Endothelium-dependent and -independent vasodilation of the superficial femoral artery in spinal cord-injured subjects

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1Department of Physiology, Institute of Fundamental and Clinical Movement Sciences, and 2Research Institute for Sport and Exercise Science, Liverpool John Moores University, Liverpool, United Kingdom; 3Rehabilitation Centre St. Maartenskliniek, and 4Department of Pharmacology-Toxicology, Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands; and 5School of Sports Science, Exercise and Health, The University of Western Australia, Crawley, Western Australia, Australia

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Thijsse DH, Kooijman M, de Groot PC, Bleeker MW, Smits P, Green DJ, Hopman MT. Endothelium-dependent and -independent vasodilation of the superficial femoral artery in spinal cord-injured subjects. J Appl Physiol 104: 1387–1393, 2008. First published February 28, 2008; doi:10.1152/japplphysiol.01039.2007.—Extreme inactivity of the legs in spinal cord-injured (SCI) individuals does not result in an impairment of the superficial femoral artery flow-mediated dilation (FMD). To gain insight into the underlying mechanism, the present study examined nitric oxide (NO) responsiveness of vascular smooth muscles in controls and SCI subjects. In eight healthy men (34 ± 13 yr) and six SCI subjects (37 ± 10 yr), superficial femoral artery FMD response was assessed by echo Doppler. Subsequently, infusion of incremental dosages of sodium nitroprusside (SNP) was used to assess NO responsiveness. Peak diameter was examined on a second day after 13 min of arterial occlusion in combination with sublingual administration of nitroglycerine. Resting and peak superficial femoral artery diameter in SCI subjects were smaller than in controls (P < 0.001). The FMD response in controls (4.2 ± 0.9%) was lower than in SCI subjects (8.2 ± 0.9%, P < 0.001), but not after correcting for area under the curve for shear rate (P = 0.35). When expressed as relative change from baseline, SCI subjects demonstrate a significantly larger diameter increase compared with controls at each dose of SNP. However, when expressed as a relative increase within the range of diameter changes [baseline (0%) – peak diameter (100%)], both groups demonstrate similar changes in response to SNP. Changes in diameter during SNP infusion and FMD response are larger in SCI subjects compared with controls. When these results are corrected, superficial femoral artery FMD and NO sensitivity in SCI subjects are different from those in controls. This illustrates the importance of appropriate data presentation and suggests that, subsequent to structural inward remodeling of conduit arteries as a consequence of extreme physical inactivity, arterial function is normalized.

flow-mediated dilation; spinal cord injury; deconditioning; nitric oxide; nitric oxide sensitivity

PHYSICAL INACTIVITY IS STRONGLY associated with a reduced cardiovascular mortality in middle age and older age (34). As a result of deconditioning, detrimental cardiovascular adaptations have been documented, which may explain the increased cardiovascular risk. For example, short-term physical inactivity associated with bed rest (3) or unilateral lower limb suspension (2) and long-term physical inactivity in the extremely inactive legs of spinal cord-injured (SCI) individuals (10, 11, 17) decrease conduit artery diameter and increase peripheral vascular resistance. Interestingly, vascular changes in the legs of SCI individuals are complete within 3 wk of initiation of the spinal cord lesion (11).

The above findings stand in contrast to observations regarding conduit artery function in deconditioned patients. Conduit artery endothelial function, assessed using flow-mediated dilation (FMD), which is largely nitric oxide (NO) dependent (20, 22), is a surrogate marker of cardiovascular disease (36) and is impaired in patients with cardiovascular risk factors (7, 9, 28). Since physical inactivity is associated with increased cardiovascular risk (34), one might logically expect to observe attenuated FMD in models of deconditioning. However, previous reports have demonstrated increased (2, 3, 5) or preserved (12) FMD in the superficial femoral artery (SFA) of deconditioned subjects. Hindlimb unweighting in rats indicates that deconditioning can lead to a downregulation of endothelial NO synthase expression (37). Accordingly, one explanation for the paradoxical FMD outcomes during physical inactivity relates to the possibility that smooth muscle cell sensitivity to NO is enhanced in the setting of deconditioning.

