Shear stress levels in paralyzed legs of spinal cord-injured individuals with and without nerve degeneration

CÉCILE R. L. BOOT,¹ JAN. T. GROOTHUIS,¹ HERMAN VAN LANGEN,² AND MARIA T. E. HOPMAN¹

¹Department of Physiology, and ²Clinical Vascular Laboratory, University Medical Center Nijmegen, 6500 HB Nijmegen, The Netherlands

Received 12 April 2001; accepted in final form 21 January 2002

The purpose of this study was to assess the relationship between inactivity and shear stress, the frictional force of blood against the endothelium, in spinal cord injury (SCI) subjects. SCI group offers a unique "model of nature" to study the effects of inactivity. Nine SCI subjects with upper (SCI-U) and 5 with a lower (SCI-L) motoneuron lesion and 10 able-bodied controls (C) were included. A venous blood sample was withdrawn to determine blood viscosity. Red blood cell velocities and arterial diameters of the common carotid artery (CCA) and common femoral artery (CFA) were measured by using echo-Doppler ultrasound in a supine position. No differences were observed in wall shear stress in the CCA between groups. In the CFA, peak and mean wall shear stress were significantly increased in SCI (14.1 and 1.2 Pa, respectively) compared with C (10.2 and 0.9 Pa, respectively). Because SCI-U and SCI-L showed no differences in shear stress levels, inactivity and nerve degeneration seems to cause the elevated shear stress levels in the CFA in SCI. However, the lack of central neural control as a causal factor cannot be ruled out.

Cardiovascular diseases are thought to be related to the arterial endothelium, the innermost cell layer of blood vessels continuously in contact with the bloodstream. A high correlation between risk factors for cardiovascular disease and endothelial dysfunction has been reported (15, 26). Shear stress is considered to be a key factor in the regulation of endothelial function (21). It is defined as the frictional force of blood against the endothelium. Shear stress is directly proportional to the viscosity of blood and to red blood cell velocity and is inversely related to vessel diameter.

Usually shear stress levels are kept at a constant level by adjusting the arterial diameter (17, 21). It is known that the diameter of conduit arteries adapts to changes in blood flow induced by activity and training as well as by inactivity and muscle atrophy (8, 11, 12). Research on animals demonstrates that a decrease in blood flow leads to arterial constriction, followed by remodeling of the arterial wall, which results in a reduced internal diameter within 2–3 wk (3, 12, 13). Although inactivity is a known risk factor for cardiovascular disease (24), the effect of inactivity and concomitant flow reduction on shear stress levels and endothelial function is unknown.

In individuals with spinal cord injury (SCI), the part of the body below the lesion level is paralyzed and is thus extremely inactive. The extremely low level of inactivity of the paralyzed legs is independent of the fitness status of the individual (18). The SCI population therefore offers a unique “model of nature” to study the effect of extreme inactivity on the peripheral circulation and, more specifically, the effect of inactivity on shear stress. As far as we know, no studies have been conducted on shear stress levels in SCI. Recently, Schmidt-Trucksäss et al. (25) reported high shear rate levels in the femoral artery in SCI compared with able-bodied controls (C). Examining shear stress levels in the extremely inactive legs of SCI will be of great physiological and clinical value.

To assess whether changes in shear stress levels in vessels below the lesion level in SCI are the result of loss of perivascular nerves (nerve degeneration) or of inactivity, SCI individuals with an upper (SCI-U; intact nerves but no central command) and SCI individuals with a lower (SCI-L; nerve degeneration and no central command) motoneuron lesion were included.

The purpose of this study was to investigate the effect of inactivity on shear stress levels in the peripheral circulation by comparing shear stress levels in the common femoral artery (CFA) between SCI individuals with paralyzed legs and C with normal leg muscle activity. To distinguish between the effect of nerve degeneration and inactivity, shear stress levels in SCI-U and SCI-L individuals were compared.

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
Subjects. Twenty-five individuals volunteered to participate in this study: 15 individuals with SCI of traumatic origin at thoracic level and 10 C. In the SCI group, 10 subjects had an SCI-U between T4 and T12, resulting in paralysis without nerve degeneration. Five SCI subjects had a SCI-L between T10 and T12, which results in paralysis with nerve degeneration. Percutaneous electrical stimulation was used to divide SCI individuals in SCI-L and SCI-U, with SCI-L individuals showing no muscle contraction on stimulation due to nerve degeneration.

