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Serum testosterone/cortisol ratio in people with obstructive sleep apnea

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Abstract

Objectives: Obstructive sleep apnea (OSA) is a major health problem that has been associated with endocrine dysfunction in the hypothalamic-pituitary-gonadal (HPG) and hypothalamic-pituitary-adrenal (HPA) axes. This study investigated cortisol, testosterone, and the testosterone/cortisol ratio in patients with OSA compared to normal sleepers.

Methods: Thirty-nine OSA patients diagnosed by overnight polysomnography (PSG) were divided into three groups, including ten mild OSA patients, 16 patients with moderate OSA, and 13 patients with severe OSA according to the apnea-hypopnea index (AHI). In addition, 13 normal sleepers with normal PSG findings were recruited as the control group. Serum levels of cortisol, testosterone, and sex hormone-binding globulin (SHBG) were measured using enzyme-linked immunosorbent assay (ELISA).

Results: There were no significant differences between the normal sleepers and the three subtypes of OSA in terms of total and free testosterone levels ($P > .1$). The results showed significantly higher levels of cortisol in the severe OSA group compared to the normal sleepers and the two other subtypes of OSA ($P < .01$). In addition, the testosterone/cortisol (T/C) ratio was significantly lower among the severe OSA compared to the moderate OSA patients ($P = .01$). A significant correlation was observed between minimal SpO2 and AHI ($r=-0.69$, $P < .01$), cortisol and AHI ($r = .47$, $P < .01$) and cortisol and minimal SpO2 ($r = -.26$, $P = .06$).

Conclusion: According to the findings, OSA is linked to HPA axis activity in severe OSA patients but not among the mild and moderate subtypes of the disorder.

KEYWORDS
cortisol, HPA axis, obstructive sleep apnea, testosterone

1 | INTRODUCTION

Obstructive sleep apnea (OSA) is the repetitive occurrence of complete (apnea) or partial (hypopnea) obstructions of the upper airway during sleep, which leads to sleep fragmentation and reduced oxygen saturation.[1] OSA is a major public health problem. The prevalence of OSA differs from 14% to 36%, depending on age and ethnicity, and is higher in males than in females.[2-4] The disorder affects ~26% of the middle-aged population.[5,6] It is, however, estimated that up to 90% of those with OSA remain undiagnosed and untreated.[1] OSA is associated with many potential adverse consequences, including motor vehicle accidents and cognitive and behavioral deficits leading to impaired

work performance and productivity, and could ultimately lead to metabolic and cardiovascular disorders, such as hypertension, diabetes, and dyslipidemias.[7-12]

Studies have revealed a connection between the endocrine system and sleep disorders and characteristics, but controversies have also been observed.[13-15] Evidence has shown that OSA is associated with endocrine dysfunction and hypothalamic-pituitary-gonadal (HPG) and hypothalamic-pituitary-adrenal (HPA) axis disorders.[1,4,7,15] OSA has been known as a sex dimorphic and male-biased sleep condition.[16] The interaction of androgen and OSA has been investigated, and testosterone was found to be an important endogenous factor that could have a causal interaction with OSA.[17] Testosterone is a sex hormone and the terminal part of the HPG axis. Several studies have found low serum testosterone levels in patients with OSA.[18,19] The apnea-hypopnea index (AHI) is one of the factors reported to be significantly associated with a low testosterone level among patients with severe OSA.[20] Some studies, however, have shown that the administration of testosterone worsens the severity of OSA.[21,22]

The HPA axis is the main mediator of the body's response to stress for helping reinstate homeostasis. Corticotropin-releasing hormone (CRH) and arginine vasopressin (AVP) are released from the hypothalamic paraventricular nucleus and regulate the secretion of the adrenocorticotropic hormone (ACTH) from the anterior pituitary that sequentially stimulates the adrenal cortex to secrete glucocorticoid hormones, mainly cortisol.[23] The association between sleep problems and the HPA axis could affect cortisol metabolism.[24,25] OSA can induce nightly hypoxic stress and the activation of the stress system via the HPA stress axis,[26] but the association between OSA and HPA activity remains controversial in human subjects.[27] Previous studies have not shown any evidence of the association of OSA with cortisol.[25,28] Another line of evidence has reported that intervention for OSA with continuous positive airway pressure (CPAP) can decrease cortisol in patients with severe OSA.[29]

