Cardiovascular effects of partial sleep deprivation in healthy volunteers

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1Sleep Laboratory of Pulmonary Division of Heart Institute (InCor), University of São Paulo Medical School, São Paulo, Brazil; 2Hypertension Unit of Heart Institute (InCor), University of São Paulo Medical School, São Paulo, Brazil; 3Federal University of Rondônia, Rondônia, Brazil; 4Nove de Julho University, São Paulo, Brazil; and 5Medical School of University of Campinas, São Paulo, Brazil

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Dettoni JL, Consolin-Colombo FM, Drager LF, Rubira MC, de Souza SB, Irigoyen MC, Mostarda C, Borile S, Krieger EM, Moreno Jr, Lorenzi-Filho G. Cardiovascular effects of partial sleep deprivation in healthy volunteers. J Appl Physiol 113: 232–236, 2012. First published April 26, 2012; doi:10.1152/japplphysiol.01604.2011.—Sleep deprivation is common in Western societies and is associated with increased cardiovascular morbidity and mortality in epidemiological studies. However, the effects of partial sleep deprivation on the cardiovascular system are poorly understood. In the present study, we evaluated 13 healthy male volunteers (age: 31 ± 2 yr) monitoring sleep diary and wrist actigraphy during their daily routine for 12 nights. The subjects were randomized and crossover to 5 nights of control sleep (>7 h) or 5 nights of partial sleep deprivation (<5 h), interposed by 2 nights of unrestricted sleep. At the end of control and partial sleep deprivation periods, heart rate variability (HRV), blood pressure variability (BPV), serum norepinephrine, and venous endothelial function (dorsal hand vein technique) were measured at rest in a supine position. The subjects slept 8.0 ± 0.5 and 4.5 ± 0.3 h during control and partial sleep deprivation periods, respectively (P < 0.01). Compared with control, sleep deprivation caused significant increase in sympathetic activity as evidenced by increase in percent low-frequency (50 ± 15 vs. 59 ± 8) and a decrease in percent high-frequency (50 ± 10 vs. 41 ± 8) components of HRV, increase in low-frequency band of BPV, and increase in serum norepinephrine (119 ± 46 vs. 162 ± 58 ng/ml), as well as a reduction in maximum endothelial dependent venuodilation (100 ± 22 vs. 41 ± 20%; P < 0.05 for all comparisons). In conclusion, 5 nights of partial sleep deprivation is sufficient to cause significant increase in sympathetic activity and venous endothelial dysfunction. These results may help to explain the association between short sleep and increased cardiovascular risk in epidemiological studies.

sympathetic activity; endothelial dysfunction

INTENTIONAL SLEEP CURTAILMENT is increasingly common in our society. “Normal” average sleep duration has decreased from ~9 h per night in 1910 to ~7 h currently (28). There is however, increasing evidence that sleep loss may be harmful not only to neurocognitive functions but also to the cardiovascular system (3, 7, 16). Recent epidemiological studies (10, 31) have linked short hours of sleep (<5 h) to increased risk of developing hypertension. In the Nurse Health Study (2), short sleep was associated with an increased risk of death due to cardiovascular events, mainly acute myocardial infarction and stroke. A recent large epidemiological study showed that sleep duration is inversely associated with incident coronary artery calcification over 5 yr of followup (15).

Among the possible mechanisms linking sleep disturbances and cardiovascular disease, sympathetic activation and inflammation are suspected to induce endothelial dysfunction, a key factor in the increased risk of cardiovascular disease. One recent study (23) showed decreased endothelial-dependent and -independent cutaneous vascular reactivity after acute exposure to 40 h of total sleep deprivation in health subjects. The effects of acute total sleep deprivation on sympathovagal balance brought conflicting results. While some studies (14, 20) reported no significant changes in resting heart rate (HR), increased blood pressure (BP), and a reduction in muscle sympathetic nerve activity, others (17) showed increased sympathetic activity and HR.