Group characteristics, including time since injury and physical activity, are summarized in Table 1. Neurological examinations of the motor and sensor neurological system revealed completeness and level of the lesion. All but two subjects with SCI had a complete sensor and motor lesion (ASIA A) (16). One of the subjects had a complete motor lesion but was sensory incomplete (ASIA B). The other subject had an incomplete sensor and motor lesion, although the motor activity was not functional (ASIA C).

All subjects completed a medical health questionnaire. Three SCI-U individuals used medication: atenolol, metoprolol, lisinopril (for treatment of hypertension), and terazos (to prevent bladder spasms). The study was approved by the Faculty Ethics Committee, and all subjects signed an informed consent before starting the tests.

Protocol. All subjects refrained from caffeine, nicotine, and alcohol for at least 12 h before starting the test. A venous blood sample was obtained from all subjects for determination of blood viscosity. The tests were performed in one experimental room, in which the temperature was kept constant between 22 and 24°C, and were started between 11 AM and 2 PM. After completing the medical health questionnaire, subjects were positioned on a bed in the supine position for the measurements.

After a 15-min supine rest period, red blood cell velocity of the common carotid artery (CCA) was measured for 2 min, followed by a measurement of the diameter of the artery. This was followed by a measurement of both entities of the CFA.

Measurements. Peak (Vpeak), mean (Vmean), and minimal (Vmin) red blood cell velocity and longitudinal systolic and diastolic vessel diameter were measured with a pulsed color-coded Doppler device (ATL 5000 HDI). A 7-MHz broadband linear array transducer (L7-4) was used. The sample volume was placed in the center of each vessel, 2 cm before bifurcation of the left CFA into the deep and superficial femoral artery, and 1.5 cm before the bifurcation of the left CCA into the external and internal carotid artery. All measurements were performed by the same experienced examiner. In the CCA and CFA, red blood cell velocity was measured beat to beat for 2 min and stored once every 10 s. An average of 12 values (2 min) was calculated for Vpeak and Vmean of CCA and Vpeak and Vmin of CFA. Vmean in the CFA was calculated off-line by using the equation 0.5[0.25Vpeak + (Vmin/6)](60/heartrate) (27). Systolic (the largest) and diastolic (the smallest) vessel diameters of the CCA and CFA were measured three times off-line, and averaged values were calculated. Mean diameter was calculated as one-third of systolic diameter plus two-thirds of diastolic diameter.

Blood viscosity was measured with a rotational viscometer (50 s⁻¹) (Emilia Rheometer, Reciprotor, Denmark). Calibration was performed with a physiological solution (0.9% NaCl) before every measurement. All measurements were performed in duplo by the same analyst, and the two values were averaged.

Data analysis. Peak wall shear rate (PWSR; in s⁻¹) was calculated as the Vpeak (m/s) multiplied by four and divided by the systolic diameter (m). Mean wall shear rate (MWSR; in s⁻¹) was obtained from the Vmean (m/s) multiplied by four and divided by the average of systolic and diastolic diameter (m). Peak wall shear stress (PWSS; in Pa) and mean wall shear stress (MWSS; in Pa) were calculated by multiplying PWSR and MWSR (s⁻¹) with blood viscosity (Pa·s⁻¹).

Statistical analysis. Differences in wall shear (PWSR, MWSR, PWSS, and MWSS), diameter (systolic and diastolic), and Vpeak and Vmean in CCA and CFA and in blood viscosity were assessed between the C and SCI group and between the SCI-L and SCI-U group by using Student’s t-tests for independent groups. Normal distribution was verified by calculating skewness. P values of <0.05 were considered to indicate statistical significance. Results are expressed as means ± SD.

RESULTS

No significant differences were found in any of the subject characteristics between the SCI and C groups nor were any differences observed in these variables between SCI-U and SCI-L (Table 1). One subject of the SCI-L group was excluded because of high blood pressure (165/115) (n = 9) because it is known that high blood pressure affects shear stress levels. The other two subjects on medication did not show any deviating results from the total SCI group. Two subjects of the SCI-U group did not have a three-phase spectrum in the CFA. As a result, the formula used for off-line calculation was not applicable, and therefore Vmean could not be calculated for these two subjects.

