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Sympathetic Overactivity Based on Heart-Rate Variability in Patients with Obstructive Sleep Apnea and Cerebral Small-Vessel Disease

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Background and Purpose Obstructive sleep apnea (OSA) is associated with cerebral white-matter changes (WMC), but the underlying mechanisms are not completely understood. Our aim was to identify the cardiovascular autonomic characteristics during sleep that are associated with cerebral WMC in OSA patients.

Methods We recruited subjects from our sleep-center database who underwent both polysomnography and brain MRI within a 1-year period. Sixty patients who had OSA with WMC (OSA+WMC), 44 patients who had OSA without WMC (OSA−WMC), and 31 control subjects who had neither OSA nor WMC were analyzed. Linear and nonlinear indices of heart-rate variability (HRV) were analyzed in each group according to different sleep stages and also over the entire sleeping period.

Results Among the nonlinear HRV indices, the Poincaré ratio (SD12) during the entire sleep period was significantly increased in the OSA+WMC group, even after age adjustment. Meanwhile, detrended fluctuation analysis 1 during non-rapid-eye-movement sleep tended to be lowest in the OSA+WMC group. These indices were altered regardless of the presence of hypertension or diabetes. In the subgroup analysis of middle-aged OSA patients, approximate entropy during rapid-eye-movement sleep was significantly lower in OSA+WMC patients than in OSA−WMC patients. Overall, the nonlinear HRV indices suggest that sympathetic activity was higher in the OSA+WMC group than in the OSA−WMC and control groups.

Conclusions Our findings suggest that dysregulation of HRV, especially overactivation of sympathetic tone, could be a pathophysiologic mechanism underlying the development of WMC in OSA patients.

Key Words obstructive sleep apnea, white-matter changes, heart-rate variability, nonlinear indices, sympathetic overactivation.

INTRODUCTION

Obstructive sleep apnea (OSA) is considered to be a significant risk factor for cardiovascular morbidity[1,2]; the main pathophysiologic mechanisms include cardiovascular autonomic dysfunction, which is predominantly represented by sympathetic overactivation.[3,4] There is growing evidence that OSA is associated with cerebral small-vessel diseases,[5,6] including cerebral white-matter changes (WMC),[7–9] which are associated with an increased risk of cerebrovascular events and vascular cognitive impairments.[10] However, the mechanisms underlying cerebral WMC in patients with OSA are not completely understood.

Heart-rate variability (HRV) has been widely analyzed as a noninvasive method for evaluating cardiovascular autonomic conditions in various conditions.[11] Among the frequency-

domain HRV indices, the low-frequency (LF) component is considered to represent both sympathetic and parasympathetic activity, while the high-frequency (HF) component is accepted as a marker of parasympathetic activity. The LF/HF ratio is considered to reflect sympathovagal balance, with a higher LF/HF indicating a predominance of sympathetic activity and a lower LF/HF suggesting parasympathetic predominance.[12] It is well document that sympathetic activity during rapid-eye-movement (REM) sleep is increased during physiologic sleep,[13] and excessive sympathetic activity during both wakefulness and sleep has been reported in OSA patients based on HRV studies.[14] It has recently been demonstrated that morbidity and mortality can be predicted more accurately using several nonlinear HRV indices than standard linear parameters in certain diseases, including cardiovascular disease.[15,16]

We previously reported a significant association between moderate-to-severe OSA and indicators of WMC, including an increased burden of white-matter hyperintensities, asymptomatic lacunar infarctions, cerebral microbleeds, or perivascular spaces.[6] Although WMC are suggested to be associated with increased sympathetic activity, and HRV analyses are useful for monitoring sympathetic activities during sleep stages, HRV analyses in association with WMC during sleep—including in OSA patients—are lacking.

In the current study we assessed linear and nonlinear HRV indices according to different sleep stages in order to identify the cardiovascular autonomic characteristics during sleep that are associated with cerebral WMC in OSA.

METHODS

Patients

In total, 135 subjects were analyzed in this study (Fig. 1). Among the patients who visited the Sleep Center of Ewha University Medical Center and underwent overnight polysomnography (PSG) between April 2005 and May 2014, we selected 185 patients in whom 1.5- or 3.0-T brain FLAIR and T2-weighted MRI images were obtained within 1 year before or after PSG. The indication for performing PSG was the pres-
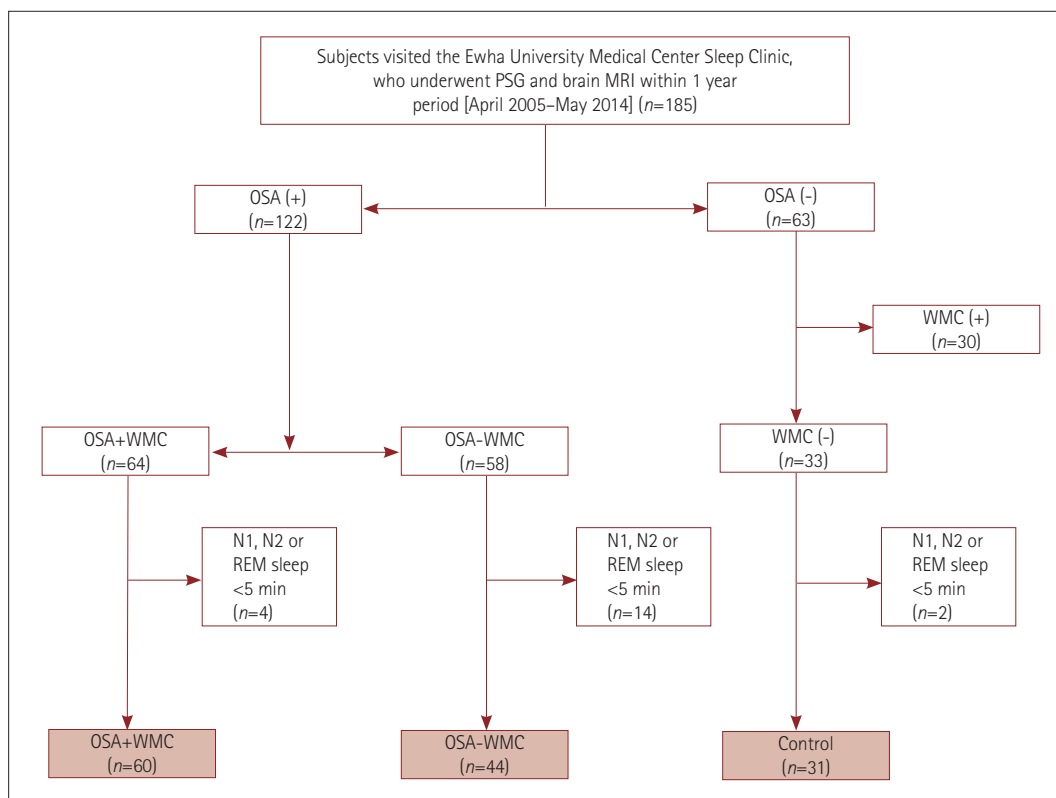