Partial sleep deprivation is increasingly common in modern societies and has been less studied than acute total sleep deprivation. Therefore, the main objective of the present study was to investigate the effects of five nights of partial sleep deprivation on cardiac autonomic balance, on peripheral sympathetic activity, and on vascular reactivity in young subjects engaged in their routine activities. To this end, we performed a randomized and crossover study that compared a period of control sleep (>7 h) with a period of partial sleep deprivation (<5 h) in young healthy subjects free of comorbidities. We reasoned that 5 nights of partial sleep deprivation is a relevant model and corresponds to a common situation experienced in a working week in western societies. We hypothesized that partial sleep deprivation for only 5 nights will promote significant impairment on cardiac autonomic balance, on peripheral sympathetic activity, and on vascular reactivity.

METHODS

Volunteers. Healthy young male volunteers were recruited for the study. All subjects were clinically evaluated, including measurements of weight and height. Exclusion criteria included age <21 and >45 yr, overweight and obese participants (body mass index: >25 kg/m²), smoking, use of chronic medications, and any established medical condition including diabetes mellitus, hypertension, dyslipidemia, heart diseases, and sleep disordered breathing (apnea-hypopnea index: ≥5 events/h by full polysomnography). Subjects that failed to comply with the sleep schedule for >30 min for 2 nights or >1 h for 1 night were further excluded. The Institutional Review Committee of the Hospital das Clínicas da Universidade de Sao Paulo, Brazil, approved this study after written informed consent was obtained from all study subjects.

Polysomnography. All volunteers underwent overnight polysomnography using a digital system (17 channels, EMbla Medicare; Flaga hf. Medical Devices), as previously described in our laboratory (4). Briefly, the following variables were monitored: electroencepha-
logram, electrooculogram, electromyogram using surface electrodes, electrocardiogram, snoring, and body position. Airflow was detected by thermocouple and by pressure flow transducer. Chest and abdominal piezo sensors monitored respiratory effort. Arterial oxygen saturation (SaO₂) and pulse were recorded with a pulse oximeter. All polysomnography was performed and scored based on the guidelines for sleep studies (21). Apnea was defined as cessation of inspiratory airflow for ≥10 s associated with at least a 3% drop in arterial oxyhemoglobin saturation, arousal, or both (8). Hypopnea was defined as a significant (>50%) reduction in airflow lasting 10 s or more. The apnea-hypopnea index was defined by the number of apneas and hypopneas per hour of sleep.

**Sleep monitoring.** During the study period, all subjects were continuously monitored by a wrist actigraphy device (Basic Mini Motionlogger Actigraph Ambulatory Monitoring; Ardsley, New York, NY) worn on the nondominant hand. The Actigraph was set to sample movements in 1-min periods. Actigraph records were automatically processed by use of AW2 software version 2.30.1 program (Ambulatory Monitoring) to extract information on sleep duration. Nap duration per day (≥5 min) was estimated by the remaining daytime sleep periods, which was confirmed by diaries entries.

**Blood samples.** Venous blood sample was collected from all participants between 8 and 10 AM after 30 min of rest in the supine position for the measurement of plasma norepinephrine (measured by high-performance liquid chromatography) and other routine laboratory measurements (glucose, total cholesterol, low-density lipoprotein, high-density lipoprotein, and red blood cell count).

**BP, HR, and autonomic measurements.** BP waveforms were obtained by a digital photoplethysmograph device (Finometer; Finapres Medical System) while subjects were awake in a supine position; all studies were performed in the morning between 8 and 10 AM during a 10-min period. A software program (BeatScope) used the BP curves and patient age, sex, weight, and height values to calculated systolic and diastolic BP, HR, cardiac output, and peripheral vascular resistance. The waveforms were simultaneously recorded on another computer equipped of acquisition and conversion of biological signals AT/MCA-CODAS (DATA Instruments, Akron, OH). The sampling frequency of signals was 1,000 Hz as previously described (5). These stored data underwent a routine analysis to provide the values of HR and BP variability.

Each heart beat was identified by the use of specialized algorithm implemented for the Matlab MT (MATLAB 6.0, Mathworks), which makes the automatic detection of events of systolic and diastolic pressure waves. Pulse interval or R-R interval was calculated as the difference between the beginning and end points of the cycle (t₁–t₀).

