Sympathetic Nonadrenergic Transmission Contributes to Autonomic Dysreflexia in Spinal Cord–Injured Individuals


_Hypertension_. 2010;55:636-643; originally published online January 25, 2010; doi: 10.1161/HYPERTENSIONAHA.109.147330

_Hypertension_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2010 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/55/3/636

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in _Hypertension_ can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to _Hypertension_ is online at:
http://hyper.ahajournals.org//subscriptions/
Sympathetic Nonadrenergic Transmission Contributes to Autonomic Dysreflexia in Spinal Cord–Injured Individuals


Abstract—Autonomic dysreflexia is a hypertensive episode in spinal cord–injured individuals induced by exaggerated sympathetic activity and thought to be α-adrenergic mediated. α-Adrenoceptor antagonists have been a rational first choice; nevertheless, calcium channel blockers are primarily used in autonomic dysreflexia management. However, α-adrenoceptor blockade may leave a residual vasoconstrictor response to sympathetic nonadrenergic transmission unaffected. The aim was to assess the α-adrenergic contribution and, in addition, the role of supraspinal control to leg vasoconstriction during exaggerated sympathetic activity provoked by autonomic dysreflexia in spinal cord–injured individuals and by a cold pressure test in control individuals. Upper leg blood flow was measured using venous occlusion plethysmography during supine rest and during exaggerated sympathetic activity in 6 spinal cord–injured individuals and 7 able-bodied control individuals, without and with phenolamine (α-adrenoceptor antagonist) and nicardipine (calcium channel blocker) infusion into the right femoral artery. Leg vascular resistance was calculated. In spinal cord–injured individuals, phenolamine significantly reduced the leg vascular resistance increase during autonomic dysreflexia (8 ± 5 versus 24 ± 13 arbitrary units; \(P = 0.04\)) in contrast to nicardipine (15 ± 10 versus 24 ± 13 arbitrary units; \(P = 0.12\)). In controls, phenolamine completely abolished the leg vascular resistance increase during a cold pressure test (1 ± 2 versus 18 ± 14 arbitrary units; \(P = 0.02\)). The norepinephrine increase during phenolamine infusion was larger (\(P = 0.04\)) in control than in spinal cord–injured individuals. These results indicate that the leg vascular resistance increase during autonomic dysreflexia in spinal cord–injured individuals is not entirely α-adrenergic mediated and is partly explained by nonadrenergic transmission, which may, in healthy subjects, be suppressed by supraspinal control. (Hypertension. 2010;55:636-643.)

Key Words: autonomic dysreflexia ▪ spinal cord injury ▪ sympathetic nervous system ▪ neurotransmitters ▪ leg vascular resistance

Autonomic dysreflexia (AD) is a potentially life-threatening episodic hypertension that develops in 80% to 90% of spinal cord–injured (SCI) individuals with a spinal cord lesion at or above the sixth thoracic spinal segment (T6). AD occurs in these SCI individuals because a large part of the sympathetic nervous system is without central inhibitory pathways. An arterial pressure increase is induced by exaggerated sympathetic activity caused by visceral, noxious, or nociceptive stimuli entering the spinal cord below the level of the lesion and can be initiated by catheterization, bladder distension, and bowel evacuation. AD is accompanied by sweating, flushing, and a pounding headache and can lead to severe morbidity and even mortality. Clinically, AD has been well documented, but the mechanisms that mediate AD remain unclear. Because AD is induced by exaggerated sympathetic activity, it is thought to be α-adrenergic mediated. Therapy with an α-adrenoceptor blocker has, therefore, been a rational first choice in AD management. However, α-adrenoceptor blockers may leave a residual vasoconstrictor response to sympathetic neurotransmitters, such as adenosine triphosphate (ATP) and neuropeptide Y, unaffected. These neurotransmitters may still cause a vasoconstrictor response during α-adrenoceptor blockade, although their exact role during exaggerated sympathetic activity is unclear. Because SCI individuals lack supraspinal sympathetic control, their responses could differ from able-bodied individuals in the contribution of nonadrenergic transmission during exaggerated sympathetic reflexes. Nowadays, a calcium channel blocker (nifedipine) is most commonly used as a primary agent in the management of AD. A calcium channel blocker may be useful to prevent or control AD, indicated by a lower blood pressure response.
whether an
than a calcium channel blocker. To test this hypothesis, the
striction during AD and would, therefore, be more effective
the effect of phentolamine on leg vasoconstriction during
more pronounced in SCI individuals. To test this hypothesis,
vasoconstriction during AD. We hypothesized that an
was compared with control individuals by means of a cold
measurement), free of overt cardiovascular diseases, and did not
individuals participated in this study (Table 1). All of the subjects
were normotensive (<140/90 mm Hg; auscultatory blood pressure
measurement), free of overt cardiovascular diseases, and did not report orthostatic hypotension. Two SCI and 2 control individuals
smoked and 4 SCI individuals used medication, none of which are
known to substantially interfere with vascular reactivity (rectal
laxantia [n=2], furosemide, and tolterodine). All of the SCI individu-
als had long-standing traumatic spinal cord injury with a motor and sensory complete spinal cord lesion above T6 (American Spinal
Injury Association Impairment Scale A, zone of partial preservation
above T6)2.3 The level of spinal cord injury was assessed by clinical
examination. The study was carried out in accordance with the
Declaration of Helsinki and was approved by the medical ethical
commitee of our institution. All of the subjects gave written
informed consent.