Fig. 1. Flow chart for the selection of the study subjects. Among the patients who visited the Sleep Center of Ewha University Medical Center and underwent overnight PSG between April 2005 and May 2014, 185 patients were selected who underwent brain MRI within 1 year before or after PSG. Sixty-four of the 122 OSA patients had WMC. Thirty-three of the subjects without OSA did not have WMC. Subjects were excluded from the heart-rate variability analysis if the artifact-free and arousal-free periods of their N1, N2, or REM sleep periods were shorter than 5 min on PSG. Finally 135 subjects were included in the current study: 60 were assigned to the OSA+WMC group, 44 to the OSA–WMC, and 31 to the control group. OSA: obstructive sleep apnea, OSA+WMC: obstructive sleep apnea with white-matter changes, OSA-WMC: obstructive sleep apnea without white-matter changes, PSG: polysomnography, REM: rapid-eye-movement, WMC: white-matter changes.

ence of at least one OSA-related symptom (e.g., witnessed loud snoring between apnea episodes, episodes of gasping for air, or choking), sleep fragmentation/insomnia, nonrefreshing sleep, and being in the high-risk category for OSA according to the Berlin Questionnaire, as described previously.[6,17,18] Brain MRI was routinely recommended in our sleep center for evaluating underlying organic brain lesions.

Based on the obtained PSG results, 122 of the 185 patients were diagnosed with OSA, of which 64 had WMC on brain MRI. Thirty of the patients without OSA who had WMC were excluded since they were considered not appropriate for inclusion in a control group when investigating the impact of OSA on the development of WMC. Subjects were excluded from the HRV analysis if the artifact-free and arousal-free periods of their N1, N2, or REM sleep were less than 5 min on PSG. Finally, 60 patients were assigned to the OSA with WMC (OSA+WMC) group, 44 to the OSA without WMC (OSA–WMC) group, and 31 subjects to the control group. This study was approved by the Institutional Review Board of our hospital (IRB No. 11-21-016). The need to obtained informed consent from the patients was waived due to the retrospective design and observational nature of the study.

MRI protocol and assessment of white-matter changes

The brain MRI protocol applied in this study has been described in detail previously.[6,19] Brain MRI image slices were acquired using the following parameters: for FLAIR, repetition time (TR)/echo time (TE)=12,000 ms/120 ms, pixel spacing=0.449 mm/0.449 mm, field of view (FOV)=183×230 mm^2, and slice thickness=5 mm; for T2-weighted images, TR/TE=15,000 ms/90 ms, pixel spacing=0.240 mm/0.240 mm, FOV=176×220 mm^2, and slice thickness=5 mm. The extent of WMC was determined on brain FLAIR and T2-weighted MRI images according to the modified Fazekas scoring system[20]: a Fazekas score of ≥1 was considered to indicate WMC, while score of 0 indicated the absence of WMC. The presence of WMC was independently investigated by two neurologists (T.J.S. and J.H.P.) while they were blinded to clinical information. Consensus was reached in cases of discrepancy regarding the presence of WMC.

Polysomnography

PSG was performed in accordance with a previously described protocol.[6,18] Overnight PSG was performed with a comprehensive device (TWin® PSG Clinical Software, Grass Technologies, West Warwick, RI, USA) at the Sleep Center of Ewha University Medical Center. The electrocardiogram (ECG) was recorded at a sampling rate of 256 Hz. The PSG data were initially scored by an experienced sleep technician and then reviewed thoroughly by expert sleep physicians (H.W.L. and J.H.P.) following the 2014 American Academy of Sleep Medicine manual for sleep scoring and respiratory event criteria.[21]

Briefly, apnea episodes were defined when the airflow was reduced to ≥90% of the baseline values for at least 10 sec, and they were further classified as either the obstructive type if respiratory effort was noted on either the chest or abdominal belt channel, or the central type if no respiratory effort was noted. Hypopnea episodes were defined as a ≥30% reduction of airflow for at least 10 sec and accompanied by a decrease of ≥3% in oxygen saturation (SaO$_2$) or arousal. The apnea-hypopnea index (AHI) was calculated by averaging the total number of obstructive apnea episodes and hypopnea episodes per hour of sleep, with OSA being diagnosed when AHI ≥5.

Assessment of heart-rate variability

For each patient we carefully selected artifact-free, apnea-free, awakening-free, 5-min ECG samples from each sleep stage: N1, N2, REM sleep, and wakefulness. ECG samples were selected from the earliest period within a particular sleep stage, and for the wakefulness period we analyzed a 5-min segment at the beginning of the PSG recording that was free of ectopic beats and artifacts. For the HRV analysis according to different sleep stages, one 5-min segment was analyzed for each sleep stage, to give a total of three 5-min segments for each patient. The HRV analysis during the entire sleep period utilized the entire ECG data set recorded between the onset and the end of sleep on PSG.

The ECG data were analyzed using Kubios HRV software.[12] The RR variability was analyzed in both the time and frequency domains. Each RR interval was calculated using Welch's periodogram method. Mean RR intervals, the standard-deviation of normal to normal RR intervals (SDNN), and the root mean square of successive differences (RMSSD) were acquired as time-domain indices. Spectral components of HRV were classified as very low frequency (VLF, <0.04 Hz), LF (0.04−0.15 Hz), and HF (0.15−0.4 Hz). LF- and HF-variability components were calculated in both power and normalized units (LFnu and HFnu, respectively). LFnu and HFnu were calculated as LFnu=LF/(total power−VLF)×100 and HFnu=HF/(total power−VLF)×100.