The power spectral density of R-R interval was obtained by the fast Fourier transformation using the Welch’s method over 16,384 points with a Hanning window and 50% overlapping. The spectral bands for Fourier transformation using the Welch’s method over 16,384 points presented as normalized dose-response curves in which the diameter of the vein during saline infusion was defined as 100% dilatation. The vein was preconstricted to 20% of the baseline size by infusing increasing doses of phenylephrine, a selective α₁-adrenergic receptor agonist (99–3,166 ng/min). The infusion rate of phenylephrine inducing 80% venoconstriction was kept constant during the entire study, and this degree of constriction was defined as 0% dilatation for the purpose of subsequent calculations. The vasodilator response expressed in this study was calculated as a percentage of the range between 0 and 100% dilatation (24). Drugs were infused using a Harvard infusion pump (Harvard Apparatus, South Natick, MA) at a flow rate of 0.3 ml/min.

**Experimental design.** The entire study period was 12 nights. The participants were randomized to 5 nights of extended sleep (Control period) or 5 nights of partial sleep deprivation (SD period) that was crossed over and interposed by 2 nights of unrestricted sleep (wash out). During the control period, subjects were instructed to sleep 8 h, ranging from a minimum of 7 h to a maximum of 9 and 1/2 h. Partial sleep deprivation consisted of a target sleep of <5 h and always ≥3 h and 30 min. The subjects were instructed to sleep later and wake up at the same time. We reasoned that this would be an easier protocol to follow and would better correspond to the sleep restriction regimen frequently experienced by our society. Subjects who did not comply with the sleep schedule were excluded from the final analysis. The subjects were instructed to keep regular activities and not travel and should not enter the study if any stress factor such as study exams were expected during the study period. In addition, the subjects were instructed not to take caffeine or alcohol during the study period. All measurements described above were made in the morning of the 6th day, i.e., at the end of control and partial sleep deprivation periods.

**Statistical analysis.** Continuous variables were normally distributed and described as means ± SD. The comparison between control and partial sleep deprivation was made by using the paired Student t-test. Comparisons of dose-response curves of endothelium venodilation were made by means of repeated ANOVA (Instat, version 3, GraphPad; La Jolla, CA). Based on a pilot study of 5 subjects, we calculated that 12 subjects would be necessary to show significant differences (α = 0.05) in endothelial function with a power of 80%. A value <0.05 was considered statistically significant.

**RESULTS**

We initially recruited 18 healthy male volunteers for the study. Two subjects were excluded before study entry because of mild sleep apnea. Three subjects were excluded from the final analysis because they did not comply with the sleep schedule. We therefore included in the final analysis 13 Caucasian subjects, aged 31 ± 2 yr, and with a body mass index of 23.7 ± 0.8 kg/m². All subjects included were kept in their regular routine and did not travel during the study period. The polysomnography of the subjects included confirmed absence of sleep apnea (apnea-hypopnea index of 2.3 ± 0.8 events/h). During regular sleep, subjects slept 8.0 ± 0.5, while in partial sleep deprivation they slept 4.5 ± 0.3 h (P < 0.003).

Partial sleep deprivation did not change office measurements of resting HR, systolic and diastolic BP, respiratory rate, and cholesterol levels (Table 1). However, partial sleep deprivation was associated with a significant increase in plasma norepinephrine levels (Table 1). Furthermore, import alterations were observed in HR and BP variability associated with sleep pattern modifications. After partial sleep deprivation, a significant increase in sympathetic and a decrease in parasympathetic modulation of cardiac autonomic balance, characterized by increased percent LF component (Fig. 1A), decreased percent HF component (Fig. 1B), and increased LF-to-HF ratio (Fig. 1C) was observed. The LF component of BP variability also had a significant increase after sleep deprivation (6.42 ±
1.83 and 11.49 ± 2.7 normalized units, respectively). No changes were observed on HF component of BP variability (1.58 ± 0.3 and 2.11 ± 1.3 normalized units, comparing control and after partial sleep deprivation, respectively).