No differences were observed in PWSS and MWSS in the CCA between any of the groups. In the CFA, PWSS and MWSS were higher in the SCI (PWSS: 14.1 ± 3.9 Pa (P = 0.01); MWSS: 1.2 ± 0.5 Pa (P = 0.07)) than in the C group (PWSS: 10.2 ± 2.9 Pa; MWSS: 0.9 ± 0.3 Pa). No differences in PWSS and MWSS were observed between the SCI-U and SCI-L groups (Fig. 1).

PWSR and MWSR of the CCA showed no differences between groups. In the CFA, PWSS and MWSS were significantly higher in SCI [PWSS: 525 ± 165 s⁻¹ (P =

Table 1. Subject characteristics

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>BM, kg</th>
<th>Height, m</th>
<th>Age, yr</th>
<th>PA, h/wk</th>
<th>TI, yr</th>
</tr>
</thead>
<tbody>
<tr>
<td>C</td>
<td>10</td>
<td>70.8 ± 7.6</td>
<td>1.78 ± 0.1</td>
<td>34.0 ± 12</td>
<td>2.95 ± 2.3</td>
<td></td>
</tr>
<tr>
<td>SCI</td>
<td>14</td>
<td>71.5 ± 14</td>
<td>1.82 ± 0.1</td>
<td>36.3 ± 8.5</td>
<td>2.32 ± 1.5</td>
<td>10.0 ± 6.3</td>
</tr>
<tr>
<td>SCI-U</td>
<td>9</td>
<td>72.8 ± 11</td>
<td>1.82 ± 0.1</td>
<td>36.4 ± 7.6</td>
<td>2.61 ± 1.2</td>
<td>10.9 ± 7.2</td>
</tr>
<tr>
<td>SCI-L</td>
<td>5</td>
<td>69.2 ± 18</td>
<td>1.81 ± 0.1</td>
<td>36.0 ± 11</td>
<td>1.80 ± 2.1</td>
<td>8.5 ± 7.9</td>
</tr>
</tbody>
</table>

Values are means ± SD. Body mass (BM), physical activity (PA), and time since injury (TI) of the control group (C), the group with spinal cord injury (SCI) with upper motoneuron lesion (SCI-U), and the group with lower motoneuron lesion (SCI-L).
Comparison with C (PWSR: 34.2 ± 7.7 s⁻¹; MWSR: 29.6 ± 8.9 s⁻¹), whereas no differences were observed in PWSR and MWSR between SCI-U and SCI-L (Fig. 2).

No differences between groups were observed in the diameter of the CCA. Systolic as well as diastolic diameter of the CFA was smaller in the SCI group compared with the C group (P = 0.05) (Fig. 3). No differences in diameters were observed between the SCI-U and SCI-L group.

No differences between groups were observed in V_peak and V_mean in the CCA or in the CFA (Table 2 and 3). Blood viscosity was not significantly different between C (2.96 ± 0.48 Pa·s) and SCI (2.72 ± 0.56 Pa·s) or between SCI-U (2.84 ± 0.38 Pa·s) and SCI-L (2.51 ± 0.20 Pa·s). Duplo measurements of blood viscosity showed a relative error of 2%.

DISCUSSION

This is the first study, as far as we know, to examine the effect of inactivity on vascular wall shear stress in SCI subjects. Individuals with SCI offer a unique model of nature to study the effects of inactivity. The main finding of this study is that arterial wall shear stress increases after SCI, which may be caused primarily if not exclusively by inactivity.

CCA. Shear stress in the CCA was examined as a reference value. Because it is positioned above the lesion level, no differences were expected between C and SCI subjects. The present study confirmed this expectation by demonstrating no differences in vessel characteristics of the CCA between SCI and C. However, a recent study of Schmidt-Trucksäss et al. (25) showed a lower shear rate in CCA in SCI individuals compared with sedentary and trained individuals without SCI. This discrepancy may be explained by differences in level of activity of SCI individuals between studies and in the duration of the lesion and thus in the duration of the wheelchair-bound lifestyle.