Experimental Procedures and Protocol
All of the subjects refrained from caffeine-containing food and
beverages, vitamin C supplements, nicotine, and alcohol for >12
hours before the experiment and from heavy physical activity for
>24 hours before the experiment. Subjects had been fasting for >12
hours and had emptied their bladder in the hour before the experi-
ment. All of the experiments were performed in the morning in a
quiet, temperature-controlled room (23 ±1°C). Each subject was
studied on 2 different occasions, separated by 1 week. On the first
eperimental day, subjects were screened with a health question-
naire, physical examination, and a resting ECG. Subsequently, a
measurement of leg vascular resistance (LVR) in supine rest and
during exaggerated sympathetic activity, that is, AD in SCI and a
CPT of the hand in control individuals. On the second experimental
day, nicardipine, a calcium ion influx inhibitor (calcium channel
blocker), and phentolamine, a nonselective competitive antagonist
of α-adrenergic receptors, were successively infused into the right
femoral artery. LVR was measured during supine rest, as well as
during AD in SCI and during a CPT in control individuals.

On both days, subjects were positioned comfortably in a supine
position on a bed with an anti- ulcer mattress. Experimental proce-
dures on the first experimental day started after a supine resting
period of ≥30 minutes. First, baseline upper leg blood flow was
measured for 10 minutes in a supine position, and, subsequently, AD
was provoked in SCI individuals for 5 minutes. In control individ-
uals, a CPT of the hand was applied for 3 minutes, during which
upper leg blood flow was measured. On the second experimental
day, an intra-arterial cannula (Angiocath 16-gauge, Becton Dickin-
son Infusion Therapy Systems Inc) was introduced after local
anesthesia (0.4 mL of 10-mg/mL lidocaine hydrochloride, Fresenius
Kabi Nederland BV) using a modified Seldinger technique into the
right femoral artery at the level of the inguinal ligament for arterial
blood pressure measurement (HP monitor type 78353B, Hewlett
Packard GmbH) and intra-arterial drug administration by an auto-
matic syringe infusion pump (Type P2000, IVAC Medical Systems).
The measurements started after a supine resting period of ≥30
minutes after cannulation of the right femoral artery. First, baseline
upper leg blood flow was measured during a 5-minute saline (0.9%
NaCl, Baxter BV) infusion period, followed by nicardipine infusion
(1 mg/mL of Cardene, Astellas Pharma BV) for 10 minutes in a dose
of 0.5 μg/min per 100 mL of leg volume.13,14 In a pilot study, higher
doses of nicardipine did not further increase blood flow in a control
individual. In the last 5 minutes of nicardipine infusion, AD was
provoked in SCI and a CPT in control individuals. After a resting
period of ≥30 minutes with only saline infusion, the same protocol
was performed but this time with phentolamine. The schedule of the
protocol is shown in Figure 1.