The mean doses of phenylephrine to reach 20% constriction of the vein was significantly lower after partial sleep deprivation compared with normal control sleep (846 ± 189 vs. 332 ± 107 ng/ml, respectively; P < 0.01). Moreover, a significant reduction in maximum venous vasodilatation induced by acetylcholine was elicited in response to partial sleep deprivation (Fig. 2). Also, with the analysis that the venodilation of the acetylcholine dose-response curve (0.36 –3,600 ng/min), it was possible to show that for each dose of Ach tested the venodilation was lower after partial sleep deprivation compared with control sleep (Fig. 3; P < 0.001). No significant changes in maximum dorsal hand vein vasodilation in response to sodium nitroprusside were detected.

**DISCUSSION**

Supporting our hypothesis, the major finding in this study is that partial sleep deprivation causes a significant increase in cardiac and peripheral sympathetic modulation, associated with a decrease in endothelial-dependent venodilation in healthy, young male subjects. To the best of our knowledge, this is the first study to demonstrate that partial sleep deprivation for 5 nights is able to cause a decrease in endothelial-dependent venodilation induced by acetylcholine. Together, these results may help to explain the association between short sleep and increased cardiovascular risk in epidemiological studies.

Total sleep deprivation has been extensively studied and causes an increase in BP with variable effects on sympathetic activity (14, 19, 20, 23, 29, 34). In contrast, few studies have evaluated the cardiovascular effects of partial sleep deprivation. Moreover, these few studies used a wide range of protocols. For instance, Meier-Ewert et al. (18) found significant rise in resting HR but not in BP after an aggressive protocol of partial sleep deprivation for 10 nights. Takase et al. (27) observed increased levels of plasma norepinephrine in college students after 4 wk of studying for final examinations, who experienced both sleep deprivation (evaluated by questionnaires) and stress. In contrast to the previous studies, we evaluated young, healthy subjects free of comorbidities who were engaged in their normal routine in a relatively mild model of sleep deprivation.

**Table 1.** Hemodynamic and laboratory measurements after control sleep and partial sleep deprivation periods

<table>
<thead>
<tr>
<th>Variables</th>
<th>Control Sleep Period</th>
<th>Partial Sleep Deprivation Period</th>
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<tbody>
<tr>
<td><strong>Hemodynamic parameters</strong></td>
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<tr>
<td>Heart rate, beats/min</td>
<td>61 ± 6</td>
<td>63 ± 8</td>
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<tr>
<td>SBP, mmHg</td>
<td>108.8 ± 9.8</td>
<td>112.9 ± 11.7</td>
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<tr>
<td>DBP, mmHg</td>
<td>63.6 ± 4.8</td>
<td>65.3 ± 6.9</td>
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<tr>
<td>Respiratory rate, min</td>
<td>16.0 ± 4.3</td>
<td>17.6 ± 4.3</td>
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<td><strong>Laboratory measurements</strong></td>
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<tr>
<td>Cholesterol, mg/dl</td>
<td>162 ± 20</td>
<td>160 ± 22</td>
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<tr>
<td>LDL cholesterol, mg/dl</td>
<td>92 ± 7</td>
<td>93 ± 6</td>
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<tr>
<td>HDL cholesterol, mg/dl</td>
<td>42 ± 2</td>
<td>43 ± 3</td>
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<tr>
<td>Glucose, mg/dl</td>
<td>78 ± 10</td>
<td>81 ± 11</td>
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<tr>
<td>Norepinephrine, ng/ml</td>
<td>119 ± 46</td>
<td>162 ± 58*</td>
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Values are means ± SE. SBP and DBP, systolic and diastolic blood pressure. *P < 0.01 partial sleep deprivation vs. control sleep period.

**Fig. 1.** Heart rate variability after control sleep and partial sleep deprivation periods. A: %low frequency (LF) component. B: %high frequency (HF) component. C: LF-to-HF ratio. *P < 0.05 vs. control sleep.