The nonlinear properties of HRV were analyzed using Poincaré plots, detrended fluctuation analysis (DFA), approximate entropy (ApEn), and sample entropy (SampEn). A Poincaré plot is a graphical presentation of the correlation between consecutive RR intervals, and the standard-deviations of the points perpendicular to and along the line of identity (the line where RRj=RRj+1) are denoted by SD1 and SD2,

respectively. SD1 represents the short-term beat-to-beat variability, and SD2 measures the long-term variability.[12] SD12 represents the relationship between these components, which is the ratio of short-term variation to long-term variation (i.e., SD1/SD2).[15,22] DFA measures correlations within the data over different time scales.[23] DFA1 (α1) and DFA2 (α2) are obtained from the plot by default within ranges of 4−16 beats and 16−64 beats, which represent short-term and long-term fluctuations, respectively. For the entropy-derived measures of ApEn and SampEn, lower values of entropy reflect data that are more predictable, while more-random and complex data are described by higher values of entropy.[24,25]

Statistical analysis

Clinical variables were compared among the groups using the chi-square test for categorical variables and analysis of variance (ANOVA) for continuous variables. The Tukey honestly significant difference (HSD) test was used for post-hoc analysis of ANOVA. HRV indices were compared among the groups using ANOVA with the post-hoc Tukey HSD test for three-group comparisons, and the independent t-test for comparisons of control and combined OSA groups. Analysis of covariance (ANCOVA) was performed to control for the effect of age on HRV, and the Bonferroni test was applied for post-hoc comparisons. In the subgroup analysis of OSA patients selected by specific age ranges, the OSA+WMC and OSA−WMC groups did not conform to a normal distribution, and so the Mann-Whitney test was used to compare the mean values. The statistical analyses were conducted using SPSS software version 13.0 (SPSS Inc., Chicago, IL, USA),

Table 1. Demographic characteristics and PSG findings in control subjects, patients with OSA−WMC, and patients with OSA+WMC

	Control (*n*=31)	OSA−WMC (*n*=44)	OSA+WMC (*n*=60)	*p*
Demographic characteristic				
Age (years)	47.8±10.7	49.1±10.2	66.8±9.6	<0.001*†
Sex (male)	14 (45.2)	36 (81.8)	32 (53.3)	0.002
BMI (kg/m²)	24.0±3.3	25.9±3.2	25.3±3.2	0.047*‡
Neck circumference (cm)	36.6±3.3	39.1±3.3	37.3±3.9	0.009*‡
HTN (yes)	7 (22.6)	10 (22.7)	39 (65)	<0.001
Diabetes (yes)	2 (6.5)	4 (9.1)	17 (28.3)	0.007
Hyperlipidemia (yes)	2 (6.5)	5 (11.4)	8 (13.3)	0.611
Alcohol consumption (yes)	12 (38.7)	18 (40.9)	19 (31.7)	0.676
Current smoker (yes)	9 (29.0)	10 (22.7)	9 (15.0)	0.417
ESS	7.1±5.4	9.0±5.6	6.8±4.7	0.083
BDI	13.2±7.5	12.8±10.5	13.7±12.7	0.916
PSG findings				
Total sleep time (min)	368.0±108.9	367.8±78.2	337.6±91.3	0.166
Sleep latency (min)	18.4±26.6	7.6±10.7	16.8±28.6	0.084
AI (n/h)	13.4±6.6	26.1±16.3	28.1±14.9	<0.001*†
RAI (n/h)	0.2±0.4	2.6±7.0	5.0±10.8	0.032*
AHI (n/h)	1.4±1.3	23.7±19.8	31.8±20.2	<0.001*†
REM AHI (n/h)	3.8±6.8	23.8±17.2	31.9±21.8	<0.001*†
NREM AHI (n/h)	1.1±1.6	23.4±21.6	32.0±21.3	<0.001*†
RDI (n/h)	3.7±13.5	27.2±22.4	31.8±19.9	<0.001*†
Minimum SaO₂ (%)	86.3±10.8	82.0±7.3	80.1±9.4	0.011*
N1 sleep (%)	22.4±11.1	28.0±16.0	32.7±16.0	0.009*
N2 sleep (%)	51.8±17.8	47.5±13.1	45.3±16.5	0.185
N3 sleep (%)	7.1±8.2	7.3±8.2	6.0±7.6	0.678
REM sleep (%)	18.0±12.0	15.6±7.0	14.2±7.0	0.129

p-values were calculated using the chi-square test for categorical variables and analysis of variance for continuous variables. Data are *n* (%) or mean± standard-deviation values.

*$p<0.05$ between control and OSA+WMC in post-hoc Tukey test, †$p<0.05$ between OSA−WMC and OSA+WMC in post-hoc Turkey test, ‡$p<0.05$ between control and OSA−WMC.

AHI: apnea-hypopnea index, AI: arousal index, BDI: Beck Depression Inventory, BMI: body mass index, ESS: excessive daytime sleepiness, HTN: hypertension, n/h: number per hour, NREM: non-rapid-eye-movement, OSA+WMC: obstructive sleep apnea with white-matter changes, OSA-WMC: obstructive sleep apnea without white-matter changes, PSG: polysomnography, RAI: respiratory arousal index, RDI: respiratory disturbance index, REM: rapid-eye-movement, SaO₂: oxygen saturation.

and $p<0.05$ was considered statistically significant.

RESULTS

Demographic characteristics and polysomnography findings

Age, sex, and body mass index (BMI) differed among the three groups (Table 1). Patients were significantly older in the OSA+WMC group (age 66.8±9.6 years, mean±standard deviation) than in the OSA–WMC group (age 49.1±10.2 years) and the control group (age 47.8±10.7 years). BMI was significantly lower in the control group than in either of the OSA groups. The incidence of hypertension and diabetes differed across the groups, being highest in the OSA+WMC group.

Sleep characteristics including daytime sleepiness, total sleep time, sleep latency, and percentages of time in the REM, N2, and N3 sleep periods did not differ significantly between the groups, with the exception of the percentage of N1 sleep being higher in the OSA+WMC group than in the control group (Table 1). The arousal index, AHI, REM AHI, non-REM (NREM) AHI, and respiratory disturbance index were significantly higher in both the OSA+ WMC and OSA–WMC groups than in the control group, as expected. The minimum SaO_2 differed significantly among the groups, with the minimum SaO_2 being markedly lower in the OSA+WMC group than in the control group.

