ORIGINAL ARTICLES

Hemodynamic and Inflammatory Markers of Sleep Apnea-Hypopnea Syndrome and Nocturnal Hypoxemia: Effects of Treatment With Nasal Continuous Positive Airway Pressure

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OBJECTIVE: In this study, we assessed factors associated with cardiovascular risk in patients with sleep apnea-hypopnea syndrome (SAHS) through analysis of plasma concentrations of N-terminal prohormone brain natriuretic peptide (NTproBNP) and high-sensitivity C-reactive protein (hsCRP). In addition, we analyzed the effect of nasal continuous positive airway pressure (nCPAP) on these markers.

PATIENTS AND METHODS: Forty-two patients with SAHS (mild to moderate in 15 cases and severe in 27) were compared with 14 individuals without SAHS. The participants were not receiving drug treatment and they did not have diabetes, hypertension, marked dyslipidemia, or cardiovascular disease, which was ruled out both clinically and by echocardiography and ^{99m}Tc-tetrofosmin scintigraphy at rest and during exercise. The effects of nCPAP in patients with severe SAHS were analyzed after 6 months of treatment.

RESULTS: Following adjustment for age, body mass index, and smoking habit, the mean concentrations of markers were not significantly higher in patients with severe SAHS than in those with mild-to-moderate SAHS or in control subjects. Nevertheless, in patients with SAHS the main factor influencing NTproBNP concentrations was the percentage of time with a nocturnal arterial oxygen saturation of less then 90% (r=0.37, P=.017). No variables predictive of hsCRP concentration were identified. The concentrations of the markers were reduced by nCPAP, but the differences were not statistically significant.

CONCLUSIONS: While nocturnal hypoxemia in SAHS is responsible for variations in the plasma concentration of NTproBNP (as a result of cardiovascular changes), SAHS appears not to be associated with the inflammatory marker hsCRP when patients with heart disease, cardiovascular risk factors, or those receiving pharmacologic treatment are excluded.

Key words: *Sleep apnea. Hypoxemia. ProBNP. Inflammation.*

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Marcadores hemodinámicos e inflamatorios del síndrome de apneas-hipopneas durante el sueño e hipoxemia nocturna. Efectos del tratamiento nasal con presión positiva continua de la vía aérea nasal

OBJETIVO: Investigamos los factores del síndrome de apneas-hipopneas durante el sueño (SAHS) que activan los mecanismos de riesgo cardiovascular, a través del estudio de las concentraciones plasmáticas del fragmento N-terminal del precursor del péptido natriurético cerebral (NTproBNP) y de la proteína C reactiva de alta sensibilidad (PCRas), así como el efecto que sobre ellos tiene el tratamiento con presión positiva continua de la vía aérea nasal (CPAPn).

PACIENTES Y MÉTODOS: Se estudió a 42 pacientes con SAHS (leve-moderado en 15 casos y grave en 27), comparados con 14 personas sin SAHS. No tomaban fármacos ni presentaban diabetes, hipertensión, dislipemia importante o enfermedad cardiovascular, que se descartó tanto clínicamente como por ecocardiografía y tomografía computarizada por emisión de fotón cínico-esfuerzo con ^{99m}Tc-tetrofosmina. En los pacientes con SAHS grave se estudiaron los efectos de 6 meses con CPAPn.

RESULTADOS: Ajustando por edad, índice de masa corporal y tabaquismo, las medias de los biomarcadores no fueron significativamente más altas en los pacientes con SAHS grave que en aquéllos con SAHS leve-moderado o en los controles. Sin embargo, en los pacientes con SAHS el principal factor que influyó en las concentraciones de NTproBNP fue el porcentaje de tiempo con saturación arterial de oxígeno nocturna menor del 90% (r = 0,37; p = 0,017), sin que se encontrara ningún predictor de los valores séricos de la PCRas. La aplicación de CPAPn hizo descender, pero no significativamente, las concentraciones de los biomarcadores.

CONCLUSIONES: Mientras que la hipoxemia nocturna en el SAHS es la responsable de las variaciones en los valores del NTproBNP, derivado de la afectación cardíaca, el SAHS no parece estar asociado con el biomarcador inflamatorio PCRas, cuando se excluye a los pacientes con alteraciones cardíacas, factores de riesgo cardiovascular o en tratamiento farmacológico.

Palabras clave: *Apnea del sueño. Hipoxemia. ProBNP. Inflamación.*

Introduction

Sleep apnea-hypopnea syndrome (SAHS) is associated with increased cardiovascular morbidity and mortality.[1] However, many risk factors for SAHS, such as male sex, advanced age, and, particularly, obesity, are shared with cardiovascular disease, making it difficult to assess whether SAHS is itself an independent risk factor. When these factors are taken into consideration, the cardiovascular risk associated with SAHS is reduced.[2] The only well-established association with SAHS is currently that observed with hypertension.[3] However, cardiovascular disease represents the most important complication. There is increasing evidence that SAHS affects left-ventricular function.[4] The degree of diastolic dysfunction in patients with diastolic heart failure is greater when associated with sleep-disordered breathing. Fung et al[5] showed that 36.8% of patients with SAHS had diastolic dysfunction. In addition, Arias et al[6] reported that SAHS can affect left ventricular diastolic function independently of other possible risk factors.