**Fig. 2.** Maximum venous vasodilatation induced by acetylcholine after control sleep and partial sleep deprivation periods.
deprivation that was carefully monitored. We reasoned that the present protocol therefore mimics the effects of sleep curtailment frequently experienced in our society. Compared with an 8-h-night sleep average period, 4.5 h of sleep for 5 nights was apparently well tolerated and elicited no significant changes in resting HR and BP in young subjects. However, partial sleep deprivation caused significant changes in autonomic cardiovascular function characterized by increased sympathetic modulation estimated by increased LF component of HR and BP variability, as well as increased levels of plasma norepinephrine. In our study, plasma concentration of norepinephrine, commonly used as an index of systemic sympathetic activity (12, 13), also increased.

In the present study, resting HR and BP did not change significantly after partial sleep deprivation, suggesting that these variables were adequately buffered by the control system. On the other hand, sympathetic activity, estimated by HR variability, showed that partial sleep deprivation caused a clear shift towards sympathetic activity as evidenced by an increase in LF, decrease in HF, and increase in LF-to-HF ratio. These results suggest that these are early and subtle cardiovascular effects that would not be detected by routine clinical evaluation. Our results are in line with results of a previous study (26) that evaluated sleep curtailment in healthy subjects who participated in a strict protocol under laboratory conditions. We extend these findings by showing increased sympathetic activity in subjects submitted to a relatively mild regimen of sleep restriction engaged in normal activities for only 5 nights. We also show an increased resting BP variability at LF frequency, further suggesting increased sympathetic activity.

In our protocol, the LF and HF HR variability components were higher and lower, respectively, after sleep deprivation, indicating a shift toward sympathetic cardiac modulation as expressed by an increase in sympathetic-vagal balance. Likewise, we also showed an increase in LF component of BP variability, further suggesting increased systemic sympathetic modulation to the vessels. Similar observation was obtained in previous studies, when subjects were not recumbent during sleep deprivation period (26, 29, 34). Emotional stress, cognitive, and physical work, among other variables, separately or together may influence hormones and catecholamine that act on BP regulation (24). This knowledge highlights the importance of maintaining a similar environmental condition and activities undertaken when comparing BP and sympathetic control in different periods of time. The present protocol therefore mimics the effects of sleep curtailment frequently experienced in our society, since the volunteers were engaged in their normal routine in a relatively mild model of sleep deprivation.

Acute sleep deprivation is able to impair arterial endothelial function in cardiologists after 24 h on call (11). However, in these studies, the participants were potentially under significant physical and mental stress, which are well-established causes of endothelial dysfunction (11). Our study is, therefore, the first to analyze the effects of partial sleep deprivation on venous endothelial function in young subjects engaged in their routine activities. The endothelium is involved in a number of pathological conditions, and different cardiovascular risk factors are associated with impairment of arterial and venous endothelium-dependent dilation (33, 22, 25, 32). Therefore, our study showed that partial sleep deprivation is also able to promote vein endothelial dysfunction. The mechanisms associated with endothelial dysfunction may be explained by increase in sympathetic activation as well as other factors such as a decrease in bioavailability of nitric oxide (9, 18, 33). Endothelial cells have high functional demands and are highly sensitive to environment changes, but differ in morphology and function depending on the vascular tree (6). Therefore, the functional impact of arterial dysfunction associated with partial sleep deprivation, as opposed to the venous dysfunction, needs to be addressed in further studies.

Our manuscript was designed to mimic “real world” sleep deprivation and has several limitations. First, several potential confounding factors, including stress, food intake, and environmental stimuli, were not monitored. Second, the study was not designed to address whether 2 days of washout with ad libitum sleep are sufficient to return the studied cardiovascular parameters to baseline. Finally, since our protocol was restricted to males, these results may differ in females.

In conclusion, our study may help to increase awareness of the potential harmful effects of apparently benign self-imposed sleep deprivation commonly experienced during a typical 5-day work week. Sleep curtailment may be a major public health problem in our modern society. Our study shows that sleep curtailment for 5 nights is sufficient to cause a significant increase in global sympathetic activity and also endothelial dysfunction.

ACKNOWLEDGMENTS
This study is registered as ClinicalTrials.gov No. NCT00669513.

DISCLOSURES
No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS

Fig. 3. Venodilation response of acetylcholine dose-response curve after control and partial sleep deprivation periods. *P < 0.05 vs. control sleep.
REFERENCES