The aim of this paper was, therefore, to assess the role of smooth muscle responsiveness to a NO donor in the legs of SCI individuals. To this end, increasing doses of sodium nitroprusside (SNP) were infused into the SFA, and changes in arterial diameter were recorded. We hypothesized that an increase in NO sensitivity of the vascular smooth muscle cells in SCI patients may contribute to the preserved or enhanced FMD responses as a consequence of deconditioning.

METHODS

Subjects

Eight healthy male controls and six male SCI individuals participated in this study. Baseline characteristics are shown in Table 1. Subjects with a history of cardiovascular diseases, diabetes, insulin resistance, hypercholesterolemia, or hypertension were excluded from the study. Control subjects were nonsmokers and used no medication. Two SCI subjects continued their medication, which did not interfere with our study purpose (methylphenidate and imipramine by one subject, whereas another subject used naproxen). Two SCI individuals smoked, but they refrained from smoking 3 days before testing. The

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SCI individuals had complete motor and sensor spinal cord lesions of traumatic origin varying from thoracic 1 to 12 (American Spinal Injury Association). The level of the spinal lesion was assessed by clinical examination. The Medical Ethics Committee of the Radboud University Nijmegen has approved the study, and all subjects gave their written, informed consent before the study.

### Protocol

Measurements were carried out in the morning after an overnight fast. Subjects were asked to empty their bladder before examination and refrained from alcohol, caffeine, vitamin C, and exercise at least 18 h before the test. Room temperature was controlled at 23–24°C.

**Day 1: Peak arterial dilatation to nitroglycerine + ischemia.** After a 30-min acclimatization period in the supine position, measurements of vessel diameter and red blood cell velocity of the SFA were performed using an echo Doppler device (Megas Esaote, Firenze, Italy). To obtain the absolute peak change in diameter of the SFA, a 13-min suprasystolic arterial occlusion (220 mmHg) was applied by a cuff placed proximally around the right thigh (but placed below the site of vascular measurement). Diameter and blood flow velocity were measured with an echo Doppler device (Megas Esaote, Firenze, Italy). To obtain the absolute peak change in diameter of the SFA, a difference in vasodilation of the SFA. Diameter and blood flow velocity were measured with a 5- to 7.5-MHz broadband linear array performs an echo Doppler device with a 5- to 7.5-MHz broadband linear array (Megas Esaote, Firenze, Italy). To obtain the absolute peak change in diameter of the SFA, a difference in vasodilation of the SFA. Diameter and blood flow velocity were measured with an echo Doppler device (Megas Esaote, Firenze, Italy).

Blood velocities and vessel diameter of SFA were measured with an echo Doppler device with a 5- to 7.5-MHz broadband linear array.

| Table 1. Subject characteristic and baseline vascular values in controls and SCI subjects |
|-----------------|--------|--------|
| n               | Controls | SCI |
| Age, yr         | 34 ± 12 | 37 ± 10 |
| Height, cm      | 185 ± 7  | 179 ± 3  |
| Weight, kg      | 82 ± 16  | 72 ± 6   |
| Systolic blood pressure, mmHg | 128 ± 9 | 121 ± 13 |
| Diastolic blood pressure, mmHg | 76 ± 4 | 78 ± 9 |
| Diameter, cm    | 0.78 ± 0.05 | 0.55 ± 0.04* |
| Resting blood flow, ml/min | 75 ± 34 | 98 ± 43 |
| Resting vascular conductance, AU | 0.79 ± 0.43 | 0.99 ± 0.48 |
| Mean wall shear rate, s⁻¹ | 13 ± 4 | 50 ± 29* |
| Peak diameter, cm | 0.85 ± 0.06 | 0.67 ± 0.04* |
| Diameter dilator range, cm | 0.08 ± 0.02 | 0.11 ± 0.02* |

Values are means ± SD; n, no. of subjects; SCI, spinal cord injured; AU, arbitrary unit. Peak diameter is examined after 13-min occlusion and spray of a nitric oxide donor (day 1). *P < 0.01 (unpaired Student’s t-test).

