Effects of Statin on Arrhythmia and Heart Rate Variability in Healthy Persons With 48-Hour Sleep Deprivation

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Background—It has been reported that sleep deprivation is associated with cardiac autonomic disorder, inflammation, and oxidative stress. Statins have significant cardiovascular protective effects in patients with cardiovascular disease. This study aimed to investigate the protective effect of statins on arrhythmia and heart rate variability in young healthy persons after 48-hour sleep deprivation.

Methods and Results—This study enrolled 72 young healthy participants aged 26.5±3.5 years. All participants received 48-hour continuous ambulatory electrocardiogram monitoring. Arrhythmia, time, and frequency domain parameters were analyzed for all participants. The primary end point, low/high frequency ratio, was significantly lower in the statin group than in the control group (2.48±1.12 versus 3.02±1.23, $P<0.001$). After 48-hour sleep deprivation, low frequency—the frequency of premature atrial complexes and premature ventricular complexes—was significantly decreased in the statin group compared with the control group ($P<0.05$). There was also a significant increase in high frequency in the statin group compared with the control group ($P<0.05$). There was a significant decrease in serum high-sensitivity C-reactive protein and malondialdehyde levels after 48-hour sleep deprivation in the statin group compared with the control group ($P<0.05$).

Conclusions—Statin use might be associated with improvement in arrhythmia and heart rate variability in healthy persons with 48-hour sleep deprivation. This finding should be confirmed by larger scale trials.

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Key Words: arrhythmia • heart rate variability • sleep deprivation • sleep disorders • statin

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During earthquakes, floods, or fire disasters, rescue workers usually perform their work without sleep. Sleep deprivation, which is a strong stressor, can exert a large effect on the cardiovascular system of rescue workers. It has been reported that sleep deprivation is associated with cardiac autonomic disorder, inflammation, and oxidative stress.[1–4] Heart rate variability (HRV) is acknowledged as a reliable marker of cardiac autonomic control, and the frequency of premature ventricular complexes (PVCs) can be an indicator of arrhythmogenicity. Our previous study

found that HRV was significantly decreased after 24-hour sleep deprivation. Metoprolol could improve HRV and reduce the frequency of premature atrial complexes (PACs) and PVCs,[2] but sleepiness and hypotension occurred frequently in participants treated with metoprolol. Statins have significant cardiovascular protective effects in patients with cardiovascular disease.[5] Statin use could not only regulate the autonomic nervous system[6] but also have antioxidant and anti-inflammatory properties[7]; however, the effects of statins on sleep deprivation remain unclear. This study aimed to investigate the effects of statin on HRV and arrhythmia after administration prophylactically before 48-hour sleep deprivation.

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Methods

Study Site and Ethics

This single-center, randomized, double-blind, placebo-controlled trial was performed at the Chinese PLA General Hospital in Beijing, People's Republic of China. The study was approved by the Beijing Ethics Association and the ethics committee of the Chinese PLA General Hospital and complied

with the Declaration of Helsinki. All participants provided written informed consent to participate in the study. The trial was registered at ClinicalTrials.gov (identifier NCT02496962).

Study Population

Participants of both sexes aged 18 to 30 years were recruited from the army by advertisement. The study enrolled 72 participants (15 women and 57 men) aged 26.5±3.4 years. All participants gave written informed consent in accordance with the PLA General Hospital before taking part in this study. All participants received financial compensation for participation. This study was carried out in the clinical investigation unit of the PLA General Hospital. Participants were free of any medical conditions (eg, hypertension, diabetes mellitus, and hyperthyroidism) and medication known to affect cardiovascular, metabolic, gastrointestinal, or immune function (including over-the-counter medication). Participants with sleep, depression, or anxiety disorders were excluded from the study based on self-reported prestudy questionnaires and written confirmation from the participant's general practitioner. All participants were nonsmokers at the time of the study and did not consume alcohol.