Studies have demonstrated that the stress-related suppression of testosterone can be a consequence of the elevation of cortisol, and low testosterone levels are a crucial part of chronic psychological and physical stress responses.[30,31] Decreased testosterone, increased cortisol, and the testosterone/cortisol (T/C) ratio have been commonly used as endocrinological indicators of stress.[32-34] Testosterone and cortisol act as part of the biological balance that modulates psychologically and physically integrated human responses and is well studied in sports medicine by measuring T/C ratio as a marker of anabolic/catabolic activities.[35] Furthermore, the T/C ratio has been introduced as a hormonal marker of psychopathologies.[36,37] In addition, the reduction of the T/C ratio has been reported to be associated with ischemic disease.[30] Ghicius et al reported higher T/C ratios among OSA subjects compared to controls in the morning. Imbalances in anabolic-catabolic symmetry suggest the dysregulation of the HPA and HPG axes among OSA subjects.[20] Regardless of the considerable amount of theories on the endocrinological response to OSA, there have been few prospective studies on the interaction between HPG-HPA axes in patients with OSA.

The present study was conducted to measure morning serum T and C levels among a group of OSA patients with different severities and normal sleepers. The relationship between the HPA and HPG axes was also investigated by measuring the T/C ratio in patient and control groups.

2 | MATERIALS AND METHODS

2.1 | Participants

This case-control study was approved by the ethics board of Kermanshah University of Medical Sciences (KUMS) in Kermanshah, Iran, and was conducted from May 2016 to September 2017 at the Sleep Disorders Research Center (SDRC) of KUMS (Ethical Code: KUMS.REC.1395.264). All the participants signed informed consent forms. Fifty-five patients who were referred for complaints of witnessed apnea and/or snoring were consequently invited to participate in the study. Each participant underwent an overnight polysomnography (PSG). From the 55 patients with complaints of excessive daytime sleepiness who underwent an overnight PSG, 39 (30 males; 76.9%, aged 47.64 ± 10.39 years) were diagnosed with OSA. Thirteen normal sleepers (10 males; 76.9%, aged 42.10 ± 11 years) matched for age, gender, and BMI were recruited as the control group.

In the next step, an experienced sleep clinician interviewed the subjects in both the patient and control groups based on the ICSD-II. The participants with chronic medical conditions, any substance abuse problems, psychiatric disorders and chronic neurological, cardiovascular and respiratory disorders were excluded. Those who used medications such as benzodiazepines and antihistamines and consumed alcohol and took any other drugs that could adversely affect sleep were also excluded. Other sleep disorders, including circadian sleep-wake disorders, hypersomnia, parasomnia, and the restless legs syndrome, identified by physician's examinations were also excluded from the study. The female subjects were in the follicular phase of their ovarian cycle. PSG and blood sampling were performed after the end of their menstrual cycle. The women did not have any menstrual disorders, and postmenopausal symptoms were taken as the exclusion criteria.

2.2 | Polysomnography and OSA diagnosis

All the participants completed a nocturnal PSG (SOMNOscreen plus®; Somnomedics) procedure. PSG was performed according to the 2014 American Academy of Sleep Medicine (AASM) guideline (AASM, 2014) with electroencephalographic monitoring using frontal, central, and occipital leads according to the 10-20 system, electrooculogram (EOG), electromyogram (EMG), flow measurement (by oronasal thermistor and nasal air pressure transducer), thoracic and abdominal respiratory efforts (induction plethysmography), oximetry and body position measurement. Thoracoabdominal movements were monitored using piezoelectric strain gauges. Oronasal thermocouples and nasal pressure transducers were used for the monitoring of respiration. Continuous pulse oximetry was also performed.