AD in SCI individuals was provoked by inflating a blood pressure
cuff to 220 mm Hg on the contralateral upper leg, which gives a
nociceptive stimulus.2,3 According to the literature, AD was achieved
when there was a systolic blood pressure response to the stimulus of
≥20 mm Hg,2,17,18 or a 20% increase in blood pressure with visual-
ized vasoconstriction.2 On the first experimental day we attempted to
provoke AD with different stimuli (bladder percussion, CPT of the
foot, and inflating a blood pressure cuff) to see which stimulus would

Methods
Subjects
Six male SCI individuals and 7 healthy male able-bodied control
individuals participated in this study (Table 1). All of the subjects
were normotensive (<140/90 mm Hg; auscultatory blood pressure
measurement), free of overt cardiovascular diseases, and did not report orthostatic hypotension. Two SCI and 2 control individuals

Table 1. Subject Characteristics, Including the Specific Characteristics of the SCI and Control Individuals

<table>
<thead>
<tr>
<th>Subject</th>
<th>SCI Level</th>
<th>DOI, y</th>
<th>Age, y</th>
<th>Height, cm</th>
<th>Weight, kg</th>
<th>SBP, mm Hg</th>
<th>DBP, mm Hg</th>
</tr>
</thead>
<tbody>
<tr>
<td>SCI1</td>
<td>C5</td>
<td>17</td>
<td>46</td>
<td>181</td>
<td>105</td>
<td>130</td>
<td>78</td>
</tr>
<tr>
<td>SCI2</td>
<td>T5</td>
<td>28</td>
<td>46</td>
<td>198</td>
<td>65</td>
<td>108</td>
<td>78</td>
</tr>
<tr>
<td>SCI3</td>
<td>C7</td>
<td>45</td>
<td>69</td>
<td>179</td>
<td>81</td>
<td>108</td>
<td>60</td>
</tr>
<tr>
<td>SCI4</td>
<td>T5</td>
<td>10</td>
<td>39</td>
<td>178</td>
<td>70</td>
<td>106</td>
<td>62</td>
</tr>
<tr>
<td>SCI5</td>
<td>C7</td>
<td>34</td>
<td>52</td>
<td>182</td>
<td>92</td>
<td>108</td>
<td>62</td>
</tr>
<tr>
<td>SCI6</td>
<td>C7</td>
<td>12</td>
<td>36</td>
<td>183</td>
<td>58</td>
<td>110</td>
<td>50</td>
</tr>
<tr>
<td>SCI</td>
<td>n=6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C1</td>
<td></td>
<td></td>
<td></td>
<td>180</td>
<td>67</td>
<td>118</td>
<td>76</td>
</tr>
<tr>
<td>C2</td>
<td></td>
<td></td>
<td></td>
<td>194</td>
<td>110</td>
<td>124</td>
<td>90</td>
</tr>
<tr>
<td>C3</td>
<td></td>
<td></td>
<td></td>
<td>174</td>
<td>65</td>
<td>118</td>
<td>82</td>
</tr>
<tr>
<td>C4</td>
<td></td>
<td></td>
<td></td>
<td>188</td>
<td>73</td>
<td>128</td>
<td>64</td>
</tr>
<tr>
<td>C5</td>
<td></td>
<td></td>
<td></td>
<td>188</td>
<td>88</td>
<td>118</td>
<td>78</td>
</tr>
<tr>
<td>C6</td>
<td></td>
<td></td>
<td></td>
<td>180</td>
<td>76</td>
<td>136</td>
<td>70</td>
</tr>
<tr>
<td>C7</td>
<td></td>
<td></td>
<td></td>
<td>187</td>
<td>85</td>
<td>138</td>
<td>70</td>
</tr>
<tr>
<td>Control</td>
<td>n=7</td>
<td></td>
<td></td>
<td>184±2</td>
<td>81±5</td>
<td>126±3*</td>
<td>76±3</td>
</tr>
</tbody>
</table>

Values represent mean±SD. DOI indicates duration of injury; C in column 1 indicates control subjects; SBP, systolic blood pressure; DBP, diastolic blood pressure; C, cervical spinal segment; T, thoracic spinal segment.