5-min periods (SNP, 0.075, 0.15, 0.3 μg·min⁻¹·dl⁻¹ of leg volume) to examine the NO sensitivity of the femoral artery. A difference in sensitivity of the smooth muscles was reported when the dilatation to a predetermined dosage of SNP differs between groups (larger dilator response corresponds with increased NO sensitivity). Anthropometric determination was used to calculate leg volume (19). The NO donor SNP directly affects the vascular smooth muscle cell, resulting in a vasodilatation of the SFA. Diameter and blood flow velocity were examined during the last 2 min of each infusion. In a previous study, the diameter of the radial artery reached a maximum at a dose of 0.3 μg·min⁻¹·dl⁻¹ (18). Systemic effects are not to be expected using these doses in the leg (4, 24). During the last minute of each infusion dosage of SNP, SFA diameter and flow velocity profiles were recorded using echo Doppler.

### Measurements and Analyses

Blood velocities and vessel diameter of SFA were measured with an echo Doppler device with a 5- to 7.5-MHz broadband linear array.

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transducer. The sample volume was placed in the center of the artery, and images were made ~3 cm distal of the bifurcation into the deep femoral artery and the SFA. The angle of insonation for the blood velocity measurements was consistently below 60°. During measurement of the diameter, the vessel area was adjusted parallel to the transducer. For baseline diameter measurements, two consecutive images in the longitudinal view were frozen at the peak systolic (Dₚ) and end-diastolic phase (Dₑ). Offline, three measurements were performed per diameter image, and the mean diameter (D) was calculated by using the formula: $\frac{1}{3} \cdot Dₚ + \frac{2}{3} \cdot Dₑ$. For baseline velocity measurements, four images with a total of 10–12 velocity profiles were obtained and manually traced afterwards by a single investigator. The average of these 10–12 Doppler spectra waveforms was used to calculate peak velocity ($V_{\text{peak}}$) and mean velocity ($V_{\text{mean}}$).

Hyperemic velocity was recorded on videotape for the first 45 s after cuff release. After 45 s, SFA diameter was recorded continuously until 8 min for determination of the peak diameter (day 1) and until 5 min after cuff release for assessment of the FMD (day 2). Reactive peak and mean hyperemic blood velocity was calculated from the flow velocity integral every 5 s, from 15 to 45 s after cuff release, which was manually traced by a single investigator. For resting and hyperemic responses, mean blood flow in milliliters per minute was calculated as $\pi \cdot (\text{radius})^2 \cdot V_{\text{mean}} \cdot (\text{cm/s}) \cdot 60$, mean wall shear rate (MWSR) was calculated as $4 \cdot V_{\text{mean}} / D$ (s⁻¹), and regional peak wall shear rate (PWSR) was calculated as $4 \cdot V_{\text{peak}} / D$ (s⁻¹).

Vessel diameters of the SFA after reactive hyperemia were measured offline from videotape at 50, 60, 90, and 120 s, and thereafter every 30 s until 8 min after cuff release to determine peak SFA diameter (day 1) and at 50, 60, 90, 120, 240, and 300 s after cuff release to measure FMD (day 2). All diameters were measured at the end-diastolic phase of the cardiac cycle, corresponding to the R-wave of a simultaneous ECG signal. FMD in the SFA and peak dilation of the SFA were expressed as both the maximal absolute and relative diameter change in end-diastolic baseline diameter. The ratio between the relative FMD response (%FMD) and the primary stimulus for vessel dilation (MWSR) was calculated. Regional MWSR was calculated as $(4 \cdot V_{\text{mean}} / D)$ (s⁻¹). MWSR area under the curve (AUC) was calculated from 15 to 40 s after cuff release to correct for the shear stress stimulus that elicits conduit artery dilation. NO sensitivity was calculated as the absolute and relative diameter increase of the SFA from baseline at each SNP dose. In addition, the increase was also calculated as the relative change within the diameter dilator range (baseline is 0% and peak diameter is 100%). Peak diameter was identified as the largest diameter change for each individual, whether this occurred in response to local high-dose SNP infusion or NTG combined with 13 min of ischemia. Reproducibility of the measurements in the SFA in our laboratory are reported to be 1.5% for baseline diameter, 14% for blood flow, and 15% for FMD (12).

### Statistical Analysis

Statistical analyses were performed using SPSS 12.0 computer software (SPSS, Chicago, IL). Kolmogorov-Smirnov test indicated a normal (Gaussian) distribution of data. Results are expressed as means ± SD, unless stated otherwise. Differences in blood flow and diameter during the NO donor infusion were examined using a two-way, repeated-measures ANOVA. To assess differences between groups, a Student’s t-test for unpaired groups was used. To examine differences between groups, an unpaired Student’s t-test was used. Differences were considered to be statistically significant at a two-sided probability value of $\leq 0.05$.