The AHI was used as the main index for OSA diagnosis and severity assessment based on the AASM.[38] Hypopnea is defined as a minimum of 30% reduction in airflow from baseline in the amplitude of the nasal airflow signal detected by a pressure transducer during sleep that lasts ≥10 seconds and which is associated with either an oxygen desaturation of 3% or an arousal. The complete cessation of breathing for ten or more seconds was taken as indicative of apnea. The AHI was calculated as the number of apneas and hypopneas per hour of sleep. The participants with AHI > 5 were diagnosed as cases of OSA. The AHI was also used for grouping the samples into mild, moderate, and severe cases. The participants were categorized into non-OSA (AHI ≤ 5), mild (5 < AHI ≤ 15), moderate (15 < AHI ≤ 30), and severe (30 < AHI) OSA groups based on the global AHI.[39]

2.3 | Biochemical analysis

Five milliliters of fasting venous blood samples were collected in the morning after the PSG recordings under standard conditions.[40] The samples were centrifuged, and their serum was stored at –20°C to undergo analysis.

Serum levels of cortisol (Code 3625-300; Monobind Inc), testosterone (Code 3725-300; Monobind Inc), and sex hormone-binding globulin [SHBG] (Code 9125-300; Monobind Inc) were measured using enzyme-linked immunosorbent assay kits. The level of absorbance was read using the Awareness Technology STAT FAX 2100 Microplate Reader (Awareness Technology). Hormone levels were calibrated according to the standard calibration curve of each hormone in the corresponding kit. Data were presented in nmol/L for SHBG, ng/mL for testosterone and μg/dL for cortisol.

2.4 | Statistical analysis

Serum levels of cortisol, testosterone, SHBG, free testosterone, and T/C were compared between the patients with OSA and the normal sleepers either by the independent t test for the normally distributed variables or Mann-Whitney's U test for the non-normally distributed variables. The comparisons were made separately for the male and female subjects. The OSA participants were classified into mild, moderate, and severe groups according to the AHI, and all the comparisons were performed between the normal sleepers group and the three OSA groups. The data obtained from the scale were presented as mean and standard deviation. The categorical data were presented as frequency and percentage. According to

the results of the Kolmogorov-Smirnov test, the distribution of the data was normal (P > .05) and the groups were compared using the parametric analysis of variance (ANOVA) and analysis of covariance (ANCOVA). The gender distribution among the normal sleepers and the OSA groups was compared using the chi-square test. In addition, age and BMI were compared among the four groups using the ANOVA. Free testosterone levels were determined by dividing the total testosterone level by the SHBG level. In addition, the level of testosterone was divided by the level of cortisol to obtain the T/C index. The hormone levels and sleep characteristics were compared among the four groups using the ANCOVA, and age, gender, and BMI were used as the covariates. Post hoc Sidak multiple comparisons were used to detect significant differences among the groups. Pearson's correlation coefficient was also used to investigate the relationships between the hormones, SPO2, and AHI. The Statistical Package for Social Sciences (SPSS Inc) version 18.0 was used for the statistical analysis.

3 | RESULTS

3.1 | Demographic findings

From the 55 patients with complaints of excessive daytime sleepiness who underwent an overnight PSG, 39 (30 male patients with a mean age of 47.64 ± 10.39) were diagnosed with OSA. Ten of them had mild OSA (AHI = 8.88 ± 2.15), 16 moderate OSA (AHI = 20.98 ± 4.11), and 13 severe OSA (AHI = 48.16 ± 12.34). Thirteen normal sleepers (ten male patients with a mean age of 42.07 ± 11.14) with normal PSG findings (AHI = 3.00 ± 1.11) were recruited as the control group. Table 1 presents the demographic characteristics of the four groups. As shown in the table, the four groups were matched for age, gender and BMI.

3.2 | Objective sleep findings

Table 2 presents the sleep-related data on the study participants. Sleep characteristics were compared among the four groups using the ANCOVA, and age, gender, and BMI were used as the covariates. There were significant differences in the frequency percentage of stage 1 among the groups. The Sidak post hoc analysis indicated significant differences between the normal sleepers and the moderate OSA groups (P = .01). There were also significant differences in the frequency percentage of stage 3 among the groups. The post hoc

TABLE 1 Comparison of gender distribution, age and BMI between normal sleepers and OSA groups

	Normal (n = 13)	Mild (n = 10)	Moderate (n = 16)	Severe (n = 13)	P-value
Gender (male)	10 (76.90%)	8 (80.0%)	10 (62.50%)	12 (92.30%)	.30[a]
Age (y)	42.10 ± 11.00	42.80 ± 8.00	47.80 ± 17.00	48.10 ± 90[a]	.47[b]
BMI (kg/m^2)	26.60 ± 4.00	29.00 ± 4.70	30.00 ± 5.40	30.80 ± 3.50[a]	.11[b]

Note: Mean ± standard deviation and count (percentage %) are presented for parametric and categorical data, respectively.
[a]Chi-square.
[b]ANOVA.