*Data are significantly different from SCI.
result in the highest increase in blood pressure and fulfill the criteria.3,4,17,18 Bladder percussion could not be performed in all of the SCI individuals (sacral rhizotomy and electric bladder stimulator), and the frequency and pressure of the bladder percussion was difficult to standardize. A CPT of the foot did not increase mean arterial pressure (MAP) in 5 consecutive SCI individuals and was, therefore, considered an inappropriate stimulus. Inflating a blood pressure cuff to 220 mm Hg on the contralateral leg resulted in the highest increases in blood pressure, was consistent in provoking AD, appeared easy to standardize, and was applicable for each SCI individual. Inflating a blood pressure cuff to 220 mm Hg did not result in hemodynamic changes or in LVR of the contralateral leg in control individuals. We, therefore, decided to use this stimulus to provoke AD in SCI individuals.

To elevate sympathetic activity in control individuals, a CPT of the hand was applied.19 A CPT consisted of immersion of the right hand into ice-water (4°C) for a period of 3 minutes.19

**Measurements**

Bilateral upper leg blood flow was measured by ECG-triggered venous occlusion plethysmography, using mercury-in-silastic strain gauges (DE Hokanson, Bellevue, WA), and electrically calibrated.20 In the supine position, the legs were positioned =5 cm above heart level to facilitate venous outflow between venous occlusions.21 Strain gauges were placed 10 cm above the patella, and 12-cm width occlusion cuffs, placed on the thigh above the strain gauge, were inflated with a rapid cuff inflator (DE Hokanson), within 1 second, to 50 mm Hg.22 Occlusion pressures were sustained for 8 heart cycles, after which the cuff was deflated instantaneously (for 10 heart cycles).

Arterial blood pressure was measured continuously using a non-invasive blood pressure device (Portapres, TNO) on day 1. A finger cuff was attached to the middle phalanx of the left third finger to measure finger arterial blood pressure, which accurately reflects intraarterial blood pressure changes.23 On day 2, arterial blood pressure was continuously measured intra-arterially using the femoral artery cannula. MAP values were derived beat to beat, and heart rate was the inverse of the interbeat interval.

Leg volume was determined by anthropometry, as described and validated by Jones and Pearson.24 Venous (day 1) and arterial (day 2) blood samples were taken to determine norepinephrine levels in SCI and control individuals. In SCI individuals, renin and angiotensin II levels were determined as well to establish a possible role of the renin-angiotensin system in AD. The blood samples were taken at rest, before infusion of nicardipine and phentolamine, and directly after AD in SCI and after a CPT in control individuals (Figure 1). The samples were collected in prechilled glass tubes on melting ice containing glutathione and EDTA for determination of norepinephrine levels and nonchilled glass tubes for determination of renin and angiotensin II levels. Samples were processed immediately in a refrigerated centrifuge and stored at −80°C until further analysis. Plasma norepinephrine was measured by sensitive and specific high-performance liquid chromatography with fluorometric detection, as described previously.25 Plasma renin was measured by immunoradiometric assay provided by CISbio International, and angiotensin II levels in medium were measured by radioimmunoassay (detection limit: 0.5 pmol/L) as described previously.26

**Data Analysis**

A data acquisition system digitalized the data with a sample frequency of 100 Hz. (Medical Information Data Acquisition, Instrumentation Department, Radboud University Nijmegen Medical Centre). Upper leg blood flow was calculated as the slope of the volume change over a 4-second interval using a customized computer program (MATLAB 6.1, Mathworks). MAP and heart rate values over the same intervals were averaged.

LVR was calculated as the arterial-venous pressure gradient divided by upper leg blood flow. For these calculations, we assumed that central venous pressure was 9 mm Hg in a supine position.27 During AD and during a CPT, the average of the highest 3 consecutive measurements was taken to determine LVR.

**Statistical Analysis**

Statistical analyses were performed using SPSS 16.0 (SPSS) software. Data are presented as mean±SD unless otherwise stated. The level of statistical significance was set at α=0.05. To assess differences in baseline values between SCI and control individuals, unpaired t tests were used. Repeated-measures ANOVAs were used to assess the effect of AD within the SCI group and of a CPT within the control group on the first day and the effect of infusion of nicardipine or phentolamine on the second day. Post hoc t tests were performed when the ANOVA reported a significant main or interaction effect. Bonferroni correction was used to correct for multiple comparisons.