Protocol

No caffeine or alcohol was allowed during the 48 hours preceding the laboratory studies to the completion of the study. Enrolled participants reported to the sleep laboratory at 7 AM after obtaining their normal sleep at home the previous night. Each participant remained awake in the sleep laboratory from 7 AM on day 1 to 7 AM on day 3. All participants had a designated bedroom for the entire study. All physiological measurements were performed in the bedroom with a temperature of 22°C and illumination of 100 lux during the study. Participants were continuously monitored by video camera. Those who displayed sleep onset were immediately aroused and kept awake by verbal encouragement. Caloric and fluid management was individualized according to estimated daily needs; however, snacks were permitted to be eaten in the laboratory. Participants were permitted to read; watch video movies on a DVD player; play video games; do job-related work, including using the computer and the Internet; and converse with the staff or visitors.

Participants were randomized using a computer-generated sequence to either placebo or statin at a 1:1 ratio. Investigators, participants, and other study personnel were blinded to the assigned treatment for the duration of the study. Participants underwent the following 2 stages in the laboratory: normal sleep and statin or placebo treatment before

sleep deprivation (1 week later). At the first stage (normal sleep), 48-hour continuous ambulatory electrocardiogram (48-hour Holter) monitoring was applied to all participants. At the second stage, participants accepted statin or placebo administration 3 days prior to sleep deprivation and then underwent sleep deprivation, which was scheduled 1 week after the first stage. The 24-hour data were expressed as the mean values of parameters collected over the entire 24 hours, and the 48-hour data were calculated between 24 and 48 hours. Patients in the statin group were given a supply of 20 mg atorvastatin (Pfizer) to be taken daily, whereas patients in the control group were given a placebo (Pfizer). Study treatment commenced 3 days before sleep deprivation and continued for 2 days during sleep deprivation. Good Clinical Practice training was required for all personnel involved in the trial.

Study Outcomes

The primary efficacy end point was the effect of statin on the change in the ratio of low frequency (LF) to high frequency (HF) with 48-hour sleep deprivation compared with the baseline value. Secondary efficacy variables were the frequency of PACs and PVCs, the standard deviation of N–N intervals (SDNN), and levels of total cholesterol, triglyceride, high-sensitivity C-reactive protein (hsCRP), interleukin 6, superoxide dismutase, and malondialdehyde (MDA; an indicator of oxidative stress).

Laboratory Tests

The laboratory data (eg, total cholesterol, hsCRP, superoxide dismutase) were obtained at baseline (before intervention), at 24-hour sleep deprivation, and at 48-hour sleep deprivation. Total cholesterol (coefficient of variation [CV] 2.3%, normal range <200 mg/dL) was determined by the cholesterol esterase method.[8] Triglycerides were determined by enzyme colorimetry (CV 3.0%, normal range <150 mg/dL).[9] Levels of hsCRP were measured using a sandwich enzyme-linked immunosorbent assay (CV 2.0%, normal range <0.8 mg/dL; R&D Systems Inc). Serum interleukin 6 concentrations were measured using an enzyme-linked immunosorbent assay (CV 2.8%, normal range <8 pg/mL; R&D Systems Inc). Superoxide dismutase activity was estimated as the inhibition of a colorimetric reaction using an assay kit (CV 3.3%, normal range 129–216 U/mL; Cayman Chemicals). Serum MDA levels were measured using a thiobarbituric acid–reactive substance method.[10] The pink adduct formed by samples was extracted in *n*-butanol. Each sample was placed in a 96-well plate and read at 535 nm in a microplate spectrophotometer reader (CV 4.1%, normal range 3.46–4.66 nmol/mL; Benchmark Plus, Bio-Rad Laboratories).

Heart Rate Variability

All participants received Holter monitoring using a 12-channel ambulatory electrocardiogram recorder (pace recorder model MIC-12H, Beijing Jinco Medical Co., Ltd) with a sampling rate of 250 Hz (4 ms). The P waves and QRS complexes were automatically classified and manually verified as normal sinus rhythm, PACs or PVCs, or noise by comparison with adjacent waves. The R-R intervals were deduced from the adjacent normal sinus beats (ie, N-N intervals). For the entire study population, time domain measurements, including mean N-N intervals, SDNN, and root mean square of successive differences, were calculated automatically every 5 minutes. The power spectrum densities were estimated by Welch's averaged periodogram method, whereas very LF power (0.01–0.05 Hz), LF power (0.05–0.15 Hz), and HF power (0.15–0.5 Hz) were derived for each 5-min segment. HF power is considered a function of cardiac parasympathetic nervous system activity to the heart.[11] LF, although not modulated by a single arm of the autonomic nervous system,[12] is considered to be normalized for total power as a representative index of sympathetic activity to the heart.[13–15]