### RESULTS

**Baseline Vascular Characteristics and FMD**

Resting and peak (after ischemia and NTG) SFA diameter were significantly lower in SCI subjects compared with controls, whereas MWSR was increased in SCI subjects (Table 1). The diameter dilator range (difference between maximal and resting diameter) was significantly larger in SCI subjects than

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**Table 2. Superficial femoral artery diameter responses to increasing dosages of SNP and during the combination of 13-min ischemia + nitroglycerine (peak) in controls and SCI subjects**

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>SNP1</th>
<th>SNP2</th>
<th>SNP3</th>
<th>Peak</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Controls</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absolute diameter, mm</td>
<td>7.8±0.2</td>
<td>8.3±0.2</td>
<td>8.5±0.2</td>
<td>8.5±0.2</td>
<td>8.6±0.2</td>
</tr>
<tr>
<td>Absolute change, mm</td>
<td>0.55±0.06</td>
<td>0.69±0.04</td>
<td>0.75±0.05</td>
<td>0.79±0.06</td>
<td></td>
</tr>
<tr>
<td>Relative change, %</td>
<td>7.1±0.8</td>
<td>8.8±0.5</td>
<td>9.7±0.7</td>
<td>9.3±1.1</td>
<td></td>
</tr>
<tr>
<td>Diameter dilator range, %</td>
<td>0</td>
<td>69±8</td>
<td>88±3</td>
<td>96±2</td>
<td>89±7</td>
</tr>
<tr>
<td><strong>SCI subjects</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absolute diameter, mm</td>
<td>5.6±0.2</td>
<td>6.2±0.2</td>
<td>6.6±0.2</td>
<td>6.6±0.2</td>
<td>6.7±0.2</td>
</tr>
<tr>
<td>Absolute change, mm</td>
<td>0.62±0.06</td>
<td>1.02±0.08</td>
<td>1.06±0.09</td>
<td>1.13±0.08</td>
<td></td>
</tr>
<tr>
<td>Relative change, %</td>
<td>11.3±1.2</td>
<td>18.4±1.5</td>
<td>19.2±1.7</td>
<td>20.3±1.5</td>
<td></td>
</tr>
<tr>
<td>Diameter dilator range, %</td>
<td>0</td>
<td>57±6</td>
<td>90±2</td>
<td>94±3</td>
<td>100±1</td>
</tr>
</tbody>
</table>

Values are means ± SD; $n = 8$ controls and $n = 6$ SCI subjects. SNP1, SNP2, SNP3: sodium nitroprusside doses 1, 2, and 3, respectively. Responses are presented as absolute diameter, absolute and relative change from baseline, and relative change within the diameter dilator range. In controls as well as in SCI subjects, each parameter demonstrated a significant effect of the increasing SNP dose using a one-way ANOVA ($P < 0.01$).
of the MWSR after cuff deflation was significantly larger in SCI subjects (1.110 ± 214) than in controls (823 ± 72, P = 0.004). After correction for the AUC of the MWSR, no difference was observed between SCI subjects and controls (Figs. 2B and 3).

**Diameter Response to SNP**

In controls as well as in SCI subjects, increasing dose of SNP resulted in a significant increase of the diameter of the SFA (Table 2, Fig. 4A). Post hoc analysis revealed significant differences in SFA diameter between all SNP doses in controls. Apart from a nonsignificant difference between SNP doses 2 and 3 (P = 0.08, while all subjects showed a larger diameter at SNP dose 3), also SCI subjects show significant differences between doses. In both groups, the diameter change during SNP dose 3 did not differ from the response during 13 min + NTG. The absolute and relative increase (from baseline) in SFA diameter during SNP infusion in SCI subjects was significantly larger than in controls (Fig. 4, B and C, P < 0.01). However, when normalized for the diameter dilator range (baseline as 0% and maximal diameter as 100%), a similar increase in SFA diameter is observed at each SNP dose between controls and SCI subjects (Fig. 4D).

When the FMD response is expressed relative to the maximal SNP response, no differences are found between controls and SCI subjects (0.45 ± 0.14 and 0.41 ± 0.09, respectively; P = 0.54).