TABLE 2 Comparison of objective and subjective sleep characteristics between groups

	Normal sleepers n = 13	Mild OSA n = 10	Moderate OSA n = 16	Severe OSA n = 13	P-value
PSG's objective sleep characteristics					
TST	$7.08^a \pm 0.45$	$6.57^a \pm 0.74$	$6.51^a \pm 0.86$	$6.67^a \pm 0.95$	.58
SE	$92.53^a \pm 4.38$	$86.14^a \pm 9.55$	$83.69^a \pm 11.50$	$85.08^a \pm 10.27$	.43
REM%	$8.84^a \pm 9.20$	$10.26^a \pm 6.68$	$9.13^a \pm 11.51$	$11.00^a \pm 11.05$	.66
Stage1% (N1)	$30.98^a \pm 19.67$	$50.14^{ab} \pm 20.81$	$53.96^b \pm 16.39$	$55.19^{ab} \pm 16.21$	.01
Stage2% (N2)	$25.08^a \pm 12.76$	$25.57^a \pm 12.92$	$19.18^a \pm 11.69$	$22.15^a \pm 14.85$	.54
Stage3% (N3)	$32.91^a \pm 19.89$	$14.06^b \pm 10.26$	$17.75^{ab} \pm 13.20$	$11.02^b \pm 12.55$	<.01
REM latency	$68.62^a \pm 73.21$	$50.28^a \pm 76.09$	$51.33^a \pm 98.55$	$84.83^a \pm 114.69$	.76
Wake Index	$2.02^a \pm 0.71$	$4.39^a \pm 2.84$	$5.15^a \pm 4.08$	$5.03^a \pm 4.37$	.07
Minimum SPO2	$86.84^a \pm 2.97$	$86.60^{ab} \pm 2.87$	$80.87^{bc} \pm 5.24$	$74.08^c \pm 9.61$	<.01
Arousal Sleep	$24.25^a \pm 9.42$	$26.99^a \pm 5.87$	$26.02^a \pm 5.69$	$30.72^a \pm 11.55$	.26
Arousal REM	$20.28^a \pm 11.46$	$24.93^a \pm 17.05$	$18.87^a \pm 10.7$	$31.82^a \pm 22.42$	.14
Arousal nREM	$24.43^a \pm 9.29$	$27.16^a \pm 5.42$	$26.34^a \pm 5.98$	$30.7^a \pm 11.59$	.30
AHI	$3.00^a \pm 1.11$	$8.88^a \pm 2.15$	$20.98^b \pm 4.11$	$48.16^c \pm 12.34$	<.01
RDI	$6.24^a \pm 3.84$	$15.20^a \pm 6.68$	$27.15^b \pm 8.74$	$55.19^c \pm 16.96$	<.01

Note: ANCOVA adjusted by sex, age, BMI; means with same superscript letters within a row are not significantly different (P > .05).
Abbreviations: AHI, apnea-hypopnea index; ESS, Epworth Sleepiness Scale total; REM, rapid eye movement; nREM, non-rapid eye movement; RDI, respiratory disturbance index; SE, sleep efficiency; SQ, sleep quality; SOL, sleep onset latency; TST, total sleep time; WASO, wake after sleep onset.

analysis showed significant differences between the normal sleepers and all the OSA groups (P < .01).