The LF/HF ratio is considered an index of the balance of sympathovagal input to sinoatrial node activity and has been found to be representative of sympathetic–parasympathetic balance in both physiological and pathophysiological conditions.[15–17]

Reproducibility

To determine the reproducibility of the Holter parameters, 20 randomly selected participants were analyzed by 2 independent blinded observers, as described. The correlation coefficients of interobserver variability for LF/HF ratio, PACs, and PVCs were 0.92, 0.93, and 0.94, respectively. The correlation coefficients of intraobserver variability for LF/HF ratio, PACs, and PVCs were 0.90, 0.96, and 0.91, respectively.

Statistical Analysis

In our preliminary study, a difference in LF/HF ratio was detected between the statin and control groups after 48-hour sleep deprivation (0.5 ± 0.6, n=26). With a significance level of 5% and 90% power to detect a 0.5 difference in LF/HF ratio (SD 0.6), 36 patients were needed in each of the 2 groups.

Data are expressed as mean±SD or median (range 25–75%). After testing data for normality, we used an independent *t* test or the Mann–Whitney *U* test to compare values between the statin group and the control group. The analyses were conducted on an intention-to-treat basis with significance levels set at *P*<0.05. Missing values were replaced by the last observed value of that variable. Statistical analyses were performed using SPSS software version 18.0 (IBM Corp).

Multivariate analysis was performed to investigate the relationship between change in LF/HF ratio and change in serum hsCRP level (or MDA level) after adjustment for age, sex, body mass index, normal duration of sleeping time, hemoglobin, creatinine, PACs, PVCs, SDNN, total cholesterol, triglyceride, interleukin 6, superoxide dismutase. Given 8 secondary outcomes, we used a Bonferroni-adjusted significance level of 0.05/8=0.00625.

Results

Study Population

The characteristics of the study population are given in Table 1 and Figure. The body mass index (kg/m^2) of the participants ranged from 22 to 23.1. The participants usually needed 7 to 10 hours of sleep daily. There were no significant differences between the 2 groups. No episodes of abnormal liver function, renal insufficiency, or any other adverse effect were reported.

HRV and Arrhythmia

After 48-hour sleep deprivation, LF and the LF/HF ratio of HRV were significantly decreased in the statin group compared with the control group (*P*<0.05). There was also a significant increase in HF and SDNN of HRV in the statin group compared with the control group (*P*<0.05). The participants experienced frequent PACs and PVCs after 48-hour sleep deprivation in the control group. The frequency of PACs and PVCs was reduced with statin treatment compared with the control group (Table 2).

Table 1. Baseline Characteristics of the 72 Participants

Characteristics	Statin Group (n=36)	Control Group (n=36)
Age, y	27±3.3	26±3.5
Male	28 (78)	29 (81)
Height, m	1.72±0.04	1.69±0.03
Weight, kg	65±3	64±3
BMI, kg/m^2	22.4±0.3	22.5±0.4
Normal duration of sleeping time, h	7.4±0.8	7.6±0.9
Hemoglobin, g/dL	13.1±1.6	13.6±1.4
Creatinine, mg/dL	0.9±0.2	0.9±0.1