**Results**

**Baseline Values**

Supine resting systolic blood pressure and MAP in SCI were lower (P=0.02) and LVR was higher (P<0.01) compared with control individuals. Provoking AD in SCI and performing a CPT in control individuals on day 1 resulted in an...
increase in MAP (P<0.01 and P=0.01, respectively) and LVR (P<0.01 and P=0.01, respectively), whereas heart rate did not change in either group. The LVR increase during AD in SCI was similar to the increase during a CPT in control individuals (Tables 1 through 3 and Figure 2).

### Phenotolamine and Nicardipine Infusion During AD in SCI Individuals

LVR in SCI individuals was lower (P=0.02) during saline infusion on day 2 compared with the baseline value on day 1. Infusion of phenotolamine did not change MAP and heart rate but did lower LVR (P=0.04). Provoking AD during phentolamine infusion did not change MAP and heart rate; however, LVR increased (P=0.02). The increase in LVR was significantly lower (P=0.04) during phenolamine infusion (8.0±4.5 arbitrary units [AU]) compared with the LVR increase on day 1 (23.6±13.4 AU). Nicardipine infusion did not change MAP, heart rate, or LVR. Provoking AD during nicardipine infusion resulted in an increase in MAP (P=0.03) and LVR (P=0.04) with no change in heart rate. The increase

### Table 3. Individual Increase in MAP and LVR During AD in SCI Individuals and CPT in Control Individuals Without and With Phentolamine or Nicardipine Infusion

<table>
<thead>
<tr>
<th>Subject</th>
<th>MAP, mm Hg (%)</th>
<th>LVR, AU (%)</th>
<th>MAP, mm Hg (%)</th>
<th>LVR, AU (%)</th>
<th>MAP, mm Hg (%)</th>
<th>LVR, AU (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SCI</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SCI1</td>
<td>12 (13)</td>
<td>4 (13)</td>
<td>2 (2)</td>
<td>14 (87)</td>
<td>8 (8)</td>
<td>9 (57)</td>
</tr>
<tr>
<td>SCI2</td>
<td>15 (16)</td>
<td>43 (142)</td>
<td>4 (4)</td>
<td>3 (16)</td>
<td>9 (10)</td>
<td>9 (50)</td>
</tr>
<tr>
<td>SCI3</td>
<td>33 (44)</td>
<td>20 (54)</td>
<td>8 (12)</td>
<td>9 (30)</td>
<td>17 (25)</td>
<td>21 (72)</td>
</tr>
<tr>
<td>SCI4</td>
<td>6 (8)</td>
<td>19 (67)</td>
<td>1 (2)</td>
<td>10 (64)</td>
<td>4 (5)</td>
<td>24 (101)</td>
</tr>
<tr>
<td>SCI5</td>
<td>30 (40)</td>
<td>33 (95)</td>
<td>36 (58)</td>
<td>11 (58)</td>
<td>27 (39)</td>
<td>25 (98)</td>
</tr>
<tr>
<td>SCI6</td>
<td>29 (36)</td>
<td>22 (67)</td>
<td>3 (4)</td>
<td>3 (14)</td>
<td>8 (11)</td>
<td>2 (12)</td>
</tr>
<tr>
<td>SCI (n=6)</td>
<td>21±11 (26±15)</td>
<td>24±13 (73±40)</td>
<td>9±13* (14±22)</td>
<td>8±5* (45±27)</td>
<td>12±8* (16±13)</td>
<td>15±9* (65±30)</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C1</td>
<td>4 (5)</td>
<td>7 (51)</td>
<td>8 (10)</td>
<td>1 (9)</td>
<td>13 (16)</td>
<td>11 (58)</td>
</tr>
<tr>
<td>C2</td>
<td>14 (15)</td>
<td>8 (54)</td>
<td>−15 (−14)</td>
<td>−3 (−23)</td>
<td>13 (12)</td>
<td>5 (27)</td>
</tr>
<tr>
<td>C3</td>
<td>9 (9)</td>
<td>12 (41)</td>
<td>6 (7)</td>
<td>3 (26)</td>
<td>3 (4)</td>
<td>1 (13)</td>
</tr>
<tr>
<td>C4</td>
<td>6 (6)</td>
<td>14 (54)</td>
<td>10 (13)</td>
<td>2 (19)</td>
<td>−2 (−3)</td>
<td>1 (8)</td>
</tr>
<tr>
<td>C5</td>
<td>15 (15)</td>
<td>17 (58)</td>
<td>6 (7)</td>
<td>0 (3)</td>
<td>6 (7)</td>
<td>1 (10)</td>
</tr>
<tr>
<td>C6</td>
<td>34 (36)</td>
<td>17 (475)</td>
<td>14 (19)</td>
<td>0 (3)</td>
<td>16 (21)</td>
<td>10 (82)</td>
</tr>
<tr>
<td>C7</td>
<td>31 (32)</td>
<td>21 (110)</td>
<td>7 (9)</td>
<td>0 (4)</td>
<td>13 (16)</td>
<td>1 (14)</td>
</tr>
<tr>
<td>Control (n=7)</td>
<td>16±12 (17±12)</td>
<td>18±13 (78±45)</td>
<td>5±9* (7±10)</td>
<td>1±2* (6±14)</td>
<td>9±6* (10±8)</td>
<td>4±4* (30±26)</td>
</tr>
</tbody>
</table>

