Effect of Angiotensin AT₁ Receptor Blockade on Sympathetic Responses to Handgrip in Healthy Men

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BACKGROUND
To determine whether angiotensin II (ANG II) contributes to the reflex skeletal muscle sympathoexcitation elicited by isometric and isotonic exercise, we tested the hypothesis that angiotensin AT₁ receptor blockade (ARB) would attenuate reflex sympathoneural responses to handgrip (HG) and to post-handgrip ischemia (PHGI).

METHODS
Seventeen healthy men were studied before and 1 week after random double-blind crossover allocation to oral losartan (100 mg daily) and placebo. Heart rate (HR), blood pressure (BP), and muscle sympathetic nerve activity (MSNA) were recorded at rest, and during 2 min bouts of isometric HG at 50% maximum voluntary contraction (MVC) and isometric HG at 30% MVC, performed randomly, each followed by 2 min of PHGI.

RESULTS
At rest, losartan doubled plasma renin (P < 0.01) and ANG II (P < 0.03) concentrations, and lowered BP (P < 0.01) yet had no effect on MSNA burst frequency or incidence. HR trended higher (P = 0.060). Losartan’s hypotensive effect persisted throughout each exercise bout (P < 0.045). MSNA and HR responses to isometric exercise and postexercise ischemia were not affected by losartan. Isometric exercise and postexercise ischemia increased MSNA on both sessions (all P < 0.01). Losartan augmented the HR response (P ≤ 0.03), and after losartan MSNA burst frequency (P < 0.01) and incidence (P < 0.04) were significantly higher at all time points, but the magnitude of the MSNA response to isometric exercise and postexercise ischemia was unchanged.

CONCLUSION
In healthy men, short-term ARB does not attenuate reflex sympathoneural responses to HG or PHGI.

Keywords: adenosine; blood pressure; hypertension; losartan; metaboreflex; muscle sympathetic nerve activity

American Journal of Hypertension, advance online publication 17 February 2011; doi:10.1038/ajh.2011.14

Angiotensin II (ANG II) increases efferent sympathetic nerve discharge and neural norepinephrine (NE) release by stimulating central neural, ganglionic, and prejunctional type 1 (AT₁) receptors.¹⁻³ Although widely prescribed for the management of hypertension and heart failure, our knowledge in humans of the impact of angiotensin AT₁ receptor blockade (ARB) on sympathetic outflow is based almost exclusively upon observations acquired from subjects studied under resting conditions.⁴⁻⁷ Thus far, the impact of ARB on the muscle sympathetic nerve activity (MSNA) response to exercise has not been characterized.

In our previous experiments in healthy young men, pretreatment with losartan (100 mg daily) had no effect on total body NE spillover at rest, but attenuated, by 80%, the reflex increase in total body NE spillover elicited by brachial artery infusion (in doses without systemic effect) of adenosine,² indicating the participation of ANG II AT₁ receptors within the central nervous system, or at efferent (e.g., ganglionic) sites, or both, in the elicitation of this response. Adenosine is one of the metabolites of exercise shown to accumulate interstitially during both dynamic⁸,⁹ and isotonic¹⁰ exercise, and activate the muscle metaboreflex by stimulating thin-fiber muscle afferents that constitute its afferent limb.¹⁰,¹¹

The aim of this study was to determine whether ANG II participates in the reflex skeletal muscle sympathoexcitation elicited by isometric and intense isotonic handgrip (HG) exercise in healthy men. We hypothesized that ARB with the antagonist losartan, which has been shown to bind to AT₁ receptors at sites both within and outside the blood–brain barrier involved in cardiovascular regulation by the autonomic nervous system,¹² would attenuate the increase in muscle sympathetic nerve firing elicited by isometric and intense isotonic HG exercise and also by post-handgrip ischemia (PHGI). By studying MSNA, we excluded any confounding influence of peripheral prejunctional AT₁ receptors on neural NE release.²

METHODS
Participants. Seventeen healthy, unmedicated, nonsmoking male volunteers (mean age: 31 ± 11 years; mean height: 177 ± 7 cm;
mean weight: 75 ± 11 kg) participated in this study. The Research Ethics Board of the University Health Network approved the protocol, ensuring compliance with the Declaration of Helsinki, and all participants provided in advance written informed consent.

