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Serum S100B protein: a useful marker in obstructive sleep apnea syndrome

Stężenie białka S100B w surowicy jako przydatny wskaźnik w zespole obturacyjnego bezdechu podczas snu

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Abstract

Background and purpose: We aimed to underline the importance of serum S100B protein as a useful biochemical marker in patients with obstructive sleep apnea syndrome (OSAS).
Material and methods: Forty-three newly diagnosed patients with OSAS (median apnea-hypopnea index [AHI, events/hour]: 37.5 [range 11.3-137]) and 25 subjects with AHI < 5 (median AHI: 4.4 [range 0.7-4.8]) were included in the study. Serum S100B protein level was tested in serum samples taken after polysomnography in both groups and the difference between OSAS patients and the control group regarding that level was assessed. In addition, the association of S100B protein serum level with age, body mass index, AHI, mean O_2 saturation percentage during sleep, minimum O_2 saturation value (%) at the end of the apneas, and the time spent at an O_2 saturation less than 90% were analyzed in the OSAS patient group.
Results: Median serum S100B protein level was 133.7 pg/mL (range 20.97-230.70 pg/mL) in patients with OSAS and 16.1 pg/mL (range 10.1-22.9 pg/mL) in the control group ($p < 0.005$). Serum S100B protein level did not correlate with any studied variable ($p > 0.05$ for each correlation coefficient).
Conclusions: Serum S100B protein level is increased in patients with OSAS and may be a useful biochemical marker in those patients.

Streszczenie

Wstęp i cel pracy: Celem pracy było podkreślenie znaczenia stężenia białka S100B w surowicy jako przydatnego wskaźnika biochemicznego u chorych na zespół obturacyjnego bezdechu podczas snu (*obstructive sleep apnea syndrome* – OSAS).
Materiał i metody: W badaniu wzięło udział 43 chorych ze świeżo rozpoznanym OSAS [mediana wskaźnika bezdechów/spłyconych oddechów, AHI (epizody na godzinę): 37,5 (zakres: 11,3–137)] oraz 25 osób z AHI < 5 [mediana: 4,4 (0,7–4,8)] stanowiących grupę kontrolną. W obu grupach zmierzono stężenie białka S100B w surowicy pobranej po wykonaniu polisomnografii i sprawdzono różnicę w tym zakresie między grupami. Ponadto w grupie chorych na OSAS określono korelację między stężeniem białka S100B w surowicy a wiekiem, wskaźnikiem masy ciała, AHI, średnim wysyceniem krwi tętniczej tlenem podczas snu, najmniejszym wysyceniem krwi tętniczej tlenem na zakończenie okresu bezdechu oraz czasem, w którym wysycenie krwi tętniczej tlenem wynosiło $< 90\%$.
Wyniki: Mediana stężenia białka S100B w surowicy wyniosła 133,7 pg/ml (zakres: 20,97–230,70 pg/ml) u chorych na OSAS oraz 16,1 pg/ml (zakres: 10,1–22,9 pg/ml) w grupie kontrolnej ($p < 0,005$). Stężenie białka S100B w surowicy nie korelowało z żadną ocenianą zmienną ($p > 0,05$ dla każdego współczynnika korelacji).

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Wnioski: Stężenie białka S100B w surowicy jest zwiększone u chorych na OSAS i może być przydatnym wskaźnikiem biochemicznym u tych pacjentów.

Słowa kluczowe: białko S100B, zespół obturacyjnego bezdechu podczas snu, znacznik.

Introduction

Obstructive sleep apnea syndrome (OSAS) is characterized by episodic apnea/hypopnea due to upper airway obstruction during sleep. This syndrome is usually associated with increased cardiovascular morbidity and mortality [1]. Patients are diagnosed with OSAS if the value of the apnea-hypopnea index (AHI), obtained through polysomnography (PSG), exceeds 5. The prevalence of OSAS is approximately 2% in women and 4% in men [2]. Upper respiratory tract obstruction in OSAS occurs due to a predisposition for pharyngeal collapse caused by a small lumen or increased extraluminal pressure. Upper respiratory tract obstruction is thought to be an effect, rather than a cause, which is triggered by a factor of central origin [3]. The pathophysiological mechanisms in OSAS include nocturnal intermittent hypoxia, apneas, increased sympathetic tone, inflammation, and oxidative stress.