3.3 | Biochemical findings

Table 3 compares the data on age, BMI, and hormone levels between the OSA patients and the normal sleepers overall and separately by gender. According to the results, no significant differences were found between the patients and controls in terms of age, cortisol, testosterone, and T/C. BMI was significantly higher among the patients compared to the normal sleepers (P = .02). BMI was also significantly higher in the male patients with OSA compared to the male normal sleepers (P = .04), but the difference was not statistically significant between the female patients with OSA and the female normal sleepers. In addition, SHBG was significantly lower among the OSA patients compared to the controls (P = .01). Similarly, among the male subgroups, SHBG was lower in the patients compared to the controls (P < .01). Free testosterone was significantly higher among the OSA patients (P = .01). The difference was significant when the male OSA patients were compared with the male controls (P < .01), but was not significant when the female OSA patients were compared with the female normal sleepers (Table 3).

Table 4 presents the data on hormone levels in the study participants after dividing the OSA patients into three severity groups. All the biochemical data were analyzed using the ANCOVA after adjustment for gender, BMI, and age. Cortisol levels were also significantly higher in the severe OSA group compared to the normal sleepers and two other OSA groups (P < .01). There were no significant differences in cortisol levels among the mild OSA, moderate OSA, and normal sleepers groups. In contrast, total and free testosterone levels did not differ significantly between the study groups. Meanwhile, T/C differed significantly among the groups (P < .01). The post hoc analysis showed that T/C was significantly higher among the moderate OSA patients compared to the severe group.

Pearson's correlation test was performed for investigating any significant relationships between AHI, SpO2, and the hormonal profile of the participants. According to the results, a significant negative correlation was observed between minimal SpO2 and AHI (r = −.69, P < .01; Figure 1). A significant positive correlation was also observed between cortisol and AHI (r = .47, P < .01; Figure 2). In addition, a significant negative correlation was detected between cortisol and minimal SpO2 (r = −.26, P = .06; Figure 3). No significant correlations were observed between testosterone and AHI (r = .14, P = .29), testosterone and minimal SpO2 (r = .07, P = .57), T/C and AHI (r = .01, P = .45) and T/C and minimal SpO2 (r = .23, P = .90).

4 | DISCUSSION

The hormonal profile was investigated as well, that is, the HPA and HPG axes in the participants with sleep apnea and also in the normal controls. Taking the OSA patients as a single group and comparing them with the controls showed a significantly higher free testosterone in the OSA patients. The difference was significant when the male OSA patients were compared with the male controls. Testosterone, T/C, and cortisol were not significantly different between the OSA and control groups. Comparing the control group with the three OSA subgroups showed that cortisol levels were significantly higher and

TABLE 3 Comparison of biochemical parameters between normal sleepers and OSA groups

	Normal	OSA	P-value
Age (y)			
Total	42.07 ± 11.14	47.64 ± 10.39	.15
Male	44.80 ± 11.09	46.63 ± 11.16	.65
Female	33.00 ± 5.56	46.55 ± 18.24	.24
BMI			
Total	26.62 ± 3.96	29.98 ± 4.59	.02
Male	26.16 ± 4.25	29.02 ± 3.51	.04
Female	28.16 ± 2.92	33.21 ± 6.36	.22
Cortisol (µg/dL)			
Total	137.13 ± 57.52	202.92 ± 142.65	.11
Male	138.99 ± 65.63	211.41 ± 145.64	.14
Female	130.93 ± 19.89	174.64 ± 136.41	.86
Testosterone (ng/mL)			
Total	2.85 ± 2.38	3.82 ± 2.64	.25
Male	3.66 ± 2.12	4.80 ± 2.11	.14
Female	0.16 ± 0.09	0.51 ± 0.99	.56
Testosterone/Cortisol			
Total	0.022 ± 0.020	0.256 ± 0.230	.63
Male	0.028 ± 0.019	0.032 ± 0.022	.79
Female	0.0012 ± 0.0008	0.0033 ± 0.006	.58
SHBG (nmol/L)			
Total	47.69 ± 25.64	31.49 ± 19.04	.01
Male	45.85 ± 18.27	28.09 ± 14.74	<.01
Female	53.72 ± 48.68	42.83 ± 27.33	.86
Free testosterone (ng/mL)			
Total	0.064 ± 0.052	0.17 ± 0.17	.01
Male	0.082 ± 0.045	0.22 ± 0.17	<.01
Female	0.0046 ± 0.0048	0.0079 ± 0.0087	.37

Note: ANCOVA adjusted by sex, age, BMI; means with same superscript letters within a row are not significantly different (P > .05).

testosterone/cortisol levels significantly lower in the patients with severe sleep apnea compared to the controls or the participants with less severe forms of sleep apnea. Testosterone levels did not differ among the groups. The AHI predicted significantly higher cortisol levels in the participants.