Only in controls at the highest SNP dose was mean arterial pressure significantly decreased (Table 3). Heart rate was increased in SCI subjects during infusion of SNP at the dosages 0.15 and 0.3 μg·min⁻¹·dl⁻¹, while controls showed an increase in heart rate at SNP infusion of 0.3 μg·min⁻¹·dl⁻¹. In controls as well as in SCI subjects, blood flow increased during incremental dosages of SNP (Table 4). A two-way, repeated-measures ANOVA demonstrated no group effect for the change in blood flow (P = 0.75). Shear rate did not change significantly during the SNP infusion (repeated-measures ANOVA, P = 0.34), while no group effect was evident (P = 0.55).

**DISCUSSION**

In the present study, we demonstrated an inward remodeling of the SFA in SCI individuals by examining both resting and peak conduit vasodilator responses. The smaller diameter in SCI subjects seems to have important consequences for examining vascular function. First, the inward remodeling leads to an increased shear rate at baseline and after ischemia. While a

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**Table 3. Mean arterial pressure and heart rate at baseline and during the SNP infusion protocol in controls and SCI subjects**

<table>
<thead>
<tr>
<th>Heart rate, beats/min</th>
<th>Baseline</th>
<th>SNP1 (0.075 μg·min⁻¹·dl⁻¹)</th>
<th>SNP2 (0.15 μg·min⁻¹·dl⁻¹)</th>
<th>SNP3 (0.3 μg·min⁻¹·dl⁻¹)</th>
<th>ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls</td>
<td>55±7</td>
<td>57±6</td>
<td>57±6</td>
<td>63±6*</td>
<td>0.004</td>
</tr>
<tr>
<td>SCI</td>
<td>57±8</td>
<td>59±9</td>
<td>60±8*</td>
<td>64±6*</td>
<td>0.005</td>
</tr>
<tr>
<td>Mean arterial pressure, mmHg</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controls</td>
<td>93±7</td>
<td>94±8</td>
<td>90±7</td>
<td>87±5*</td>
<td>0.001</td>
</tr>
<tr>
<td>SCI</td>
<td>101±7</td>
<td>99±8</td>
<td>97±8</td>
<td>95±9</td>
<td>NS</td>
</tr>
</tbody>
</table>

Values are means ± SD; n = 8 controls and n = 6 SCI subjects. NS, not significant. A repeated-measured ANOVA was used to examine differences between the separate infusions. *Post hoc P < 0.05 baseline compared with SNP infusion.
larger FMD response is present in SCI subjects than in controls, this difference is abolished after correction for the eliciting stimulus. In addition, to examine smooth muscle cell sensitivity for NO, we constructed a dose-response curve to a NO donor. After correction for the differences in diameter between controls and SCI subjects, we found a comparable NO sensitivity of the smooth muscle cells, rather than enhanced responses to this stimulus. Thus long-term physical inactivity of the legs of SCI individuals is associated with structural changes in the conduit artery, while function of the artery is normal.

An ~30% smaller baseline SFA diameter was reported in the paralyzed legs of SCI individuals compared with their able-bodied peers. The magnitude of this difference is equivalent to that reported in previous studies (10, 12, 25). Based on the smaller SFA diameter in SCI subjects, one may suggest the presence of an inward remodeling process in the extremely inactive legs of SCI subjects. However, peak artery diameter has recently been demonstrated to provide a better index of structural artery remodeling than baseline diameter, because the latter is impacted by sympathetic tone and other competitive vasoactive influences (23). In the present study, the largest SNP dose and ischemia combined with sublingual NTG both result in a marked increase in diameter (23). Comparing these peak diameters, SCI subjects still demonstrate a smaller arterial diameter than their able-bodied peers. This clearly indicates the presence of an inward remodeling in the SFA of SCI subjects.

In keeping with recent reports relating to prolonged [SCI (12)] and short-term deconditioning [unilateral lower limb suspension (2), bed rest (3, 5)], the uncorrected FMD data were enhanced in the deconditioned limb. While this approach is still widely applied to calculate the FMD, studies in the brachial artery recommend correction for the eliciting stimulus (21, 26). Using the postocclusive AUC of the MWSR, the results of the present study indicate that FMD is similar in controls and SCI subjects. This finding parallels a previous study during inactivity (12) that used the PWSR to correct for the eliciting stimulus. As a result of the inward remodeling, a larger shear stress is present in the SFA of SCI subjects at rest and after cuff release. After correction for postocclusive deflation shear stress stimulus, similar FMD responses between SCI individuals and controls are observed. This indicates that vascular function is preserved in conduit arteries supplying deconditioned limbs, notwithstanding the fact that the SFA arteries exhibited significant inward remodeling.