Obstructive sleep apnea syndrome causes various problems, from simple snoring to serious pulmonary, cardiovascular, endocrine, and psychiatric disorders. Patients with OSAS experience daytime sleepiness, impaired memory, disordered cognitive functions, and difficulty in performing their daily activities.

In recent years, interest in the analysis of neurobiochemical markers, such as S100B protein, glial fibrillary acidic protein, and neuron-specific enolase, for various central nervous system diseases has been gradually increasing [4]. S100B protein is a member of the S100 family. The S100 family, which performs many intracellular and extracellular regulating activities, is a broad subgroup of calcium binding proteins [5]. The members of this protein family interact with many effector proteins, regulate enzyme activities, affect the structure of the cytoskeleton, and ensure cell growth, cell differentiation and calcium homeostasis. S100B protein is coded via the 22.3 locus of chromosome 21 [6]. S100B is produced principally by astrocytes and has autocrine and paracrine effects on glia, neurons and microglia [7].

In recent studies, a correlation was found between the size of the infarct resulting from ischemic brain damage and the serum S100B protein level [8,9], and it is possible to use protein S100B measurements to monitor stroke treatment [10]. Increased S100B levels in cases of hypoxia are similar to those noted in traumatic damage. In a few recent studies performed in OSAS patients, increased serum S100B protein levels were found [11,12].

In the current study, the serum S100B protein levels of patients with OSAS were measured and compared with the control group. We also aimed to demonstrate the relationship between serum S100B protein levels and sleep-related variables such as AHI values, mean O_2 saturation during sleep, minimum O_2 saturation value at the end of the apnea and the time spent at an O_2 saturation below 90% in patients with OSAS.

Material and methods

Patients

In total, 68 patients (26 women) who presented to the sleep outpatient clinic in the Chest Diseases Department of Diskapi Yıldırım Beyazıt Training and Research Hospital between January 2011 and July 2011 with complaints of snoring, witnessed apnea at night and excessive daytime sleepiness were enrolled in the study. Patients with comorbid diseases, acute infections, malignancies, recent myocardial infarction, recent surgery, chronic use of sleep-affecting drugs (such as hypnotics), alcohol intake, restless leg syndrome, sleep phase disorder, and/or periodic leg movements during sleep were excluded from the study.

The study was planned in compliance with the Helsinki Declaration and with the recommendations of our hospital's ethics committee. All patients signed a voluntary informed consent form before the study commenced.

Methods

Demographic data, smoking habits and alcohol consumption habits of the patients in the study were investigated. A detailed physical examination was performed in each patient, and height, body weight and neck cir-

cumference measurements were taken. After the patients were assessed by an ear-nose-throat specialist, those found to have anatomical nasal and/or chin problems (such as septal deviation, conchal hypertrophy, or retromicrognathia) were excluded from the study. Apnea was defined as complete cessation of airflow for 10 seconds. The Epworth Sleepiness Scale (ESS) was applied to the patients to evaluate daytime sleepiness. The validity of the Turkish version of the scale was previously documented [13].

The body mass index (BMI) of each patient was calculated by dividing body weight in kilograms by the square of the patient's height in meters. To exclude additional diseases, levels of plasma aspartate and alanine aminotransferase, bilirubin, urea, creatinine, total T3 and T4, thyroid-stimulating hormone, total cholesterol, triglyceride, high density lipoprotein, low density lipoprotein, hemogram, and C-reactive protein, as well as erythrocyte sedimentation rate, were examined using commercial kits and standard laboratory tests. In addition, respiratory function tests, blood gas analysis, electrocardiography, and chest X-rays were assessed. Respiratory function tests were performed in the hospital's respiratory laboratory according to American Thoracic Society (ATS) criteria and using a Jaeger spirometer. Forced expiratory volume in one second (FEV_1) and forced vital capacity (FVC) were measured at least three times and the optimal ratios (FEV_1/FVC, %) were recorded. All patients were hospitalized in our sleep clinic for one night to undergo PSG. Fasting venous blood samples of 10 cc were taken from patients between 7:00 a.m. and 8:00 a.m. after PSG. Then, the blood samples were centrifuged at 5000 rpm for 10 minutes. Separated serum samples were stored at –70°C. Serum S100B protein levels were measured using the ELISA (Dia Metra, Italy) kit.

