation in rats compared to its effects in dogs and monkeys [7,23,24]. In the current study, we examined the hemodynamic effects of NO produced by basal as well as β-AR-stimulation of eNOS enzyme. We did not, however, study the effect of iNOS induction, e.g., by lipopolysaccharides [25].

In summary, our data show that in sham rats, iNOS modulates arterial hemodynamics in large, conduit arteries while eNOS is the primary NOS regulator of peripheral resistance vessels. The control of arterial hemodynamics by NOS is markedly attenuated in MI rats. In addition, we present evidence that endothelial dysfunction is mediated by alterations in the NOS system and contributes to β-AR hypo-responsiveness in heart failure due to MI.

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