Our finding, suggesting that FMD is preserved in SCI subjects, is, therefore, consistent with most of the previous studies in the literature. However, a recent study that focused on the posterior tibial artery reported impaired FMD data in SCI subjects compared with controls (29). However, the FMD data in this study were not corrected for the eliciting shear rate stimulus, and vessel imaging was performed distal from the occlusion cuff in a relatively small artery, which may account for the different findings compared with other studies in the literature. Previous studies in the upper limb suggest that distal cuff occlusion induces FMD, which is largely NO and endothelium dependent (13), whereas dilation of arteries that were within the ischemic zone is less NO dependent (13). Furthermore, our laboratory recently proved that distal cuff placement relative to the imaging site in the SFA causes largely NO-mediated dilation in this artery (20). No such evidence exists, so far, for dilation of smaller arteries in the lower limb, which may not produce as much NO. Hence, methodological differences between previous studies (Refs. 1–4 vs. 16) of lower limb FMD responses in physical inactivity, and in particular the fact that different arteries were examined, could explain the different outcomes.

The traditional sublingual single-dose administration of a NO donor to examine vascular smooth muscle function is potentially confounded by differences between subjects in body composition (i.e., fat free mass and plasma volume differ between SCI subjects and controls) and pharmacokinetic and -dynamic factors. Using intra-arterial infusion of increasing dosages of SNP to examine the NO responsiveness using a dose-response relationship, absolute and relative changes from baseline diameter at each SNP dose (Fig. 4, B and C) revealed a larger diameter increase in SCI subjects than in controls. The peak dilatation during this response corresponds with previous reports in SCI subjects and controls using sublingual NTG (2, 3, 12, 31, 32). This suggests, in keeping with previous studies (2, 3), that responsiveness to endothelium-independent nitrovasodilators may be enhanced during deconditioning. However, the inward remodeling in SCI subjects raises important issues regarding the comparison of vascular function data between groups with different arterial structure characteristics. As stated above, SCI subjects and controls possess important differences in baseline and peak SFA diameter. Simply presenting the effect of increasing SNP dosages as relative or absolute changes does not take these differences in baseline and peak diameter measurements into account. To correct for the differences in baseline and peak diameters between groups, we choose to express the SNP-mediated diameter change as a proportion of the dilator range (baseline is set at 0% and peak diameter at 100%). Using this approach, SCI subjects and controls demonstrate a similar NO sensitivity of the SFA. According to this analysis, smooth muscle cell NO sensitivity of the SFA in SCI subjects is similar between SCI and control subjects. These results emphasize the importance of using...

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Table 4. Baseline blood flow and mean wall shear rate at baseline and during the SNP infusion protocol in controls and SCI subjects

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>SNP1 (0.075 μg·min⁻¹·dl⁻¹)</th>
<th>SNP2 (0.15 μg·min⁻¹·dl⁻¹)</th>
<th>SNP3 (0.3 μg·min⁻¹·dl⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood flow, ml/min</td>
<td></td>
<td>75 ± 34</td>
<td>119 ± 30*</td>
<td>146 ± 44*</td>
</tr>
<tr>
<td>Controls</td>
<td></td>
<td>98 ± 43</td>
<td>137 ± 66*</td>
<td>169 ± 59*</td>
</tr>
<tr>
<td>Mean wall shear rate, s⁻¹</td>
<td></td>
<td>13 ± 4</td>
<td>18 ± 5</td>
<td>21 ± 7</td>
</tr>
<tr>
<td>Controls</td>
<td></td>
<td>50 ± 29</td>
<td>48 ± 19</td>
<td>50 ± 14</td>
</tr>
<tr>
<td>SCI</td>
<td></td>
<td></td>
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</tbody>
</table>