Values represent mean±SD. Δ indicates increase; %, percentage increase; C, control subject.
*Data are significantly different from day 1.
†Data are significantly different from phenolamine infusion.
Phentolamine and Nicardipine During CPT in Control Individuals

Phentolamine infusion in control individuals did not change MAP but did increase heart rate ($P=0.03$) during nicardipine infusion (11.9±8.4 mm Hg) compared with the MAP increase on day 1 (21.3±10.9 mm Hg). Nicardipine did not significantly attenuate the LVR increase during AD ($P=0.12$), whereas the LVR increase provoked by AD was significantly lower ($P<0.05$) during phentolamine (8.0±4.5 AU) compared with nicardipine infusion (15.1±9.9 AU). In the noninfused leg, no change in LVR was seen during infusion of phenolamine (31.3±10.3 versus 30.0±10.0 AU) or during nicardipine (33.0±15.8 versus 33.8±16.8 AU; Tables 2 and 3 and Figure 2).

Blood Samples

Baseline norepinephrine levels were significantly lower ($P<0.01$) in SCI compared with control individuals. During AD, norepinephrine did not significantly increase in SCI in contrast to control individuals during a CPT ($P=0.01$). Norepinephrine increased significantly during AD, in combination with phenolamine and nicardipine infusion in SCI ($P=0.04$ and $P=0.03$, respectively) and during a CPT in control individuals ($P<0.01$, both conditions). The increase in norepinephrine during phenolamine infusion was significantly more pronounced ($P=0.04$) in control than in SCI individuals. There were no significant increases in renin and angiotensin II levels during AD in SCI individuals without or with phenolamine or nicardipine infusion (Table 4).

Discussion

The major finding of this study is that AD in SCI individuals is not entirely mediated through the $\alpha$-adrenergic pathway, indicated by the presence of a remaining residual leg vasoconstrictor response during AD in SCI individuals during phenolamine infusion. Nevertheless, a more pronounced attenuation of the LVR increase during AD was present during phenolamine infusion compared with a nicardipine infusion. In contrast to SCI individuals, phenolamine infusion completely abolished the vasoconstriction response to a CPT in control individuals, which could indicate that the presence of supraspinal sympathetic control in control individuals may suppress the role of nonadrenergic transmission during exaggerated sympathetic activity.
Contribution of Nonadrenergic Transmission

The LVR increase during AD in SCI individuals did not differ from the LVR increase during a CPT in control individuals. Nevertheless, phentolamine completely abolished the LVR increase in control individuals during a CPT, whereas a significant residual leg vasoconstrictor response occurred in SCI individuals during AD with phentolamine infusion. These results suggest that nonadrenergic transmission contributes to the leg vasoconstriction during exaggerated sympathetic activity in SCI but not in control individuals. This interpretation is supported by the more pronounced increase in plasma norepinephrine levels in control individuals compared with SCI individuals in response to exaggerated sympathetic activity. This observation challenges the widespread view that the vasoconstrictor response during AD in SCI individuals is entirely \(\alpha\)-adrenergic mediated.2–4.28