**Study design.** Participants were allocated by pharmacy-prepared double-blind randomization to either a losartan-start group (100 mg daily) or a placebo-start group. All tablets were taken once daily for 7 days. After a washout period of at least 3 weeks, subjects then ingested daily for 7 days the last dose of losartan or placebo.

**Experimental protocol.** Data were acquired in the morning, in a quiet, temperature-controlled room, after 72 h of abstinence from caffeine and a light caffeine-free breakfast. After voiding (to minimize any effects of progressive bladder distension on blood pressure (BP)), participants lay supine for the duration of the protocol. First, the force of each participant’s maximum voluntary contraction (MVC) was determined by averaging the values achieved by the nondominant arm during three maximum HG efforts (handgrip dynamometer, model 78010; Lafayette Instrument, Lafayette, IN). A polyethylene catheter was then inserted into a right antecubital vein for blood sampling (plasma NE, epinephrine, dopamine, plasma renin, and ANG II concentrations). Lead II of the electrocardiogram was recorded continuously to derive heart rate (HR). Noninvasive oscillometric determinations of brachial artery pressure in the dominant arm (standard adult cuff: 23–33 cm; Dinamap Pro 100; Critikon, Tampa, FL), were acquired each minute. Breathing patterns were recorded by a pneumobelt connected to a pressure transducer. Multi-unit recordings of postganglionic MSNA were obtained with a unipolar tungsten electrode inserted selectively into a muscle–nerve fascicle of the right fibular (peroneal) nerve.

After 15–20 min of supine rest and spontaneous breathing, these data were acquired over a 7 min minimum baseline period, after which venous blood was drawn. Next, to determine whether there was a dose–response relationship between graded mechanoreceptor activation plus tissue hypoperfusion in the presence or absence of losartan (as stimulus) and MSNA (as response), using their nondominant hand subjects performed in random order 2 min bouts of 50% MVC isometric HG (during which relaxations between contractions allow efflux of ischemic metabolites from the forearm), and 30% MVC isometric HG (a stimulus characterized by sustained muscle contraction, hypoperfusion, and ischemia). To dissociate metabolic stimulation of muscle afferents from the potentially confounding sympathoexcitatory effects of muscle contraction or central command, both exercises were followed immediately by PHGI. An upper arm cuff was inflated to 200 mm Hg for 2 min to trap within the forearm metabolites generated by exercise. After cuff deflation, recordings continued over an additional 2 min (recovery) period.

Each experimental session concluded with a 1.5 min cold pressor test (CPT); immersion of the dominant hand in ice water) as an internal positive control for reflex sympathoexcitatory capacity.

**Data analysis.** Signal output was inscribed by a Gould Viper recorder (Gould Instrument Systems, Madison, WI), sampled at a frequency of 200 Hz (with the exception of the electrocardiogram: 1,000 Hz) and following conversion from analogue to digital format stored on a PC desktop for subsequent offline analysis.

Average MSNA burst frequency (bursts/min), burst incidence (bursts/100 cardiac cycles) and integrated MSNA (bursts frequency × mean amplitude; expressed in arbitrary units) were determined using LabVIEW based custom software (National Instruments, Austin, TX). Integrated MSNA was normalized to the maximum burst amplitude (peak height) detected during each recording (normalized integrated MSNA; expressed in arbitrary units/min). If the baseline signal was affected by electrode drift, bursts were recalibrated against the subsequent maximum amplitude.

As in our previous HG experiments, values for data acquired during each HG session were averaged over five discrete time segments for subsequent analysis: the 2 min of rest before HG; separately for the first and second minute of HG; the 2 min of PHGI; and the 2 min after occlusion cuff deflation (HG-recovery). For the CPT, average values for HR, MSNA, and minute-by-minute BP were calculated for the 2 min baseline period before, the period of, and a 2 min recovery period after hand immersion.