Heart-rate variability findings

Several indices in the time-domain and frequency-domain analyses of HRV differed significantly among the three groups (Table 2). Since most of the HRV parameters—including the frequency-domain indices and nonlinear indices—are significantly influenced by increasing age,[26,27] and patients in the OSA+WMC group were significantly older than those in the other groups, we performed ANCOVA to control for the effect of age on HRV parameters. Applying this age adjustment resulted in the differences in the time-domain and frequency-domain indices no longer being significant.

In terms of nonlinear HRV indices, SD12 was significantly higher and DFA1 was markedly lower in the OSA+WMC group than in the OSA–WMC and control groups during NREM sleep, REM sleep, and the entire sleep period (Table 2). ApEn was significantly lower in the OSA+WMC group during REM sleep.

When ANCOVA was performed with age controlled as a covariate, SD12 during the entire sleep period was the only HRV parameter that differed significantly among the three groups ($p=0.045$). However, DFA1 tended to be lowest in the OSA+WMC group during NREM sleep ($p=0.051$) (Table 2).

Subgroup analysis in subjects without hypertension or diabetes

In order to minimize the effect of confounding factors on HRV indices, we performed a subgroup analysis in subjects who did not have hypertension or diabetes, comprising 24, 33, and 18 subjects in the control, OSA–WMC, and OSA+WMC groups, respectively. ANCOVA was performed with age controlled as a covariate. SD12 during the entire sleep period differed significantly among the groups, being 0.41±0.13, 0.50± 0.16, and 0.60±0.21 in the control, OSA–WMC, and OSA+WMC groups, respectively ($p=0.028$). DFA1 during NREM sleep was also significantly different among the groups, being 1.15± 0.37, 0.97±0.28, and 0.86±0.36 in the control, OSA–WMC, and OSA+WMC groups, respectively ($p=0.025$) (Table 2).

Subgroup analysis in selected age groups

Since more than two-thirds of the general population older than 65 years have WMC,[28] we selected middle-aged patients (age 45−64 years) for inclusion in an additional subgroup analysis, involving 18 patients in each of the OSA–WMC and OSA+WMC groups. The mean AHI was higher than 20 in both groups, which indicates that most of these patients had moderate-to-severe OSA. The demographic characteristics and sleep study parameters did not differ significantly between these two groups (Table 3).

In the HRV analysis, none of the frequency-domain indices differed between the OSA–WMC and OSA+WMC groups (data not shown). Regarding the nonlinear indices, ApEn during REM sleep was significantly lower in the OSA+WMC group (0.79±0.25) than in the OSA–WMC group (0.94± 0.23) ($p=0.031$) (Fig. 2A and Table 3). Receiver operating characteristics analysis was performed to assess the effectiveness of ApEn for differentiating OSA+WMC from OSA–WMC. The value of 0.71 for the area under the curve indicated a diagnosis of OSA+WMC. When ApEn was lower than 0.85, the sensitivity, specificity, positive predictive value, and negative predictive value were 55.6, 77.8, 71.4, and 63.6%, respectively (Fig. 2B). None of the other nonlinear indices differed significantly between the two groups (data not shown).

DISCUSSION

With the aim of identifying the cardiovascular autonomic characteristics associated with cerebral WMC in OSA patients, we performed HRV analysis based on sleep stages obtained from PSG in a large number of OSA patients and control subjects. The frequency-domain indices did not exhibit any meaningful differences among the study groups. The nonlinear index of SD12 during the entire sleep period was increased in the OSA+WMC group, which was the only signif-

Table 2. Analysis of heart-rate variability in control, OSA–WMC, and OSA+WMC groups (selected indices with significant group differences)

	Control (n=31)	OSA–WMC (n=44)	OSA+WMC (n=60)	$p^{\dagger}$ (ANOVA)	$p^{\S}$ (ANCOVA)	Subgroup analysis (no HTN or diabetes)$^{\parallel}$ $p^{\S}$ (ANCOVA)
Time-domain indices (ms)						
SDNN						
NREM sleep (N1–N2)	43.4±23.5	60.7±52.5	85.6±104.8	0.038*	0.203	0.130
RMSSD						
NREM sleep (N1–N2)	37.6±29.9	61.3±68.1	105.2±134.3	0.005*	0.177	0.117
Frequency-domain indices						
HF power (ms²)						
NREM sleep (N1–N2)	534.8±713.7	2,442.8±9,075.4	10,365.9±22,009.0	0.006*†	0.090	0.181
LFnu						
Awake	45.3±17.3	50.1±17.9	40.0±17.1	0.016†	0.271	0.629
NREM sleep (N1–N2)	51.9±24.9	50.4±20.4	38.8±21.5	0.007*†	0.065	0.157
HFnu						
Awake	54.4±17.1	49.7±17.9	59.7±16.9	0.017†	0.275	0.635
NREM sleep (N1–N2)	48.0±24.8	49.5±20.3	60.9±21.4	0.007*†	0.064	0.160
Nonlinear indices (ms)						
Poincaré plot (SD1)						
NREM sleep (N1–N2)	26.6±21.2	43.4±48.3	74.5±95.2	0.005*	0.177	0.117
Poincaré ratio (SD12)						
NREM sleep (N1–N2)	0.52±0.32	0.57±0.25	0.72±0.30	0.003*†	0.167	0.140
REM sleep	0.36±0.27	0.44±0.28	0.61±0.37	0.002*†	0.305	0.479
Entire sleep period	0.44±0.18	0.51±0.16	0.60±0.19	<0.001*†	0.045	0.028
Detrended fluctuation analysis 1						
NREM sleep (N1–N2)	1.07±3.39	0.93±0.31	0.81±0.33	0.003*	0.051	0.025
REM sleep	1.17±0.34	1.06±0.31	0.90±0.37	0.002*	0.238	0.352
Entire sleep period	0.93±0.17	0.87±0.14	0.80±0.18	0.002*	0.308	0.129
ApEn						
REM sleep	0.90±0.22	0.90±0.22	0.78±0.27	0.018†	0.234	0.776