Before PSG, a standard questionnaire, including ESS, was administered. Following the physical examination, patients, who were tested for respiratory functions, underwent a sleep technician attended polysomnographic examination for a whole night using a 64-channel Comumedics® E-series polysomnography device (for 6-8 hours). Six-channel electroencephalogram (EEG; C3-M2, C4-M1, F1-M2, O1-M2, O2-M1, F2-M1), electrooculogram (EOG), electromyogram (EMG, submental and tibialis muscles), electrocardiogram (EKG), oronasal air flow (cannula and thermistor), abdominal and thoracic respiratory movements, snoring (tracheal microphone), body positions, and pulse oximeter (fingertip) parameters were evaluated using the Profusion software program version 3. Scoring of sleep and respiratory events was performed (e.g., apnea, hypopnea, and desaturation) according to American Academy of Sleep Medicine (AASM) criteria. Sleep recordings with sleep efficacy of 69% or more were considered valid. Patients were divided into three groups based on AHI and analysis was performed within each group as follows: normal (AHI < 5; $n = 25$), mild OSAS (AHI between 5 and 15; $n = 13$), moderate OSAS (AHI between 15 and 30; $n = 21$), or severe OSAS (AHI > 30; $n = 9$).

Statistical analysis

For normally distributed data, average measurements were calculated as mean ± standard deviation (SD); median was used for non-normally distributed variables. Differences between ages and BMIs of the control group and the patient group were normally distributed and investigated using the t-test for independent samples, while differences in serum S100B protein levels, AHI values, mean O_2 saturation during sleep, minimum O_2 saturation value at the end of the apnea and the time spent at an O_2 saturation below 90%, which were not normally distributed, were investigated using the Mann-Whitney *U*-test. A difference was considered statistically significant if a p-value was less than 0.05 in the two-sided statistical testing. The relationship between serum S100B protein level and other OSAS-related continuous variables was investigated using the Spearman correlation analysis. Correlations with p-values less than 0.05 were interpreted as significant.

Results

Demographic and PSG data for the 68 patients monitored overnight at our hospital are shown in Table 1.

Serum S100B concentration was significantly greater in patients with OSAS than in controls ($p < 0.005$) (Table 1).

The relationships between serum S100B protein level and the other OSAS patient variables are shown in Table 2. Serum S100B protein level did not correlate with age, BMI, AHI, mean O_2 saturation during sleep, minimum O_2 saturation value at the end of the apnea or the time spent at an O_2 saturation below 90%.

Discussion

Up to now, there have been few studies investigating serum S100B protein levels of patients with OSAS.

Table 1. Demographic, polysomnographic and laboratory data of studied patients and controls

Variables	OSAS (n = 43)	Controls (n = 25)	P-value
Age [years]; mean (SD)	47.23 (6.24)	43.76 (8.82)	> 0.05
Sex [female/male]; n	18/25	8/17	> 0.05
Cigarette smoking [pack/year], (+/–)	10/33	5/20	> 0.05
Body mass index [kg/m^2], median (range)	29.6 (24.4-46.1)	27 (21.5-33.9)	> 0.05
Apnea-hypopnea index [events/hour]; median (range)	37.5 (11.3-137)	4.4 (0.7-4.8)	0.000
Epworth Sleepiness Scale; mean (SD), range	10.8 (5.9), 8-11	9.2 (5.1), 8-10	> 0.05
Mean O_2 saturation (%) during sleep; median (range)	85 (67-89)	94 (92-96)	0.000
Minimum O_2 saturation value (%) at the end of the apneas; median (range)	69.35 (67-72)	92 (92-94)	0.000
Time spent at O_2 saturation below 90% [minutes]; median (range)	141.7 (39.3-337.5)	0	0.000
Serum S100B protein level [pg/mL], median (range):			
All patients/controls	133.77 (20.9-230.7)	16.1 (10.1-22.9)	0.000
Mild OSAS (n = 13)	135.6 (20.9-214.6)		
Moderate OSAS (n = 21)	145.53 (24.67-221.56)		
Severe OSAS (n = 9)	147.34 (29.78-230.7)		

OSAS – obstructive sleep apnea syndrome; SD – standard deviation

In this study, a significant difference was observed between serum S100B protein levels of subjects in the control group and OSAS patients with no concomitant disease which might cause cerebral damage ($p < 0.005$). However, no significant correlation was found between serum S100B protein level and OSAS related variables such as AHI, mean O_2 saturation during sleep, minimum O_2 saturation value at the end of the apnea, the time spent at an O_2 saturation below 90% or patient characteristics, like age and BMI.