Table 2. Peak red blood cell velocity

<table>
<thead>
<tr>
<th>Group</th>
<th>CCA</th>
<th>CFA</th>
</tr>
</thead>
<tbody>
<tr>
<td>C</td>
<td>1.03 ± 0.23</td>
<td>0.87 ± 0.17</td>
</tr>
<tr>
<td>SCI</td>
<td>0.97 ± 0.16</td>
<td>0.99 ± 0.22</td>
</tr>
<tr>
<td>SCI-U</td>
<td>0.97 ± 0.16</td>
<td>0.90 ± 0.29</td>
</tr>
<tr>
<td>SCI-L</td>
<td>0.98 ± 0.17</td>
<td>1.15 ± 0.19</td>
</tr>
</tbody>
</table>

Values are means ± SD. Peak red blood cell velocities (m/s) in the common carotid (CCA) and common femoral arteries (CFA) in the C and SCI groups are shown. SCI group is divided into SCI-U and SCI-L groups.
Table 3. Mean red blood cell velocity

<table>
<thead>
<tr>
<th>Group</th>
<th>CCA</th>
<th>CFA</th>
</tr>
</thead>
<tbody>
<tr>
<td>C</td>
<td>0.40 ± 0.07</td>
<td>0.10 ± 0.05</td>
</tr>
<tr>
<td>SCI</td>
<td>0.43 ± 0.09</td>
<td>0.24 ± 0.17</td>
</tr>
<tr>
<td>SCI-U</td>
<td>0.41 ± 0.04</td>
<td>0.21 ± 0.16</td>
</tr>
<tr>
<td>SCI-L</td>
<td>0.46 ± 0.13</td>
<td>0.30 ± 0.20</td>
</tr>
</tbody>
</table>

Values are means ± SD. Mean red blood cell velocities (m/s) in the CCA and CFA in the C and SCI groups are shown. SCI is divided into SCI-U and SCI-L groups.

PWSS and MWSS levels in CCA reported here are in agreement with earlier findings in a healthy population (4, 9, 10) but in contrast, i.e., markedly lower, with values of wall shear rate in the CCA reported in another study (22). This may be explained by differences in methods to calculate wall shear rate. Samijo et al. (22) calculated wall shear rate by using complete velocity profiles recorded by means of received ultrasonographic radio frequency signals, whereas in the present study red blood cell velocity was calculated from the velocity spectrum received by a 2-mm sample volume in the middle of the vessel. The inclusion of a C group, measured with the same technique and by the same examiner as in the present study, is therefore of great importance. In addition, the coefficient of variance for the measurement set up in this study, assessed in six subjects who were all measured twice within 2 wk, was 6% (diameter) and 14% (shear stress) in the femoral artery and 6% (diameter) and 10% (shear stress) in the carotid artery, which includes physiological as well as physical variation. These reproducibility data are in line with values found by Demolis et al. (5), who reported a variation coefficient of 4–5% for the diameter and 10–12% for red blood cell velocity in the femoral and carotid artery.

Absolute values of \( V_{\text{peak}} \) and \( V_{\text{mean}} \) in CCA as well as in diameters of the CCA in both groups were in agreement with results previously reported (2, 4, 9, 10, 19, 22, 28) and were not different between the C and SCI groups, indicating that a spinal cord lesion does not influence these characteristics of the CCA.

CFA. In the CFA, wall shear rate and wall shear stress levels were significantly higher in the SCI than in the C group. The fact that wall shear rate and wall shear stress did not show any differences between SCI-L and SCI-U seems to indicate that inactivity and not nerve degeneration is the major causal factor, which explains the elevated wall shear rate and wall shear stress levels.

PWSR is in agreement with the results of Schmidt-Trucksäss et al. (25), who reported PWSR in a SCI population (588 ± 120 s\(^{-1}\)) to be higher than in C (356 ± 113 s\(^{-1}\)).

This is the first study to show that wall shear stress in the CFA is higher in SCI than in C, which could be the result of either a decrease in diameter, an increase in red blood cell velocity, an increase in blood viscosity, or a combination of these factors.

The diameter of the CFA is significantly lower in the SCI group compared with the C group, which is consistent with earlier studies reporting diameters of the CFA between 8 and 10 mm in C (23) and between 5 and 7 mm in SCI (8, 11, 19). This indicates that the peripheral circulation in SCI patients adapts to the inactivity of the paralyzed muscles by vascular atrophy.

No differences in \( V_{\text{peak}} \) in the CFA were observed between the SCI and C groups, which is in contrast with a study of Hopman et al. (11) that showed \( V_{\text{peak}} \) in SCI individuals to be significantly lower (0.56 m/s) than in C (0.76 m/s). Possible explanations for the discrepancy in results may be related to differences in echo-Doppler machines, differences in examiner, and differences in analyzing the spectrum of the red blood cell velocity, i.e., tracing vs. peak detection. However, the more important variable with respect to blood flow and MWSS, \( V_{\text{mean}} \), was not different from previous studies (11) and did not show any differences between the SCI and C groups. Red blood cell velocity, therefore, seems not to be the key variable to explain the differences found in shear stress between groups.