Data are n (%) or mean±standard-deviation values.
*$p<0.05$ between control vs. OSA+WMC in post-hoc Turkey test, †$p<0.05$ between OSA-WMC vs. OSA+WMC in post-hoc Tukey test, †p-values calculated using ANOVA, §p-values calculated using ANCOVA with age controlled as a covariate, ∥subgroup analysis was performed in subjects who had neither hypertension nor diabetes. There were 24, 33, and 18 subjects in the control, OSA–WMC group, and OSA+WMC group, respectively.
ApEn: approximate entropy, ANCOVA: analysis of covariance, ANOVA: analysis of variance, HF: high frequency, LF: low frequency, NREM: non-rapid-eye-movement, nu: normalized unit, OSA+WMC: obstructive sleep apnea with white-matter changes, OSA-WMC: obstructive sleep apnea without white-matter changes, REM: rapid-eye-movement, RMSSD: root mean square of successive differences, SDNN: standard-deviation of normal to normal RR intervals.

icant difference observed after age adjustment. Additionally, DFA1 during NREM sleep tended to be lower in the OSA+WMC group. These indices were altered regardless of the presence of hypertension or diabetes. In subgroup analysis of middle-aged patients (age 45−64 years) with moderate-to-severe OSA, ApEn during REM sleep was significantly lower in the OSA+WMC group than in the OSA–WMC group. Overall our findings suggest that changes in nonlinear HRV indices reflect sympathetic overactivity in the OSA+WMC group, which might be related to the pathophysiology of WMC in OSA patients.

Changes in HRV indices according to different sleep stag-

es have been reported both in healthy subjects and in patients with OSA.[13] Parasympathetic activity normally increases as a subject approaches a deeper sleep stage during NREM sleep, resulting in increases in the HF component and decreases in the LF component as well as in the LF/HF ratio. The opposite alterations occur during REM sleep and wakefulness, characterized by the predominance of sympathetic activity, resulting in increases in the LF component and LF/HF and a decreased HF component.[11,13] Previous HRV analyses in OSA patients revealed an overall predominance of sympathetic activity during either wakefulness or sleep, characterized by lower total variability and increases in the LF component and
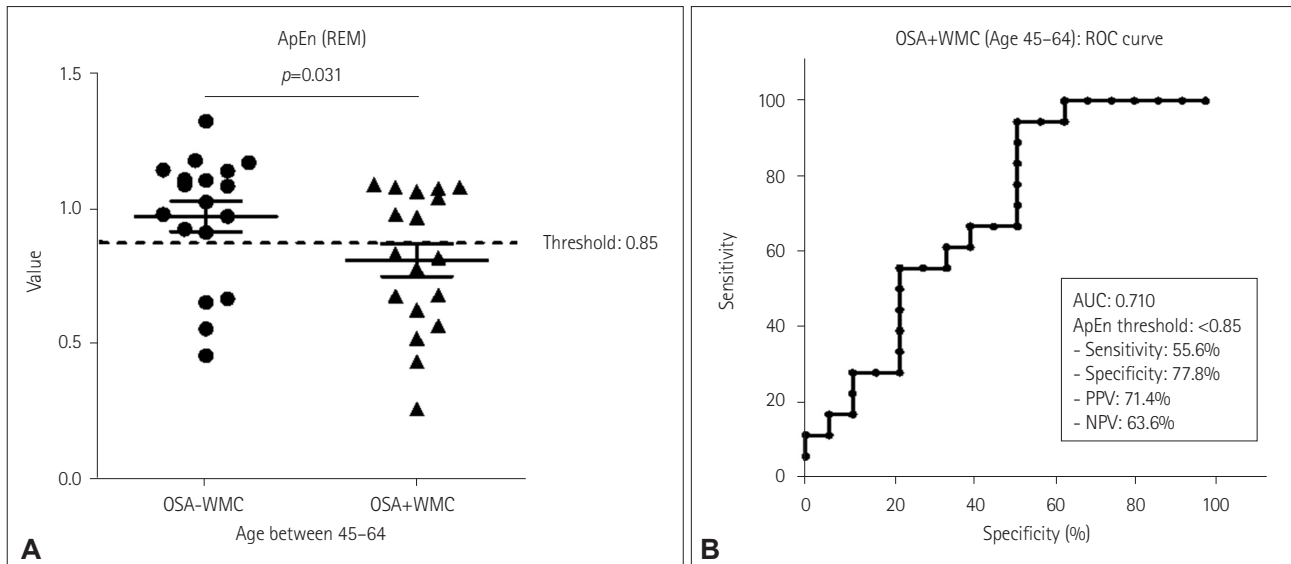
Fig. 2. ApEn values during REM sleep in middle-aged OSA patients (age 45–64 years). A: ApEn values during REM sleep of each subject in the OSA–WMC group (0.94±0.23, *n*=18) and OSA+WMC group (0.79±0.25, *n*=18) (*p*=0.031, Mann-Whitney test). B: ROC curve analysis was performed for diagnosing OSA+WMC among middle-aged OSA patients. The area under the curve was 0.71. With a cutoff of ApEn <0.85, the sensitivity, specificity, PPV, and NPV were 55.6, 77.8, 71.4, and 63.6%, respectively. ApEn: approximate entropy, NPV: negative predictive value, OSA: obstructive sleep apnea, OSA+WMC: obstructive sleep apnea with white-matter changes, OSA-WMC: obstructive sleep apnea without white-matter changes, PPV: positive predictive value, REM: rapid-eye-movement, ROC: receiver operating characteristics.

LF/HF, and a decrease in the HF component.[4,11,24,29]

Our data differed somewhat from previous findings for HRV changes in frequency-domain indices in OSA patients during sleep.[30,31] The frequency-domain HRV indices did not differ among the groups in our study. Analyzing frequency-domain HRV indices in OSA patients is especially challenging due to the presence of repetitive apnea episodes, arousals, and leg movements, which induce biologic noise and can modify the findings of HRV analyses.[11] We consider these factors to be the main reasons for the inconsistencies in findings for frequency-domain HRV indices in OSA patients between previous studies,[31,32] and the current study.

Nonlinear HRV indices may be superior for assessing autonomic cardiovascular regulation in OSA patients. Many of the heart-rate fluctuations occur over a broad frequency range, showing complex, and irregular variability. This suggests that the mechanisms involved in cardiovascular regulation occur in a nonlinear fashion over a long time period, and therefore nonlinear methods will be superior to conventional (linear) measurement techniques for detecting alterations in heart-rate dynamics.[33] Several studies have shown that nonlinear indices are more effective than conventional indices for predicting the condition of the heart and the prognosis in cardiac patients.[15,16,25,34] Moreover, nonlinear indices are known to be less affected by artifacts,[23] which makes them more appropriate for use in OSA patients who intrinsically display many artifacts on the ECG due to recurrent arousals.