Because phentolamine is a nonselective competitive \(\alpha\)-adrenoceptor antagonist, incomplete \(\alpha\)-adrenoceptor blockade could, in theory, explain the residual LVR increase in SCI individuals. A similar intra-arterial dose of phentolamine has been used previously and achieved a maximal vasodilator effect in both SCI and control individuals, indicating complete \(\alpha\)-adrenoceptor blockade.15.29,30 Moreover, the LVR increase during a CPT in control individuals was completely abolished during phentolamine infusion, despite a larger increase in plasma norepinephrine compared with SCI individuals. This confirms that intra-arterial infusion of phentolamine achieved effective intrasynaptic drug concentrations30 and excludes incomplete \(\alpha\)-adrenergic blockade as an explanation for the residual vasoconstrictor response in SCI individuals in the present study.

Because vasoconstriction during AD occurs rapidly, it is not likely that other vasoconstrictor mechanisms play an important role, such as the renin-angiotensin system. The renin-angiotensin system is a slow-acting vasoconstriction mechanism and, therefore, unlikely to cause immediate arterial vasoconstriction on a visceral, noxious, or nociceptive stimulus. Moreover, there were no increases in renin and angiotensin II levels during AD in SCI individuals, supporting the notion that there was no activation of the renin-angiotensin system. However, the local angiotensin system could still play a role, because we did not have an additional angiotensin II subtype I receptor blockade. It is unlikely that vasopressin plays a role, because vasopressin levels are low in SCI individuals and do not increase during AD.31 The instant reaction to the triggering stimulus provoking AD in combination with the lack of supraspinal control in SCI individuals causing exaggerated sympathetic activity provides enough evidence for a sympathetic-mediated mechanism. Because \(\alpha\)-adrenoceptor blockade did not abolish the LVR increase during AD, other sympathetic neurotransmitters might be involved, such as ATP and neuropeptide Y.10 ATP, neuropeptide Y, and norepinephrine are costored in the synaptic cleft and are simultaneously released.32 Receptors for these neurotransmitters are located on smooth muscle and endothelial cells of blood vessels.32 It is thought that norepinephrine and ATP have a coordinated action in neurogenic vasoconstriction, which is modulated by neuropeptide Y.32 We did not investigate the mechanism by which spinal cord injury increases sympathetic nonadrenergic transmission. Because the sympathetic nervous system below the lesion in SCI individuals is without central inhibitory pathways,2,3 in contrast to control individuals, we speculate that an intact supraspinal control of sympathetic outflow preferentially suppresses sympathetic nonadrenergic transmission. Alternatively, spinal cord injury alters the relative concentrations of costored neurotransmitters. In this regard, an intermediate role for endothelin is worth mentioning. We have shown previously that the contribution of endothelin in LVR is increased in SCI individuals.33 In rats, endothelin 1 infusion increased the relative contribution of ATP as a functional sympathetic neurotransmitter.34 Therefore, endothelin may mediate the increased contribution of nonadrenergic neurogenic vasoconstriction in SCI individuals.

Phentolamine Effect Superior to Nicardipine

Provoking AD in SCI individuals during nicardipine infusion still resulted in an LVR increase with a concurrent increase in MAP. However, the MAP increase with nicardipine infusion was lower compared with day 1, indicating an attenuation of the blood pressure response during AD in SCI individuals. This is consistent with 1 previous study demonstrating a lower blood pressure response during oral nifedipine pretreatment in SCI individuals who exhibited AD during electroejaculation.11 However, the LVR increase was similar to day 1, indicating a minor effect of nicardipine on leg vasoconstriction.

Despite the residual LVR increase during AD in SCI individuals with phentolamine infusion, this LVR increase was significantly lower than with nicardipine infusion, and, moreover, with phentolamine there was no change in MAP. These results indicate a superior effect of an \(\alpha\)-adrenoceptor antagonist compared with a calcium channel blocker on leg vasoconstriction and concurrent blood pressure response during AD in SCI individuals.