Data are presented as mean±SD or number (%) of patients. BMI indicates body mass index.
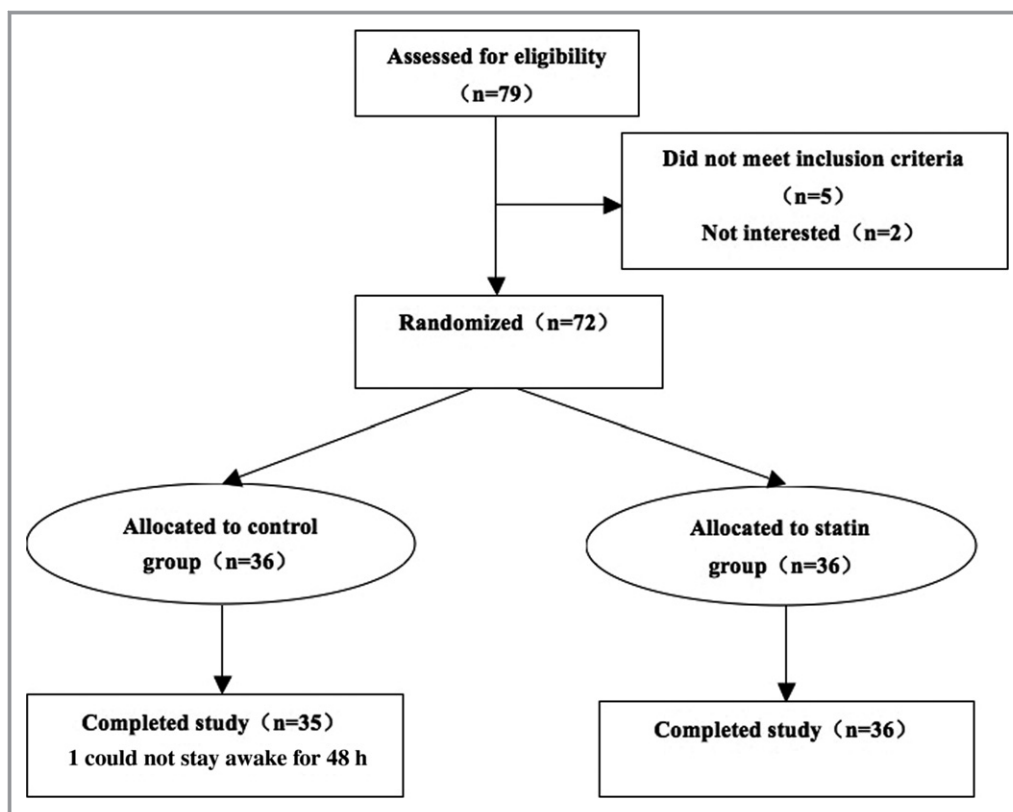
Figure. Participant flow chart.

Levels of hsCRP, Interleukin 6, Superoxide Dismutase, and MDA

The mean reductions in serum hsCRP levels were significantly greater in the statin group than in the control group (Table 2). The difference in the decrease in serum hsCRP levels was −0.11 mg/dL (95% CI −0.18 to −0.05; *P*<0.001). The difference in the decrease in serum MDA levels was −2.51 nmol/mL (95% CI −4.02 to −1.86; *P*<0.001). Change in LF/HF ratio correlated with change in serum hsCRP level (*r*=0.11, *P*=0.02) and MDA level (*r*=0.08, *P*=0.03) in adjusted analyses between the 2 groups.

When age, sex, body mass index, normal duration of sleeping time, hemoglobin, creatinine, total cholesterol, low-/ high-density lipoprotein ratio, hsCRP, MDA and use of statin were considered as explanatory variables and improvement in the LF/HF ratio was set as a dependent variable, administration of statin was consistently identified as a significant determinant for the improvement in LF/HF ratio, using a multivariate regression analysis (*P*=0.009) (Table 3).

Discussion

We observed significant salutary effects of statins on LF/HF in participants with 48-hour sleep deprivation. In addition,

statins elicited favorable changes in markers of inflammation and oxidative stress.