SD12 during the entire sleep period was significantly increased and DFA1 during NREM sleep had a tendency to decrease in the OSA+WMC group after age adjustment, which together suggest the predominance of sympathetic activity in the OSA+WMC group. These alterations were consistently observed in patients without hypertension or diabetes, suggesting that our findings are not attributable solely to the presence of hypertension or diabetes. SD12 may reflect the complex nature of HRV changes since it combines two nonlinear characteristics in a single parameter: increased SD12 can reflect increased short-term beat-to-beat variability (SD1) and/or decreased long-term variability (SD2). It is meaningful that SD12 measured during the entire sleep period was significantly higher in the OSA+WMC group, because assessing the ECG data during the entire sleep period can avoid selection bias when choosing ECG samples. Increased SD12 and decreased DFA1 have been reported to be associated with mortality after myocardial infarction,[15] and SD12 has been negatively correlated with DFA1 both in healthy subjects and in patients with coronary artery disease.[22] Increased sympathetic activity is thought to be reflected in a lower DFA1[34]; increasing the infusion doses of norepinephrine resulted in increased SD12 and decreased DFA1.[35,36]

ApEn during REM sleep was the only parameter that differed significantly between the OSA+WMC and OSA–WMC groups in a subgroup analysis of middle-aged patients with moderate-to-severe OSA. ApEn reflects the randomness of HRV, with a lower ApEn indicating decreased randomness,[12]

Table 3. Demographic characteristics and PSG findings in selected middle-aged subjects (age 45–64 years) from the OSA–WMC and OSA+WMC groups

	OSA–WMC (n=18)	OSA+WMC (n=18)	p
Demographic characteristic			
Age (years)	55.3±4.0	57.6±4.9	0.068
Sex (male)	14 (77.8)	14 (77.8)	1.000
BMI (kg/m^2)	25.5±3.0	26.6±2.7	0.226
Neck circumference (cm)	39.4±2.9	40.3±2.9	0.532
HTN (yes)	8 (44.4)	10 (55.6)	0.505
Diabetes (yes)	3 (16.7)	6 (33.3)	0.248
Hyperlipidemia (yes)	2 (11.1)	1 (5.6)	0.546
Alcohol consumption (yes)	8 (44.4)	9 (50.0)	0.738
Current smoker (yes)	4 (22.2)	2 (11.1)	0.371
ESS	10.2±6.2	7.3±3.8	0.126
BDI	14.2±10.1	10.1±8.8	0.204
PSG findings			
Total sleep time (min)	365.3±81.4	364.6±105.9	0.650
Sleep latency (min)	10.9±14.4	7.4±9.8	0.628
AI (n/h)	24.1±15.3	26.0±14.4	0.584
RAI (n/h)	2.0±3.3	3.4±7.8	0.938
AHI (n/h)	20.9±18.0	29.3±17.6	0.064
REM AHI (n/h)	18.2±13.3	26.5±20.9	0.308
NREM AHI (n/h)	21.3±20.7	30.6±19.2	0.059
RDI (n/h)	29.1±24.5	29.1±17.8	0.501
Minimum SaO$_2$ (%)	81.7±4.9	83.6±4.6	0.265
N1 sleep (%)	30.6±16.5	30.8±11.3	0.521
N2 sleep (%)	48.6±16.2	46.7±14.2	0.696
N3 sleep (%)	5.5±5.8	5.4±6.4	0.650
REM sleep (%)	14.8±7.5	15.1±6.5	0.938
Nonlinear index for HRV			
ApEn			
REM sleep	0.94±0.23	0.79±0.25	0.031

p-values were calculated using the chi-square test for categorical variables and the Mann-Whitney test for continuous variables. Data are *n* (%) or mean±standard-deviation values.
AI: arousal index, AHI: apnea-hypopnea index, ApEn: approximate entropy, BDI: Beck Depression Inventory, BMI: body mass index, ESS: excessive daytime sleepiness, HRV: heart-rate variability, HTN: hypertension, n/h: number per hour, NREM: non-rapid-eye-movement, OSA+WMC: obstructive sleep apnea with white-matter changes, OSA-WMC: obstructive sleep apnea without white-matter changes, PSG: polysomnography, RAI: respiratory arousal index, RDI: respiratory disturbance index, REM: rapid-eye-movement, SaO$_2$: oxygen saturation.

and decreased HRV randomness is associated with simplification of cardiovascular regulation, which is observed in aging and pathologic conditions.[11] Patients in the OSA+WMC group demonstrated lower ApEn, which suggests a reduction of parasympathetic tone.[25] A lower ApEn has been reported to be associated with postoperative ventricular dysfunction[25] and poor outcome in arrhythmia patients.[16] We suggest that

decreased ApEn, especially during REM sleep, is a useful marker for predicting WMC in OSA patients.

The identified nonlinear indices that were dysregulated in the OSA+WMC group were concordantly related to sympathetic overactivity and parasympathetic dysfunction, which is a well-known autonomic property of OSA. It is not yet known if increased SD12, decreased DFA1, and decreased ApEn are directly associated with the pathophysiology of cerebral WMC. However, we assume that sympathetic overactivity would play a crucial role in the development of WMC. OSA patients experience recurrent intermittent hypoxemia with consequent sympathetic activation and marked surges in blood pressure, with each of these mechanisms possibly impairing endothelial function[37] and eventually leading to cerebral WMC.[38] It has been reported that sympathetic nerve terminals exist in the tunica media of human cerebral arteries[39] and that the intracranial capillary diameter is regulated by pericyte contraction in response to noradrenergic neurotransmitters.[40] While continuous-positive-airway-pressure treatment can reportedly improve linear HRV parameters,[41,42] whether such treatment can normalize nonlinear indices remains unclear. However, we propose that these nonlinear indices can be used as indicators of WMC in OSA patients, and consequently may be useful when selecting OSA patients on whom to perform brain MRI. For example, the presence of WMC in middle-aged OSA patients could be predicted by ApEn during REM sleep with a moderate positive predictive value (71.4%) and specificity (77.8%).