An obvious explanation for the observed difference in effect could be an inefficient dosage of nicardipine. Infusion of nicardipine did not result in a significant decrease in basal LVR in SCI individuals. However, in control individuals, basal LVR decreased with the same dose, and, in a pilot study, higher dosages of nicardipine did not result in a more pronounced vasodilatory effect. We are, therefore, convinced that we used a sufficient dose of nicardipine. The vasodilatation caused by nicardipine is, however, more pronounced in hypertensive compared with normotensive individuals.16 Because blood pressure in SCI individuals was lower compared with control individuals, the effect of nicardipine on LVR could be smaller for this reason.

Baseline Values

Consistent with our results, SCI individuals with a high spinal cord lesion are prone to low resting blood pressures,1,8–3 which is thought to be attributed to a diminution in sympathetic nervous activity below the lesion as supported by low plasma norepinephrine levels.1,2 Although the reduced norepinephrine levels in SCI individuals may also be influenced by the efficiency of the reuptake by the norepinephrine transporter and changes in blood flow redistribution caused by inactivity and muscle atrophy. Despite lower sympathetic activity, SCI
individuals have a higher resting LVR, probably because of a combination of functional and structural vascular changes. During AD, SCI individuals can increase their LVR to the same extent as control individuals during a CPT. Although a CPT is not directly comparable with AD, both are strong sympathetic stimuli, notwithstanding the physiological and neurological differences, especially the absence (SCI) or presence (control) of supraspinal control.

Limitations
Although α-adrenergic responsiveness deteriorates with age, all of the SCI and control individuals demonstrated similar increases in LVR during AD and a CPT, respectively, compared with their peers. Moreover, the oldest control individual (C7, 54 years) had a complete abolished LVR increase during a CPT with phentolamine infusion, and the oldest SCI individual (SC13, 69 years) had an attenuated LVR increase during AD with phentolamine infusion.

Infusion of saline in SCI individuals lowered supine LVR compared with day 1. This is probably because of a more pronounced reaction to an increase in shear on the vascular wall, caused by the saline infusion, because of functional changes in SCI individuals. The noninfused leg did not demonstrate a lower supine LVR, indicating that the lower supine LVR in the infused leg is probably caused by the saline infusion. The lower supine LVR did not have any effect on MAP and heart rate. Because the focus of the study was the effect of phentolamine and nicardipine infusion on the LVR increase during AD and a CPT, we may assume that the lower basal LVR did not influence our results.

Perspectives
The management of AD in SCI individuals remains a challenge in clinical practice. Over the years, many different antihypertensive agents have been used. In earlier days, α-adrenoceptor antagonists were used; however, their use was limited, and nowadays nifedipine (calcium channel blocker) is used as a primary agent in the management of AD. However, the present study demonstrates that an α-adrenoceptor antagonist appears to have a more pronounced effect than a calcium channel blocker on the LVR and concurrent MAP increase during AD in SCI individuals. Moreover, serious adverse reactions after the use of immediate-release nifedipine in hypertensive emergencies in non-SCI individuals have been reported. The present study demonstrates that AD in SCI individuals is not entirely α-adrenergic mediated and that sympathetic nondrenergic transmission may partly explain the LVR increase. The development of antagonists of nondrenergic transmitters may be a target for future AD management in SCI individuals. Perhaps these antagonists may reduce the blood pressure increase during AD without lowering the blood pressure in the absence of AD.

Acknowledgments
We acknowledge the enthusiastic participation of all of the subjects in this study. In addition, we thank Jos Evers, Bregina Kersten, Suzanne Arends, and Ria Wolkorte for assistance during the experiments.

Sources of Funding
J.T.G. is financially supported by the Netherlands Organisation for Scientific Research (ZonMW AGIKO-stipend).

Disclosures
None.

References
22. Groothuis JT, van Vliet L, Kooijman M, Hopman MTE. Venous cuff pressures from 30 mmHg to diastolic pressure are recommended to measure arterial inflow by plethysmography. J Appl Physiol. 2003;95:342–347.

Downloaded from http://hyper.ahajournals.org/ at Radboud Universiteit Nijmegen on July 13, 2012