Spectral analysis techniques have been used to determine changes in central nervous system activity. Power in specific frequency bands can be related to parasympathetic and sympathetic nervous system activity. Specifically, relative power in HF areas, usually from 0.15 to 0.5 Hz, has been used to infer parasympathetic nervous system activity. A range of lower frequencies from 0.05 to 0.15 Hz has typically been related to a combination of parasympathetic and sympathetic influences.[18–20] Because LF power is a combination of sympathetic and parasympathetic effects, investigators frequently infer sympathetic nervous system activity from the ratio of low (parasympathetic and sympathetic) to high (predominantly parasympathetic) power so that parasympathetic power is extracted from the ratio to some extent,[19,21,22] providing a better indicator of sympathetic activity. SDNN and root mean square of successive differences are also considered representative indices of parasympathetic nervous system activity.[23] Acute sleep deprivation is associated with increased sympathetic activity and decreased parasympathetic modulation.[24] In addition, sleep disturbance may also result in sympathovagal imbalance[24,25] and an increase in PVCs.[25] Lower HF, SDNN, and root mean square of successive differences reflect lower parasympathetic activity, and

Table 2. Electrocardiogram and Laboratory Investigations of Patients in 2 Treatment Groups

Parameters	Statin Group (n=36)				Control Group (n=36)				P Value*
	Baseline	24 Hours	48 Hours	Change in Statin Group	Baseline	24 Hours	48 Hours	Change in Control Group	
Holter									
LF/HF ratio	1.66±0.95	2.03±1.04	2.48±1.12	0.86 (0.41–1.19)	1.60±0.91	2.31±1.09	3.02±1.23	1.45 (0.79–2.15)	<0.001
AHR, beats/min	67±6	69±7	75±9	8 (2–14)	66±6	70±7	77±10	10 (3–18)	0.19
SBP, mm Hg	120±10	122±10	125±13	6 (−2 to 16)	118±9	123±11	128±12	9 (−1 to 19)	0.21
DBP, mm Hg	70±6	73±7	77±8	7 (1–13)	69±5	74±6	79±8	9 (2–16)	0.20
PAC, beats/h	1±1	3±1	7±3	6 (3–9)	1±1	5±2	15±5	14 (7–20)	<0.001
PVC, beats/h	1±1	2±1	5±3	4 (1–7)	1±1	4±2	10±6	9 (3–15)	<0.001
HF, ms^2	771±305	675±269	607±253	−163 (−215 to −108)	782±310	630±258	549±243	−235 (−319 to −154)	<0.001
LF, ms^2	1284±503	1374±586	1506±601	216 (105–317)	1253±459	1458±595	1659±642	406 (218–592)	<0.001
VLF, ms^2	3401±515	3214±467	2957±453	−447 (−526 to −327)	3520±558	3058±410	2623±454	−890 (−1152 to −644)	<0.001
SDNN, ms	153 (115–219)	146 (92–161)	131 (101–191)	−22 (−30 to −13)	152 (99–211)	128 (85–165)	119 (82–151)	−34 (−60 to −15)	0.001
RMSSD, ms	51±25	47±29	44±24	−7 (−13 to −1)	53±24	45±25	39±23	−13 (−23 to −11)	0.003
Total cholesterol, mg/dL	162±22	153±21	141±24	−21 (−29 to −12)	164±23	176±24	188±26	23 (11–34)	<0.001
Triglyceride, mg/dL	123±20	116±19	111±18	−11 (−22 to −1)	122±18	128±20	129±22	6 (−3 to 16)	<0.001
LDL/HDL ratio	1.62±0.04	1.97±0.05	2.17±0.06	0.55 (0.51–0.59)	1.58±0.05	2.11±0.05	2.42±0.07	0.84 (0.79–0.90)	<0.001
hsCRP, mg/dL	0.42±0.12	0.45±0.11	0.52±0.13	0.12 (0.06–0.17)	0.41±0.11	0.51±0.13	0.66±0.16	0.24 (0.14–0.35)	<0.001
IL–6, pg/mL	4.2±1.3	4.9±1.6	5.9±1.9	1.9 (1.1–2.8)	4.1±1.1	5.3±1.8	6.5±2.1	2.2 (1.1–3.4)	0.19
SOD, U/mL	119±15	164±22	221±31	90 (67–119)	125±16	171±20	210±27	83 (69–98)	0.15
MDA, nmol/mL	5.07 (4.28–6.67)	6.12 (4.65–7.63)	7.82 (5.34–9.54)	2.65 (1.46–3.56)	5.13 (4.75–6.85)	7.08 (5.14–9.11)	10.11 (7.13–13.53)	5.05 (3.12–7.18)	<0.001