In addition, some of the time-domain indices differed considerably in OSA+WMC patients. It is noteworthy that SDNN and RMSSD were significantly increased during NREM sleep in the OSA+WMC group (the oldest group), because both parameters are known to decrease with increasing age.[27] Increased nighttime RMSSD has been reported to be independently associated with the progression of small-vessel disease.[43] However, the differences in SDNN and RMSSD in the OSA+WMC group were inconsistent after age adjustment, and so further studies are required to clarify the associations between these parameters and cerebral WMC.

This study was subject to a few limitations. First, the baseline demographic characteristics and the prevalence of chronic conditions such as hypertension and diabetes differed among the included groups. Patients in the OSA+WMC group were significantly older (by a mean of almost 20 years) and had a higher prevalence of hypertension or diabetes than those in the other groups. HRV indices are significantly affected by age, and hypertension and diabetes may impact HRV measures.[26,27] We therefore performed ANCOVA with age controlled as a covariate, and performed additional subgroup analyses in order to minimize the effect of these po-

tential confounders. Second, most of the subjects in the control group had multiple sleep-related complaints, including insomnia, sleep fragmentation, snoring, nonrefreshing sleep, and so they might not have constituted a reliable control group. Third, apnea-free, artifact-free, 5-min ECG samples were selected from each sleep stage for HRV analysis, which might not represent the HRV for each sleep stage. It is known that HRV changes during REM sleep, and can begin during NREM sleep several minutes earlier than the transition to REM sleep[13]; also, the degree of sympathetic activation during REM sleep is affected by the previous sleep stage.[32] We consider these limitations to be the main reasons for the inconsistencies between the findings for the frequency-domain HRV indices between the current study and previous studies.

In summary, certain nonlinear HRV indices were significantly altered in OSA+WMC patients, with SD12 being increased and DFA1 and ApEn being decreased during sleep. In particular, an increased SD12 during the entire sleep period and a decreased ApEn during REM sleep could be useful as markers for predicting cerebral WMC in OSA patients. Future work should investigate the validity of these nonlinear HRV parameters and their causal relationships with the pathophysiologic mechanisms underlying WMC.

Conflicts of Interest

The authors have no financial conflicts of interest.

Acknowledgements

REFERENCES

1. Yaggi HK, Concato J, Kernan WN, Lichtman JH, Brass LM, Mohsenin V. Obstructive sleep apnea as a risk factor for stroke and death. *N Engl J Med* 2005;353:2034-2041.
2. Gonzaga C, Bertolami A, Bertolami M, Amodeo C, Calhoun D. Obstructive sleep apnea, hypertension and cardiovascular diseases. *J Hum Hypertens* 2015;29:705-712.
3. Khoo MC, Kim TS, Berry RB. Spectral indices of cardiac autonomic function in obstructive sleep apnea. *Sleep* 1999;22:443-451.
4. Palma JA, Urrestarazu E, Lopez-Azcarate J, Alegre M, Fernandez S, Artieda J, et al. Increased sympathetic and decreased parasympathetic cardiac tone in patients with sleep related alveolar hypoventilation. *Sleep* 2013;36:933-940.
5. Eguchi K, Kario K, Hoshide S, Ishikawa J, Morinari M, Shimada K. Nocturnal hypoxia is associated with silent cerebrovascular disease in a high-risk Japanese community-dwelling population. *Am J Hypertens* 2005;18:1489-1495.
6. Song TJ, Park JH, Choi KH, Chang Y, Moon J, Kim JH, et al. Moderate-to-severe obstructive sleep apnea is associated with cerebral small vessel disease. *Sleep Med* 2017;30:36-42.
7. Nishibayashi M, Miyamoto M, Miyamoto T, Suzuki K, Hirata K. Cor-

8. Kamba M, Inoue Y, Higami S, Suto Y, Ogawa T, Chen W. Cerebral metabolic impairment in patients with obstructive sleep apnoea: an independent association of obstructive sleep apnoea with white matter change. *J Neurol Neurosurg Psychiatry* 2001;71:334-339.
9. Kim H, Yun CH, Thomas RJ, Lee SH, Seo HS, Cho ER, et al. Obstructive sleep apnea as a risk factor for cerebral white matter change in a middle-aged and older general population. *Sleep* 2013;36:709-715B.
10. Pantoni L. Cerebral small vessel disease: from pathogenesis and clinical characteristics to therapeutic challenges. *Lancet Neurol* 2010;9:689-701.
11. Tobaldini E, Nobili L, Strada S, Casali KR, Braghiroli A, Montano N. Heart rate variability in normal and pathological sleep. *Front Physiol* 2013;4:294.
12. Tarvainen MP, Niskanen JP, Lipponen JA, Ranta-Aho PO, Karjalainen PA. Kubios HRV--heart rate variability analysis software. *Comput Methods Programs Biomed* 2014;113:210-220.
13. Bonnet MH, Arand DL. Heart rate variability: sleep stage, time of night, and arousal influences. *Electroencephalogr Clin Neurophysiol* 1997;102:390-396.
14. Narkiewicz K, Pesek CA, Kato M, Phillips BG, Davison DE, Somers VK. Baroreflex control of sympathetic nerve activity and heart rate in obstructive sleep apnea. *Hypertension* 1998;32:1039-1043.
15. Stein PK, Domitrovich PP, Huikuri HV, Kleiger RE; Cast Investigators. Traditional and nonlinear heart rate variability are each independently associated with mortality after myocardial infarction. *J Cardiovasc Electrophysiol* 2005;16:13-20.
16. Khan AA, Mumtahina U, Yeasmin N. *Heart rate variability analysis using approximate entropy and detrended fluctuation for monitoring heart condition.* Piscataway (NJ): IEEE, 2013.
17. Epstein LJ, Kristo D, Strollo PJ Jr, Friedman N, Malhotra A, Patil SP, et al. Clinical guideline for the evaluation, management and long-term care of obstructive sleep apnea in adults. *J Clin Sleep Med* 2009;5:263-276.
18. Song TJ, Park JH, Choi KH, Kim JH, Choi Y, Chang Y, et al. Is obstructive sleep apnea associated with the presence of intracranial cerebral atherosclerosis? *Sleep Breath* 2017;21:639-646.
19. Song TJ, Cho HJ, Chang Y, Youn M, Shin MJ, Jo I, et al. Low-density-lipoprotein particle size predicts a poor outcome in patients with atherothrombotic stroke. *J Clin Neurol* 2015;11:80-86.
20. Fazekas F, Chawluk JB, Alavi A, Hurtig HI, Zimmerman RA. MR signal abnormalities at 1.5 T in Alzheimer's dementia and normal aging. *AJR Am J Roentgenol* 1987;149:351-356.
21. Berry RB, Brooks R, Gamaldo CE, Harding SM, Lioyd RM, Marcus CL, et al. *The AASM Manual for the Scoring of Sleep and Associated Events: Rules, Terminology and Technical Specifications, Version 2.1.* Darien (IL): American Academy of Sleep Medicine, 2014.
22. Hoshi RA, Pastre CM, Vanderlei LC, Godoy MF. Poincaré plot indexes of heart rate variability: relationships with other nonlinear variables. *Auton Neurosci* 2013;177:271-274.
23. Penzel T, Kantelhardt JW, Grote L, Peter JH, Bunde A. Comparison of detrended fluctuation analysis and spectral analysis for heart rate variability in sleep and sleep apnea. *IEEE Trans Biomed Eng* 2003;50:1143-1151.
24. Palma JA, Iriarte J, Fernandez S, Valencia M, Alegre M, Artieda J, et al. Characterizing the phenotypes of obstructive sleep apnea: clinical, sleep, and autonomic features of obstructive sleep apnea with and without hypoxia. *Clin Neurophysiol* 2014;125:1783-1791.
25. Fleisher LA, Pincus SM, Rosenbaum SH. Approximate entropy of heart rate as a correlate of postoperative ventricular dysfunction. *Anesthesiology* 1993;78:683-692.
26. Beckers F, Verheyden B, Aubert AE. Aging and nonlinear heart rate control in a healthy population. *Am J Physiol Heart Circ Physiol* 2006;290:H2560-H2570.