Toronto Medical Laboratories (Toronto, Ontario, Canada) assayed plasma NE, epinephrine, and dopamine concentrations using high-performance liquid chromatography with electrochemical detection. Plasma renin concentration was measured by an immunoradiometric sandwich procedure (Renin III generation kit; Cisbio US, Bedford, MA) with a pair of monoclonal antibodies. The first monoclonal antibody, adsorbed on a polystyrene tube, specifically recognizes both the active and the inactive form of renin. The second monoclonal antibody labeled with 125Iodine specifically recognizes the active form of renin. ANG II in plasma was measured by radioimmunoassay (Angiotensin II 125I RIA kit; Bühmann Laboratories, Schönenbuch, Switzerland) after solid phase extraction with phenylsilylsilica cartridges.

**Statistical analysis.** Data are presented as mean ± s.d., unless otherwise stated. All data were recorded, analyzed, confirmed, and tabled with all investigators blinded to tablet allocation. Paired t-tests were administered to test for differences between losartan and placebo on all normally distributed dependent resting variables. Signed rank sum tests were performed on non-normally distributed data. Two-way (drug × time) ANOVA with repeated measures were used to assess MSNA, HR, and BP in participants with paired high quality MSNA recordings during exercise, PHGI (n = 15) and CPT (n = 12) portions of the experimental protocol. Student Newman–Keuls post hoc procedures were employed to evaluate specific differences between means, where applicable. All data were analyzed
using Sigma Stat for Windows (ver 3.5; Jandel Scientific, San Rafael, CA), and an α level of ≤0.05 was considered statistically significant.

RESULTS
Resting values
Losartan increased plasma renin \((P = 0.01)\) and ANG II \((P = 0.03)\) concentrations (Table 1) and lowered both systolic and diastolic BP \((P < 0.01; \text{Table 1})\). HR trended higher \((P = 0.060)\). By contrast, no differences were observed on losartan and placebo days with respect to: MSNA burst frequency \((P = 0.32)\), burst incidence \((P = 0.51)\), normalized integrated MSNA \((P = 0.44)\), plasma NE \((P = 0.54)\), epinephrine \((P = 0.43)\), or dopamine \((P = 1.00)\) concentrations (Table 1).

Responses to isotonic HG and PHGI
On both study days, MSNA burst frequency rose significantly above resting values by the first minute of 50% isotonic HG exercise, and remained elevated during PHGI \((all \ P ≤ 0.01)\) (Figure 1), as did normalized integrated MSNA (product of burst frequency and burst amplitude, normalized for peak amplitude; all \(P < 0.04\); data not shown). The sympatheural response to isotonic HG exercise and PHGI (absolute change from baseline) was similar on the losartan and placebo days \((P = 0.41\) and 0.93, respectively) (Table 2).

Mean arterial pressure (MAP) was lower on the losartan day throughout the interventions \((P = 0.045)\), however, it rose significantly above resting values and remained elevated throughout the 50% isotonic HG bout and PHGI \((all \ P ≤ 0.01)\) (Figure 1). The MAP responses to isotonic exercise and PHGI were similar in magnitude on the 2 study days \((P = 0.33\) and 0.21, respectively).

Response to isometric HG and PHGI
On both study days, MSNA burst frequency rose significantly above resting values during 30% isometric HG and remained elevated above baseline during PHGI \((all \ P ≤ 0.01)\), as did normalized integrated MSNA \((all \ P < 0.01\); data not shown). Throughout these interventions burst frequency was significantly higher on the losartan day \((P < 0.01\); Figure 2), as was normalized integrated MSNA \((P = 0.02)\). However, the magnitude of the MSNA response to isometric HG (absolute change from baseline), whether expressed as burst frequency \((P = 0.40)\), or normalized integrated MSNA \((P = 0.21)\), was similar on the losartan and placebo study days, as was the magnitude of the MSNA response to PHGI, whether expressed as burst frequency \((P = 1.00)\) or normalized integrated MSNA \((P = 0.53)\) (Table 2).