Data are presented as mean±SD or median (range 25–75%). AHR indicates average heart rate; DBP, diastolic blood pressure; HDL, high-density lipoprotein; HF, high frequency; hsCRP, high-sensitivity C-reactive protein; IL-6, interleukin 6; LDL, low-density lipoprotein; LF, low frequency; MDA, malondialdehyde; PAC, premature atrial complex; PVC, premature ventricular complex; RMSSD, root mean square successive differences; SBP, systolic blood pressure; SDNN, standard deviation of N–N intervals; SOD, superoxide dismutase; VLF, very low frequency.
*P<0.05, change in statin group after 48-hour sleep deprivation vs change in control group after 48-hour sleep deprivation.

Table 3. Independent Variables for Improvement in the LF/HF Ratio

Independent Variables	Regression Coefficient	Hazard Ratio (95% CI)	P Value
Age	−0.009	0.97 (0.91–1.12)	0.77
Sex	−0.041	0.85 (0.76–1.26)	0.65
Heart rate	−0.002	0.71 (0.61–1.17)	0.32
Systolic blood pressure	−0.001	0.94 (0.87–1.10)	0.92
BMI	−0.007	0.90 (0.72–1.46)	0.46
Administration of statin	1.387	1.32 (1.02–2.19)	0.009
Total cholesterol	−0.017	0.86 (0.78–0.96)	0.01
LDL/HDL ratio	−0.036	0.75 (0.69–0.85)	0.007
hsCRP	−0.103	0.89 (0.75–0.95)	0.02
MDA	−0.054	0.76 (0.70–0.84)	0.008

Model adjusted for age, sex, heart rate, systolic blood pressure, diastolic blood pressure, BMI, normal duration of sleeping time, hemoglobin, creatinine, total cholesterol, LDL/HDL ratio, hsCRP, MDA, and use of statin. BMI indicates body mass index; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; LDL, low-density lipoprotein; LF/HF, low frequency/high frequency; MDA, malondialdehyde.

higher LF and LF/HF ratios indicate higher sympathetic activity. All of these factors are associated with a higher risk of cardiovascular disease.[23,26] In our study, the LF/HF ratio was improved with statin treatment of participants with 48-hour sleep deprivation. The difference in means of 0.5 in the LF/HF ratio between the 2 groups was attained ($P<0.05$). To our knowledge, this study is the first randomized trial to assess the effects of statin on arrhythmia and HRV in healthy persons with 48-hour sleep deprivation. We also found that statin could reduce inflammation and oxidative stress.

Potential Mechanisms

It has been reported that sleep deprivation is associated with cardiac autonomic disorder, inflammation, and oxidative stress.[2–4] Statin was reported to ameliorate inflammation in previous studies.[27,28] In our study, we found that hsCRP levels were significantly lower in the statin group, thus it is possible that statin reduces inflammation and improves the LF/HF ratio. Statin can ameliorate oxidative stress.[29] In the present study, the MDA levels in the statin group were significantly lower than those in the control group; therefore, it is possible that an improvement in oxidative stress contributed to the decrease of the LF/HF ratio in the statin group. In addition, statin therapy is associated with a reduction in ventricular tachyarrhythmias and atrial fibrillation.[30] Statin treatment significantly reduced vulnerability to ventricular fibrillation via the mechanism of reduction of neural and electrophysiological remodeling.[31] The mechanism will be investigated in further studies.

Study Limitations

The main limitations of this study are that it is from a single center and that the sample size was small. Moreover, a crossover study (instead of the baseline data) should be done. This approach could strengthen the study design by decreasing interindividual variability. In addition, the continual information collected over the 48-hour period (eg, 1, 2, 3 hours) could be used in the study rather than just snapshots at 0, 24, and 48 hours. There is a need for large-scale and long-term research into this issue with analysis of more laboratory indicators.

Conclusion

Statin use might be associated with improvement in arrhythmia and HRV in healthy persons with 48-hour sleep deprivation. This finding should be confirmed by larger scale trials. It might suggest that patients who have sleep deprivation or disorders or who do shift work could be put on statins to reduce the risk of heart disease.

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Disclosures

None.

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