Blood viscosity showed no differences between the C and SCI groups. No earlier studies have measured blood viscosity in SCI individuals. Because of the different methods of measurement of blood viscosity, comparison of absolute values between studies is not always valid. Previous studies (6, 14) reported blood viscosity values of 3.6 and 3.8 Pa·s, whereas in the present study, with good relative errors of 2%, values between 2.5 and 3.0 Pa·s were found, which again stresses the importance of a C group within the study. Because no differences between groups were observed, it is unlikely that blood viscosity is a determinant of changes in wall shear stress after inactivity.

It can be concluded that the smaller diameter of the CFA in SCI compared with C explains the changes in wall shear stress for the most important part. It is generally accepted that arteries adapt to chronic changes in blood flow by undergoing compensatory adjustments of their internal diameters (17, 21). The effect of this compensatory response is the maintenance of mean arterial hemodynamic shear stress. It has now been shown that this process depends on intact endothelium (13), most likely via modulation in production of vasoactive substances by endothelial cells such as nitric oxide and endothelin-1 (20). SCI seems to disturb this process and the diameter decreases more than one would expect on the basis of flow reduction, leading to elevated levels of arterial shear stress.

From animal studies (1), it is known that long-term sympathectomy and nerve degeneration may alter endothelial function, leading to a reduction in endothelial nitric oxide synthase expression and increase in endothelin-1. It may be hypothesized that perivascular nerves play a role in endothelial function and thus in the regulation of shear stress levels. The fact, however, that shear stress levels are similarly elevated in SCI subjects with SCI-U (intact nerves) as well as SCI-L (nerve degeneration) seems to dismiss this hypothesis and would suggest that inactivity plays a more important role. If nerve degeneration would have a major
effect on wall shear stress levels, wall shear stress in SCI-L should have been different from that in SCI-U. The absence of central cerebral control below the level of the lesion in both SCI-L and SCI-U may be another contributing factor. Although it has never been studied or suggested before, it may be the lack of central cerebral control that leads to endothelial dysfunction and changes in arterial wall structure (7) rather than the absence of perivascular nerve activity. This suggestion, however, needs further exploration.

The clinical consequences of chronically enhanced arterial shear stress levels in humans are unknown. It has been established that atherosclerotic lesions colocalize with regions of low shear stress throughout the arterial tree (10). From in vitro experiments, we know that supraphysiological shear stress levels induce endothelial injury, whereas it has been suggested from animal experiments that slightly elevated levels of arterial shear stress may lead to an atheroprotective endothelial phenotype (26). Extrapolation from in vitro data or animal studies to the human organism may be difficult, especially with regard to the difference in time frame, i.e., the chronic adaptive process developing over years in humans vs. the short term intervention in the animal studies. Additional research using longer time frames, including experimental models of human disease, is clearly needed.

Limitations. A potential limitation of the present study is the lack of a subject group with inactivity without the loss of central cerebral control. Human models for inactivity, like leg fracture or bed rest, to study the peripheral circulation may have the disadvantage of confounding processes like bone repair (and concomitantly enhanced blood flow) or systemic circulatory changes (like loss of plasma volume), respectively. We, therefore, chose a study design with a unique setup of SCI individuals with extreme inactivity of the lower part of the body (independent of fitness level or sport participation) with nerve degeneration (SCI-L) and without nerve degeneration (SCI-U). This enabled us to separate inactivity from nerve degeneration and to draw more firm conclusions on the causality of shear stress adaptation to inactivity in humans with SCI with and without nerve degeneration. However, with this study design, we could not assess the contribution of cerebral control in shear stress regulation. Additional research with healthy individuals casting an arm or leg would be the optimal study design to solve the above-mentioned problem.

In conclusion, the present study demonstrates that wall shear stress in the femoral artery is higher in SCI than in C resulting from inactivity rather than possible structural wall changes due to chronic sympathectomy from nerve degeneration, although the endothelial dysfunction and changes in arterial wall structure due to chronic sympathectomy or lack of central cerebral control in SCI cannot be ruled out.

Participation of all subjects was greatly appreciated. We thank Marlous Brok of the Clinical Vascular Laboratory for echo-Doppler measurements.

REFERENCES