Sleep is an important restorative neurophysiological state that plays a crucial role in body hemostasis and quality of life.[41] Any condition that disrupts sleep could affect the restorative ability of this state. OSA is one of the more prevalent sleep conditions[4] that leads to sleep fragmentation and hypoxia due to the repetitive occurrence of apnea or hypopnea during sleep.[1,42]

The OSA groups and the normal sleepers were not significantly different in terms of the various PSG-derived sleep parameters. Moreover, objective sleep quantity and efficiency derived from the PSG findings were similar in the normal sleepers and the OSA patients. In addition, the groups were not significantly different in terms of the wake and arousal indices. A more interesting finding was obtained from the sleep-stage physiologies. Although the four groups were not significantly different in terms of the REM and N2, significant differences were observed in the frequency percentage of stage 1 and stage 3 among the groups. The OSA patients stayed more in lighter sleep (N1) and less in deeper sleep (N3) than the normal sleepers. Furthermore, shifting from light to deeper and slow-wave sleep occurred later among the OSA patients and they stayed in more hemostatic and restorative slow-wave sleep less than the normal group. A previous study reported that patients with an increased severity of OSA stay longer in REM than deep sleep, but that study had not recruited normal sleepers as controls.[43] Moreover, no significant differences were observed in the percentage of REM sleep between the groups.

The relationship between cortisol levels and sleep apnea is a matter of debate. In the present study, serum cortisol levels increased slightly and non-significantly in the mild and moderate OSA groups compared to the controls; however, serum cortisol levels increased considerably in the severe OSA group. In contrast to these findings, other studies have reported lower cortisol levels and hypocortisolemia among OSA patients compared to normal sleepers.[44,45] A negative correlation has also been reported between cortisol levels and AHI.[44] Some studies have failed to find any significant correlations between cortisol and AHI or any significant differences in cortisol level between OSA patients and normal sleepers.[46,47] This study, however, found a positive relationship between AHI and cortisol. Some studies have reported a reduction in cortisol after CPAP in OSA patients compared to the placebo group.[28,48] In addition, increased morning cortisol levels have been reported among OSA patients.[48] The present findings and the results of other studies which have reported increased
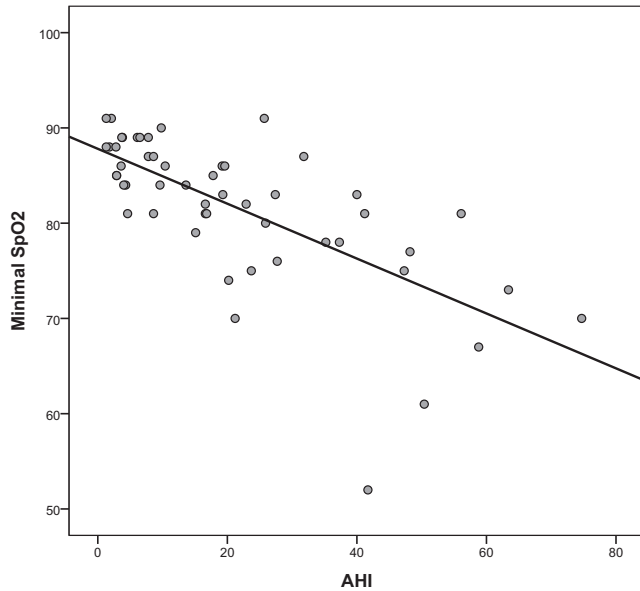
TABLE 4 A comparison of biochemical parameters between the normal sleepers and the OSA groups with regard to the severity of OSA