On the losartan study day, the HR increase from baseline was significantly greater in magnitude during both HG and PHGI (interaction \(P ≤ 0.03\)) (Figure 2). During HG, HR increased from baseline by \(14.8 \pm 10.8\) min\(^{-1}\) on the placebo day, and by \(20.3 \pm 10.7\) min\(^{-1}\) on the losartan day \((P < 0.01)\); corresponding PHGI values were +0.5 ± 4.3 and +4.1 ± 5.2 min\(^{-1}\) \((P = 0.30)\).

MSNA burst incidence was elevated above baseline by the second minute of isometric HG, and remained higher during PHGI \((all \ P < 0.01)\) (Figure 2). MSNA burst incidence was also consistently greater throughout these interventions on the losartan day \((P = 0.04)\). However, the magnitudes of both the MSNA response to isometric HG (absolute change from baseline) and to PHGI were similar on the losartan and placebo study days \((P = 0.88\) and 0.57 respectively) (Table 2).

On both study days, MAP, which was lower on the losartan day throughout these interventions \((P < 0.01)\), rose significantly above resting values by the second minute of isometric HG \((all \ P < 0.04)\) (Figure 2). MAP responses to isometric exercise and to PHGI were similar in magnitude on these 2 days \((both \ P ≤ 0.21)\).

Comparison of responses to isometric vs. isotonic HG
Absolute increases in MAP from baseline were similar for 30% isometric and 50% isotonic HG exercise (+11.2 ± 1.6 mm Hg vs. +9.6 ± 2.9 mm Hg, respectively; \(P = 0.38)\). Isometric exercise elicited a greater sympatheural response when represented

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Table 1 | Hemodynamic and humoral effects of losartan on resting values

<table>
<thead>
<tr>
<th>Variable</th>
<th>Placebo</th>
<th>Losartan</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Systolic blood pressure (mm Hg)</td>
<td>121 ± 15</td>
<td>113 ± 11</td>
<td>0.001</td>
</tr>
<tr>
<td>Mean arterial blood pressure (mm Hg)</td>
<td>84 ± 11</td>
<td>78 ± 10</td>
<td>0.001</td>
</tr>
<tr>
<td>Diastolic blood pressure (mm Hg)</td>
<td>66 ± 10</td>
<td>60 ± 10</td>
<td>0.001</td>
</tr>
<tr>
<td>Heart rate (b.p.m.)</td>
<td>60 ± 10</td>
<td>63 ± 8</td>
<td>0.060</td>
</tr>
<tr>
<td>Muscle sympathetic nerve activity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bursts/min</td>
<td>22 ± 11</td>
<td>26 ± 10</td>
<td>0.32</td>
</tr>
<tr>
<td>Bursts/100 beats</td>
<td>39 ± 18</td>
<td>41 ± 17</td>
<td>0.51</td>
</tr>
<tr>
<td>Normalized integrated (units/min)</td>
<td>9 ± 6</td>
<td>10 ± 6</td>
<td>0.44</td>
</tr>
<tr>
<td>Norepinephrine (pg/ml)</td>
<td>169 ± 101</td>
<td>186 ± 68</td>
<td>0.54</td>
</tr>
<tr>
<td>Epinephrine (pg/ml)</td>
<td>55 ± 37</td>
<td>55 ± 18</td>
<td>0.43</td>
</tr>
<tr>
<td>Dopamine (pg/ml)</td>
<td>23 (15, 31)</td>
<td>15 (15, 31)</td>
<td>1.00</td>
</tr>
<tr>
<td>Angiotensin II (pg/ml)</td>
<td>1.8 (1.2, 4.4)</td>
<td>11.8 (8.0, 43.6)</td>
<td>0.03</td>
</tr>
<tr>
<td>Plasma renin (ng/l)</td>
<td>24.6 ± 51.4</td>
<td>58.8 ± 43.1</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Values are mean ± s.d. for normally distributed data, and median with 25% and 75% confidence intervals for non-normally distributed data, \(n = 15\).

b.p.m., Beats per minute.
AT1 Receptor Blockade and MSNA

as burst frequency (+13.6 ± 9.4 bursts/min vs. +7.5 ± 4.5 burst/min, respectively; \( P = 0.03 \)) but not when expressed as burst incidence (\( P = 0.17 \)) or as normalized integrated MSNA (\( P = 0.28 \)).