In recent years, S100B protein has been largely focused on as a biochemical marker of cerebral disorders. Increased serum S100B protein level may reflect either glial damage or astrocytic reactions to neural injury [14]. Serum S100B protein levels increase after trauma and after stroke [15]. In previous studies, serum S100B protein level was associated with severity of trauma and disease prognosis [16]. In hypoxia following cardiac arrest, serum S100B protein level has been correlated with clinical manifestation and the stage of coma [17,18].

In OSAS, recurrent apnea-hypopnea episodes lead to desaturation and intermittent hypoxia with deteriorating oxygenation. Therefore, cerebral damage may occur following hypoxia. Recent studies on serum S100B protein levels of patients with OSAS have generated different results. Silva *et al.* [11] found that

Table 2. Correlation between serum S100B protein level and sleep-related variables in patients with obstructive sleep apnea syndrome (r – Spearman correlation coefficient)

Variables	Serum S100B protein level (pg/mL)	
	r	p-value
Age [years]	–0.004	0.777
Body mass index [kg/m^2]	–0.166	0.287
Apnea-hypopnea index [events/hour]	0.150	0.333
Mean O_2 saturation (%) during sleep	–0.002	0.989
Minimum O_2 saturation value (%) at the end of the apneas	–0.262	0.089
Time spent at O_2 saturation below 90% [minutes]	0.113	0.476

S100B levels were higher after PSG than before PSG. Braga *et al.* [12] reported a significantly higher serum S100B protein level in patients with OSAS compared to the control group.

In another study, serum S100B protein level in OSAS patients was slightly increased in only two patients with severe OSAS [19]. On the other hand, among 60 male patients with OSAS, a significant difference was found between morning and evening serum S100B protein levels in patients with moderate OSAS, but no such

difference was observed for patients with severe OSAS. In addition, serum S100B protein level correlated negatively with AHI values and oxygen desaturation index, and positively with basal saturation and average minimal oxygen saturation [20].

In both traumatic and hypoxic cases, degeneration may occur in astrocytes. In such cases, serum S100B protein level increases. In many clinical trials, increased serum S100B protein levels in the early post-stroke period have been associated with clinical and/or functional outcomes [21]. In prior studies [22,23], cerebrovascular and neuropsychiatric diseases caused increased serum S100B protein release from astrocytes. The other studies showed that the patients with OSAS had additional diseases. So the increases of serum S100B protein level in patients in other studies suggest that OSAS can be controlled by a complex alarm network. In this study, we determined that serum S100B protein level was increased in OSAS patients with no additional disease or morbid obesity. We propose that OSAS may increase release of S100B protein in astrocytes.

OSAS is characterized by disordered respiration patterns during sleep. In OSAS, repetitive episodes of apnea cause increased sympathetic nerve activity, increased surges in arterial blood pressure, oxidative stress, hypoxia and hypercapnia [24]. To date, the mechanism of action of S100B protein remains unclear. One could assume that those cerebral changes which occur during sleep may increase the serum S100B protein level. Different diseases produce different S100B protein levels, suggesting that it has different effects on the various parts of the body depending on the disease. For example, in nanomolar concentrations it stimulates neuron growth and protects neurons against oxidative stress. If it reaches the micromolar level in the extracellular region of the brain, it stimulates inflammation, induces apoptosis and causes neuron damage [6].

Serum S100B protein concentration does not correlate with the level of that protein in the brain and cerebrospinal fluid [25], because S100B protein cannot pass freely through the blood-brain barrier. For the reasons explained above, our study may not reflect a relation between serum S100B protein and OSAS-related variables.

OSAS may be associated with cardiovascular and metabolic diseases and conditions such as hypertension, diabetes mellitus, obesity, coronary artery disease, stroke, and heart failure. Therefore, OSAS is an important health problem, and future clinical use of the serum S100B protein level may be helpful in the evaluation of OSAS. We believe that the results of our study will contribute to the literature since only a few studies have been performed on serum S100B protein levels in patients with OSAS. The major limiting factor in our study was that it is difficult to find patients having OSAS without any additional diseases.

Conclusion

Serum S100B protein levels may increase in patients with OSAS, suggesting an astrocyte reaction in patients with OSAS. S100B protein may therefore be a useful biochemical marker in OSAS patients.

Disclosure

Authors report no conflict of interest.

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