Values are means ± SD; n = 8 controls and n = 6 SCI subjects. *Post hoc P < 0.05 baseline compared with SNP infusion.
correct methodology to assess diameter changes, particularly when significant differences exist in structural characteristics of the artery. While our data from SCI subjects suggest that the vasodilator function may not be affected, this does not rule out the possibility that shorter deconditioning stimuli (i.e., bed rest) are associated with changes in NO-mediated conduit artery dilator function. Indeed, in the context of physical conditioning, it was recently suggested that the initial phase of exercise training is associated with increased endothelial NO synthase expression, which provides a short-term buffer to the increased exercise-associated shear rates (15). Possibly as a consequence, repetitive shear stress mediated NO release, and prolonged exercise training is associated with structural arterial remodeling, which normalizes shear, causing endothelial NO activity to return toward their initial levels. In this way, it is proposed that changes in arterial function and structure exist on a continuum. The results of the present study, in which chronic deconditioning is associated with inward remodeling of the SFA, with normal arterial function, may reflect an inverse of this exercise-training phenomenon. Hence, after an initial decrease in vasodilator function due to changes in shear rate in the early phase of deconditioning, conduit arteries remodel inwardly with consequent normalization of vascular function. These hypothetical considerations will require further examination.

In the present study, NO sensitivity was calculated as the relative increase of the SFA during three dosages of SNP within the diameter dilator range. Recently, McCully and coworkers published papers (27, 29, 30) on the dilator range in SCI subjects and controls. In contrast to our study, which used baseline diameter as the lower reference value for the dilator range, McCully et al. assessed “minimal” diameters. This value was defined as the arterial diameter (posterior tibial artery) at the end of 10 min of ischemia, where the imaged artery lies within the ischemia zone. One may hypothesize that using the minimal diameter represents a better approach to calculate the dilator range. However, no difference in reproducibility is evident between baseline and minimal diameters (27), and, furthermore, it may be possible to induce smaller minimal diameters with coinfusion of vasoconstrictor agents or administration of a concomitant cold pressor test. For a practical perspective, it is impossible to place an effective occluding cuff proximal to the probe position when imaging of the SFA occurs ~3 cm distal of the bifurcation into the deep femoral artery and the SFA.

Limitations

As indicated above, our SCI results relating to smooth muscle cell function differ from some previous studies. We cannot rule out the possibility that differences in subject characteristics between studies may contribute to this disparity. However, our inclusion and exclusion criteria are similar to those of previous studies, and we think it unlikely that our subjects were unique in some way. While we excluded smokers, subjects with cardiovascular disease and risk factors (e.g., diabetes and insulin resistance), which are known to impair FMD responses (6, 7, 14, 33), we did not quantify body composition. However, we can anecdotaly report that the subjects appeared to be of similar body composition, notwithstanding their lower limb atrophy. Another limitation of our study is related to the diameter analysis, which was performed using the manual caliper placement technique, as automatic edge detection and wall tracking were not available at the time. The latter results in less potential for bias and improved reproducibility, compared with the manual caliper placement (35), and we, therefore, recommended it as an optimal approach for contemporary ultrasound studies. Nevertheless, we have established that our technique demonstrates good reproducibility (coefficient of variation = 15%) for SFA FMD studies, and our approach does not differ much from that reported in numerous other recent publications in high-impact journals (8, 16, 38). The present results are also limited to the SFA, and we cannot exclude the possibility that other lower limb arteries may respond differently. Although the SFA FMD response is NO dependent (20), there is no evidence available regarding how well SFA function serves as a surrogate for coronary function. A significant correlation is, however, present between brachial and coronary NO-mediated dilation (1). Nonetheless, superficial femoral FMD is NO mediated and, similar to coronary arteries, prone to atherosclerotic plaque formation, and may, therefore, be related to cardiovascular risk. Future studies should further examine this hypothetical consideration.

In conclusion, this study demonstrated an inward remodeling of the SFA in the extremely inactive legs of SCI individuals. The smaller diameter leads to markedly higher shear rates at rest, but also during periods of increased flow in the SFA. Correcting the diameter change after 5 min of ischemia for the increased shear rate stimulus, SCI individuals demonstrate a preserved FMD response. Moreover, taking the differences in baseline and peak diameter into account (using the diameter dilator range), NO sensitivity of the SFA in SCI is similar to that observed in able-bodied controls. This suggests that deconditioning in the extremely inactive legs of SCI individuals results in structural change of conduit arteries (i.e., an inward remodeling), while vasodilator function is preserved.

GRANTS

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