Responses to CPT
In 12 of these participants, the CPT, a nonmetaboreflex internal control, elicited significant increases in MAP (\( P \leq 0.01 \)), HR (\( P < 0.01 \)), MSNA burst frequency (\( P < 0.01 \)), MSNA burst incidence (\( P \leq 0.01 \)) and normalized integrated MSNA (\( P \leq 0.01 \)). These responses were similar on the 2 study days (all \( P > 0.22 \); Table 3).

DISCUSSION
Our previous experiments involving healthy young men (also studied in the presence or absence of losartan 100 mg daily given over a period of 1 week) had implicated ANG II as a potential neuromodulator of the skeletal muscle metaboreflex.2 The purpose of this study was to determine whether ANG II facilitates the net integrated reflex skeletal muscle sympathoexcitation elicited by intense isotonic and isometric HG exercise in healthy humans, without specifying, at this juncture, the precise neural site or sites of such participation. We observed no significant effect of losartan on the magnitude

TABLE 2 | Sympathoneural response (increase from baseline) to 2 min of isotonic or isometric handgrip (HG) and 2 min of post-handgrip ischemia (PHGI)

<table>
<thead>
<tr>
<th>Muscle sympathetic nerve activity</th>
<th>Placebo</th>
<th>Losartan</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>50% isotonic HG2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bursts/min</td>
<td>+7.5 ± 4.5</td>
<td>+7.5 ± 5.8</td>
<td>0.81</td>
</tr>
<tr>
<td>Bursts/100 beats</td>
<td>+4.0 ± 6.2</td>
<td>+2.5 ± 8.9</td>
<td>0.41</td>
</tr>
<tr>
<td>Normalized integrated (units/min)</td>
<td>+4.7 ± 3.9</td>
<td>+4.4 ± 4.5</td>
<td>0.78</td>
</tr>
<tr>
<td>PHGI following 50% isotonic HG</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bursts/min</td>
<td>+4.5 ± 4.0</td>
<td>+6.1 ± 4.4</td>
<td>0.40</td>
</tr>
<tr>
<td>Bursts/100 beats</td>
<td>+7.9 ± 2.4</td>
<td>+7.6 ± 7.2</td>
<td>0.93</td>
</tr>
<tr>
<td>Normalized integrated (units/min)</td>
<td>+2.9 ± 2.4</td>
<td>+3.7 ± 3.4</td>
<td>0.57</td>
</tr>
<tr>
<td>30% isometric HG2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bursts/min</td>
<td>+13.6 ± 9.4</td>
<td>+17.6 ± 12.4</td>
<td>0.40</td>
</tr>
<tr>
<td>Bursts/100 beats</td>
<td>+9.9 ± 14.8</td>
<td>+10.1 ± 13.1</td>
<td>0.88</td>
</tr>
<tr>
<td>Normalized integrated (units/min)</td>
<td>+6.7 ± 2.4</td>
<td>+11.5 ± 7.8</td>
<td>0.21</td>
</tr>
<tr>
<td>PHGI following 30% isometric HG</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bursts/min</td>
<td>+7.7 ± 6.8</td>
<td>+8.4 ± 7.8</td>
<td>0.00</td>
</tr>
<tr>
<td>Bursts/100 beats</td>
<td>+12.3 ± 12.6</td>
<td>+10.5 ± 12.3</td>
<td>0.57</td>
</tr>
<tr>
<td>Normalized integrated (units/min)</td>
<td>+4.6 ± 5.1</td>
<td>+5.8 ± 5.5</td>
<td>0.53</td>
</tr>
</tbody>
</table>

Values are mean ± s.d., \( n = 15 \).