	Normal (n: 13)	Mild (n:10)	Moderate (n:16)	Severe (n:13)	P-value
Cortisol (µg/dL)	137.14[a] ± 57.52	142.92[a] ± 20.14	149.14[a] ± 120.18	315.29[b] ± 159.84	<.01
Testosterone (ng/mL)	2.85[a] ± 2.40	3.81[a] ± 1.86	3.34[a] ± 2.86	4.39[a] ± 2.93	.26
Testosterone/ cortisol	0.02[ab] ± 0.02	0.03[ab] ± 0.01	0.03[b] ± 0.03	0.01[a] ± 0.01	<.01
SHBG (nmol/L)	45.25[a] ± 24.88	33.60[a] ± 27.67	33.15[a] ± 17.04	27.82[a] ± 14.74	.15
Free testosterone (ng/mL)	0.06[a] ± 0.05	0.15[a] ± 0.10	0.15[a] ± 0.23	0.18[a] ± 0.14	.11

Note: ANCOVA adjusted by sex, age, BMI; means with same superscript letters within a row are not significantly different (P > .05).

FIGURE 1 A scatter plot showing the relationship between the minimal SpO2 and apnea-hypopnea index (AHI)

FIGURE 3 A scatter plot showing the relationship between serum cortisol level and the minimal SpO2

cortisol levels in OSA patients seem biologically plausible. The reciprocal interactions between sleep homeostasis and HPA activity are well recognized. Repeated physiological stress induced by hypoxia and sleep fragmentation affects the HPA axis and cortisol secretion.[48] Stress-responsive biological systems and the sleep circadian rhythm are regulated by the hypothalamus sub-thalamic nucleus [49] and thus closely interact, and any changes in sleep quality and quantity may change HPA axis activity and cortisol levels.[50] In the present study, the severe OSA group had
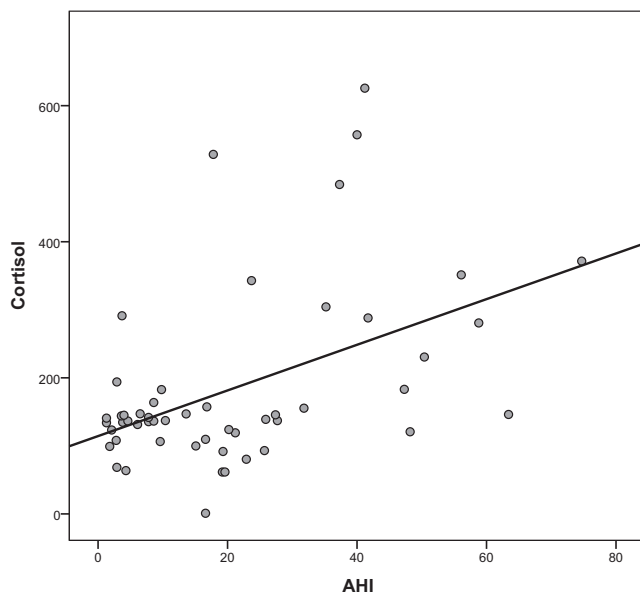

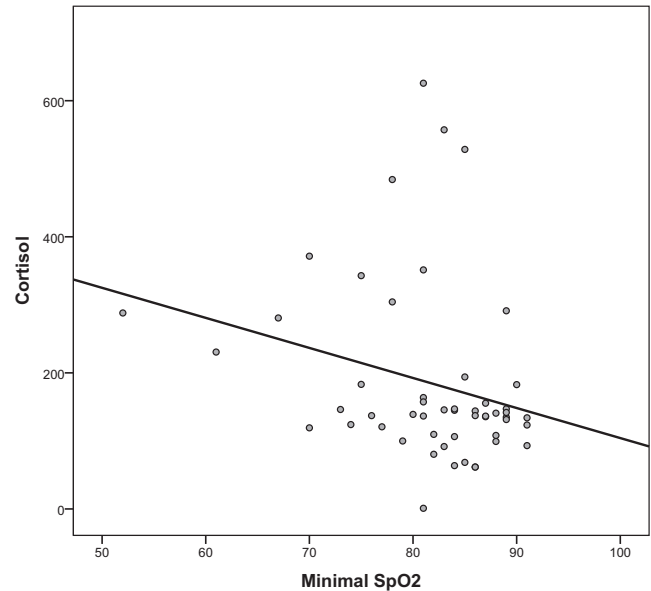**FIGURE 2** A scatter plot showing the relationship between serum cortisol level and AHI

a lower percentage of N3 compared to the other groups, and this decrease in N3 sleep may alter HPA activity and lead to higher cortisol levels. More studies with homogeneous participants, various biochemical samples, and precise sleep laboratory investigations are needed for the further analysis of the existing controversies.