27. Antelmi I, de Paula RS, Shinzato AR, Peres CA, Mansur AJ, Grupi CJ. Influence of age, gender, body mass index, and functional capacity on heart rate variability in a cohort of subjects without heart disease. *Am J Cardiol* 2004;93:381-385.

28. Smith EE, Saposnik G, Biessels GJ, Doubal FN, Fornage M, Gorelick PB, et al. Prevention of stroke in patients with silent cerebrovascular disease: a scientific statement for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke* 2017;48:e44-e71.

29. Wiklund U, Olofsson BO, Franklin K, Blom H, Bjerle P, Niklasson U. Autonomic cardiovascular regulation in patients with obstructive sleep apnoea: a study based on spectral analysis of heart rate variability. *Clin Physiol* 2000;20:234-241.

30. Gula LJ, Krahn AD, Skanes A, Ferguson KA, George C, Yee R, et al. Heart rate variability in obstructive sleep apnea: a prospective study and frequency domain analysis. *Ann Noninvasive Electrocardiol* 2003;8:144-149.

31. Lado MJ, Méndez AJ, Rodríguez-Liñares L, Otero A, Vila XA. Nocturnal evolution of heart rate variability indices in sleep apnea. *Comput Biol Med* 2012;42:1179-1185.

32. Busek P, Vanková J, Opavský J, Salinger J, Nevsímalová S. Spectral analysis of heart rate variability in sleep. *Physiol Res* 2005;54:369-376.

33. Mäkikallio TH, Tapanainen JM, Tulppo MP, Huikuri HV. Clinical applicability of heart rate variability analysis by methods based on nonlinear dynamics. *Card Electrophysiol Rev* 2002;6:250-255.

34. Jons C, Raatikainen P, Gang UJ, Huikuri HV, Joergensen RM, Johannesen A, et al. Autonomic dysfunction and new-onset atrial fibrillation in patients with left ventricular systolic dysfunction after acute myocardial infarction: a CARISMA substudy. *J Cardiovasc Electrophysiol* 2010;21:983-990.

35. Tulppo MP, Mäkikallio TH, Seppänen T, Airaksinen JK, Huikuri HV. Heart rate dynamics during accentuated sympathovagal interaction.

36. Tulppo MP, Mäkikallio TH, Seppänen T, Shoemaker K, Tutungi E, Hughson RL, et al. Effects of pharmacological adrenergic and vagal modulation on fractal heart rate dynamics. *Clin Physiol* 2001;21:515-523.

37. Kato M, Roberts-Thomson P, Phillips BG, Haynes WG, Winnicki M, Accurso V, et al. Impairment of endothelium-dependent vasodilation of resistance vessels in patients with obstructive sleep apnea. *Circulation* 2000;102:2607-2610.

38. Hoth KF, Tate DF, Poppas A, Forman DE, Gunstad J, Moser DJ, et al. Endothelial function and white matter hyperintensities in older adults with cardiovascular disease. *Stroke* 2007;38:308-312.

39. Oka N, Akiguchi I, Matsubayashi K, Kameyama M, Maeda T, Kawamura J. Density of sympathetic nerve terminals in human superficial temporal arteries: potassium permanganate fixation and monoamine oxidase histochemistry. *Stroke* 1987;18:229-233.

40. Peppiatt CM, Howarth C, Mobbs P, Attwell D. Bidirectional control of CNS capillary diameter by pericytes. *Nature* 2006;443:700-704.

41. Limphanudom P, Chierakul N, Pinyopattarakul N, Nana A, Naruman C, Tangchityongsiva S, et al. Recovery of heart rate variability in patients with moderate to severe obstructive sleep apnea after 6-month continuous positive airway pressure treatment. *J Med Assoc Thai* 2007;90:1530-1535.

42. Kufoy E, Palma JA, Lopez J, Alegre M, Urrestarazu E, Artieda J, et al. Changes in the heart rate variability in patients with obstructive sleep apnea and its response to acute CPAP treatment. *PLoS One* 2012;7:e33769.

43. Yamaguchi Y, Wada M, Sato H, Nagasawa H, Koyama S, Takahashi Y, et al. Impact of nocturnal heart rate variability on cerebral small-vessel disease progression: a longitudinal study in community-dwelling elderly Japanese. *Hypertens Res* 2015;38:564-569.

Am J Physiol 1998;274:H810-H816.