Figure 1 | Fifty percent isotonic handgrip protocol. (a) Mean arterial pressure (MAP), (b) heart rate (HR), (c) muscle sympathetic nerve activity (MSNA) burst frequency, and (d) burst incidence at rest (PRE), during first (HG1) and second (HG2) min of handgrip, during 2 min of post-handgrip ischemia (PHGI) and 2 min of recovery (REC) after 1 week of daily losartan, 100 mg, (closed circle) or placebo (open circle). Values represent means ± s.e., \( n = 15; n = 14 \) for MAP. Stated \( P \) values refer to significance level of main effect for losartan. * \( P \leq 0.01 \) vs. pre-HG. b.p.m., beats per minute.
of MSNA responses to either mode of HG, or to post-HG trapping of ischemic metabolites, such as adenosine, in the forearm. In interpreting these findings we will discuss briefly the exercise stimuli, the drug intervention, the model and population selected, and the study hypothesis itself.

These stimuli were clearly sufficient to increase significantly MSNA burst frequency and incidence on both the placebo and losartan study days (Figures 1 and 2, Table 2). The less intense isotonic protocol is of particular interest, as in this instance burst incidence was highest during PHGI ischemia. Thus, if losartan had exerted a significant inhibitory effect on the reflex sympathoneural responses to the accumulation of ischemic metabolites of exercise, this action might have been specifically evident at that time. However, as illustrated in Figure 1, no such interaction was detected, either during PHGI or during HG itself.

Losartan doubled plasma renin and ANG II concentrations and lowered BP at rest, and throughout both isotonic and isometric challenges, confirming both participant adherence to protocol and the efficacy of angiotensin ARB. Also, in so far as the reduction in baseline BP was not accompanied by any reflex increase in resting MSNA or plasma catecholamines,
the dose of losartan selected was evidently sufficient to alter neural circulatory control at rest. HR tended to be higher (3 beats/min) at baseline on the losartan day, but this finding did not achieve prespecified statistical significance. Importantly, at baseline, HR-adjusted MSNA was no different on the 2 study days. In our previous randomized double blind crossover trial, also involving healthy young men, losartan lowered resting BP significantly, also without affecting total body NE spillover, forearm NE appearance rate, forearm vascular resistance, HR, arterial baroreflex sensitivity for HR, or power in the high- or low-frequency components of the HR power spectrum.2

Consistent with the findings of Scherrer and colleagues,17 who observed higher MSNA and HR when they countered the pressor response to isometric HG using sodium nitroprusside, in the present series the lower MAPs throughout all periods of isometric exercise were accompanied by higher MSNA burst frequency and incidence (Figure 2; main effect of losartan). Lower MAPs were also evident throughout the isotonic exercise protocol, but by contrast, these were not accompanied by increases in MSNA burst incidence (Figure 1). Importantly, the magnitude of the sympathoexcitatory response to either mode of HG or of PHGI was unaffected. Recently Ogoh et al.18 reported that metaboreflex activation by PHGI following 30% MVC exercise increased low frequency transfer function gain between BP and MSNA. Their findings raise the possibility that had an inhibitory effect of losartan been present at this specific time point, this action might have been obscured by a concurrent increase in baroreflex gain.

In contrast to our prior and present finding that 7 days of losartan 100 mg lowered BP significantly but had no effect on resting total body NE spillover, plasma NE, or MSNA, other investigators found that another AT1 antagonist eprosartan, 600 mg, raised plasma ANG II concentrations of healthy young men fivefold, induced a lesser reduction in mean BP, and elicited a significant 45% increase in resting MSNA burst frequency and a 36% increase in MSNA burst incidence.4 However, as in this study, reflex MSNA responses to lower body negative pressure and mental stress were not attenuated by eprosartan. In that report, the higher MSNA at rest was attributed to the marked increase in circulating ANG II concentrations, acting on central AT1 receptors to reset the arterial baroreflex modulation of MSNA so as to enhance rather than attenuate the anticipated response to a lower BP.4