Testosterone levels were higher in the OSA patients, but the differences between the groups were not statistically significant in this regard, which might have been due to the small sample size. Several studies have reported low serum testosterone in OSA patients.[18,19,51] Nonetheless, testosterone has also been reported to increase following physical [52,53] and psychosocial stress.[54,55] If increased cortisol levels are considered a biomarker of stress, an increase is expected in testosterone levels rather than a reduction in the severe OSA group. The treatment of OSA with continuous positive airway pressure (CPAP) did not change the total and free serum testosterone levels.[15] It has also been reported that testosterone therapy worsens sleep-disordered breathing in patients with OSA.[21,22,56,57] Liu et al showed that testosterone levels rise to their peak during the first REM episode.[58] The sleep-related increase in testosterone is thus associated with REM latency.[59] The present study found no significant differences in REM latency and the percentage of REM between the groups. REM latency should therefore be considered an important factor involved in the relationship between OSA and testosterone as the main factor that affects the endogenous rhythm of testosterone production. Future studies should also consider the classification of the disorder into REM-dependent-OSA and NREM-dependent-OSA.[60] Similar to previous studies on this subject, the present study also did not recruit its participants according to this classification.

In the present study, the T/C ratio differed significantly between the groups. The post hoc analysis revealed that this difference was significant only between the moderate and severe OSA groups, as the ratio was significantly lower in the severe compared to the moderate OSA group. Regardless of the considerable theories on the endocrinological response to OSA, there have been few prospective studies on the interaction between HPG and HPA axes in patients with OSA. In contrast to the present findings, Ghicius et al reported a higher T/C ratio among the OSA subjects compared to the controls in the morning.[20] Although some studies have reported a stress-related suppression of testosterone as a consequence of cortisol elevation,[30-34] an increase in testosterone has been reported following physical [52,53] and psychosocial stress.[54,55] Given that the T/C ratio has been introduced as a hormonal marker of psychopathologies [36,37] and since a reduction in this ratio is associated with ischemic diseases,[30] it may be concluded that severe OSA patients are more prone to the serious cognitive and emotional consequences of the disorder, while there is a low risk for serious endocrinological and neuropathological consequences in those suffering from mild or moderate OSA.

The limitations of this study include measuring the hormones in one occasion due to the limited resources available. According to a previous study, the difference in the T/C ratio between the OSA and control subjects may vary depending on the time of blood sampling.[20] Future studies are recommended to investigate the association between the hormonal profile and sleep by measuring 24-hour changes in this profile according to circadian and ultradian rhythms. The concurrent investigation of objective sleep characteristics is a strength of this study, as using objective data helped reveal the association of SpO2 and AHI with the hormonal profile of the subjects. Also, by classifying the participants into mild, moderate, and severe apnea groups, the role of severity in the hormonal profile was identified with precision. Considering the role of gender in HPG activity, it is crucial to compare the sex hormones separately in male and female subjects. Nevertheless, due to the small sample size, it was not possible to perform this analysis between the OSA subgroups; therefore, the analysis was performed between the controls and the OSA patients as a whole.

5 | CONCLUSION

Serum cortisol levels were significantly higher among the severe OSA patients compared to the normal sleepers and those with less severe forms of OSA. In addition, the AHI predicted serum cortisol levels significantly. No significant differences were found in serum testosterone levels between the normal sleepers and the OSA groups. The T/C ratio was significantly lower in the severe OSA group compared to the mild and moderate OSA groups and the control group.

CONFLICTS OF INTEREST

Hiwa Mohammadi, Mohammad Rezaei, Amir Sharafkhaneh, Habibolah Khazaie and Mohammad Rasoul Ghadami declare that they have no conflicts of interest to report.

ETHICAL APPROVAL

All the procedures performed in the study involving human participants were in accordance with the ethical standards of the Ethical Research Committee of KUMS (Code: KUMS.REC.1395.264) and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

INFORMED CONSENT

Informed consent was obtained from all the participants.

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