In the present series, losartan doubled plasma ANG II concentrations. Thus, it is also conceivable that these observations reflect augmentation of the mechanoreflex, in an effort to maintain perfusion pressure within contracting muscle, by enhanced stimulation of unblocked central AT2 receptors.19 Having found arterial baroreceptor-HR reflex gain to be greater in AT2 receptor deleted mice than in wild type mice, Gross et al.20 concluded that AT1 receptor stimulation inhibits arterial baroreflex function, likely via a central action. Of note, in our study participants, there was a significant losartan interaction with respect to the HR response to the more intense stimulus of isometric HG, suggesting baroreflex-mediated chronotropic augmentation of cardiac output so as to maintain skeletal muscle perfusion in the setting of lower systemic BP. In rats anesthetized with urethane, microinjection of losartan into the paraventricular hypothalamic nucleus reduced MAP without altering renal sympathetic nerve activity, whereas the AT2 receptor antagonist PD123319 lowered renal sympathetic nerve activity,21 a finding consistent with a role for central AT1 receptors in the tonic regulation of central sympathetic outflow. These lines of evidence suggest that the main effect of losartan observed for MSNA and HR during isometric HG might reflect higher ANG II concentrations acting on unblocked central AT2 receptors, with sympathoexcitatory consequences.

Exercise induced concentrations of adenosine at afferent nerve endings, which were not assessed in this study, may not be comparable to those achieved in the direct brachial artery infusion experiment that stimulated the hypothesis now tested.2 Others have reported a reduction in MSNA during isometric HG in response to local blockade of prostaglandin synthesis in healthy humans,22 as well as to a lowering of blood lactate during hyperoxia.23 Importantly, in a previous study from our laboratory, nonspecific blockade of adenosine receptors with caffeine did not alter MSNA during HG exercise or PHGI in healthy subjects.24 By contrast, a subsequent reduction was observed in congestive heart failure patients during PHGI following 30% isometric and 50% isotonic HG.24

The principal conclusion from this study is that in healthy young men short-term ARB does not attenuate the net integrated reflex sympathoneural response to isometric or intense isotonic HG or PHGI. Although one might speculate that a higher dose of losartan might yield different results, we consider this to be both unlikely (given the magnitude of the inhibitory effect of this dose on reflex sympathoexcitation in a similar population in our previous experiments,2 its effect on the relationship between resting diastolic BP and MSNA in the present series, and the accessibility of this antagonist to AT1 receptors lying outside the blood brain barrier)25 and if indeed the case, of limited relevance to clinical practice. Although it is not possible to draw definitive conclusions as to site of action and specifically the central neural disposition of losartan from the present findings, in that the angiotensin AT1 receptor is distributed widely throughout the central nervous system and sympathetic ganglia, it is possible that this neutral net effect in healthy young men represents the summation or interaction of the effects of losartan on AT1 receptors exerting sympathoexcitatory actions at some sites, and sympathoinhibitory effects at others. Such balance may well be altered in conditions characterized by alterations in AT1 and AT2 receptor number, distribution, or affinity for the endogenous agonist. Thus, future experiments should evaluate whether the net integrative response to HG exercise is altered in sodium restricted or hypovolemic healthy subjects, in women, or in conditions such as hypertension or congestive heart failure. Replication of the present protocol in such conditions would be of particular interest, as ARB has been reported recently to increase significantly interindividual variation in systolic BP in hypertensive patients, rather than decrease such variation,
as is observed in those treated with diuretics or calcium channel antagonists.26

Acknowledgments: This investigation was supported by an Operating Grant from the Canadian Institutes of Health Research (MT9271). C.M. was the recipient of a Heart and Stroke Foundation of Ontario Fellowship. J.F. is a Career Investigator of the Heart and Stroke Foundation of Ontario and holds the Canada Research Chair in Integrative Cardiovascular Biology.

Disclosure: The authors declared no conflict of interest.