Clinical studies have shown that brain natriuretic peptide (BNP) and its precursor proBNP are biomarkers of left ventricular dysfunction and indicators of disease prognosis.[7,8] Measurement of these markers can facilitate the diagnosis of heart failure[9] and lead to suspicion of silent myocardial ishcemia[10] and other acute ischemic phenomena.[11] Secretion of proBNP occurs mainly in the ventricle, and it is processed at this point into the physiologically active BNP (77-108) and the N-terminal fragment or NTproBNP (1-76). Although derived from a common precursor, these fragments differ in a number of respects. NTproBNP is not biologically active and therefore does not activate clearance mechanisms. As a result, it has a half-life of approximately 60 to 120 minutes and is much more stable than BNP; the variability of NTproBNP concentrations within a given patient is therefore less. In addition, it can be collected directly in sample tubes and analysis of its concentration is more sensitive than analysis of BNP in certain processes.[12]

C-reactive protein (CRP) is a nonspecific marker of inflammation. Various studies have shown that it is an important predictor of risk in arteriosclerosis and coronary heart disease.[13] It has also been reported that, in patients with SAHS, CRP concentration is elevated[14] and positively correlated with the severity of the disease.[15] If obesity and SAHS are considered to be inflammatory processes, as indicated by high serum levels of markers of systemic inflammation, it is important to consider whether the high concentrations of CRP in patients with central obesity are a consequence of obesity or of SAHS.[16]

This study had the following aims: to determine whether the concentrations of NTproBNP and CRP are elevated in patients with SAHS, since from a pathophysiologic point of view SAHS can cause left ventricular dysfunction and is associated with inflammatory processes; to identify factors that are associated with or predictive of such increases; to identify variables that are independent predictors of variation in NTproBNP and CRP concentration; and to evaluate whether treatment leads to a reduction in the concentrations of these biomarkers, leading to improvements in ventricular dysfunction and reduction of the inflammatory response, and thus, a reduction in the risk of cardiovascular mortality in these patients. It remains to be determined whether the mechanisms underlying cardiovascular risk in SAHS are activated as a consequence of the apnea-hypopnea index (AHI), the magnitude of hypoxemia, or the number of arousals.

Patients and Methods

Patients

The study included 42 consecutively enrolled patients with a new diagnosis of SAHS and 14 control subjects (Table 1). All of the patients with SAHS were recruited from the Sleep Unit of Hospital Universitario de Salamanca in Salamanca, Spain, after they were referred for suspected SAHS, which was diagnosed by polysomnography. After enrollment of patients, healthy individuals were recruited from among the staff of the same hospital. The control subjects were matched for age, sex, and body mass index (BMI), and they did not have symptoms of SAHS; following conventional polysomnography they were classified as controls on the basis of their AHI. A complete history was obtained for all subjects, and all were questioned regarding smoking. Signed informed consent was obtained prior to inclusion in the study. The study was approved by the ethics committee of Hospital Universitario de Salamanca.

Study Protocol

The inclusion criterion for patients with SAHS was an AHI of at least 10, and for healthy control subjects the criteria were an AHI of less than 10 and a score of less than 10 on the Epworth Sleepiness Scale (ESS). The following exclusion criteria were used for the 2 groups: *a)* unwillingness or inability to undergo the diagnostic procedures; *b)* current use of medication; *c)* obstructive or restrictive pulmonary disease; *d)* daytime hypoxemia or hypercapnia; *e)* cardiac arrhythmias; *f)* history of hypertension (blood pressure ≥140/80 mm Hg) or moderate-to-severe dyslipidemia (cholesterol ≥239 mg/dL, triglycerides ≥400 mg/dL); *g)* diabetes mellitus; *h)* morbid obesity (BMI ≥40 kg/m^2); *i)* any degree of hepatic or renal dysfunction; and *j)* left ventricular ejection fraction less than 50%, ischemic or valvular heart disease, myocardial heart disease, pericardial heart disease, or cerebrovascular accident diagnosed on the basis of patient history and physical examination, electrocardiogram, chest x-ray, echocardiogram, conventional cardiac stress test, and ^{99m}Tc-tetrofosmin scintigraphy at rest and during exercise. The criteria for withdrawal of enrolled patients were *a)* worsening of symptoms that necessitated a change in treatment, *b)* hospital admission, and *c)* mean use of nasal continuous positive airway pressure (nCPAP) of less than 4 hours.

The examinations and diagnostic procedures included in the protocol were carried out in all subjects. In those subjects who fulfilled the first 9 criteria and who provided informed consent, echocardiography, cardiac stress testing, and ^{99m}Tc-tetrofosmin scintigraphy were used to rule out heart disease (valve disease, myocardial heart disease, pericardial effusion, heart failure, or silent myocardial infarction). Finally, polysomnography was performed in the members of the study group.

Complete diagnostic polysomnography was performed using a Medicid 4 polysomnograph (Neuronic, Saragossa, Spain). Sleep stages and respiratory events were analyzed manually according to standard international criteria.[17,18] Mild SAHS was classified as corresponding to an AHI ≥10 and <20; moderate SAHS as an AHI ≥20 and <30; and severe SAHS as an AHI

TABLE 1
Characteristics of the Study Subjects[a]

	Controls	Mild-to-Moderate SAHS	Severe SAHS
No. of patients	14	15	27
Sex, male/female	12/2	14/1	25/2
Age, y	43.9 (3.1)	43.6 (8.1)	48.4 (12.2)
BMI, kg/m^2	28.6 (2.0)	28.8 (1.8)	30.0 (2.3)
Smokers	4 (30.7%)	5 (33.3%)	8 (29.6%)
Systolic blood pressure, mm Hg	132.5 (9.0)	131.7 (6.9)	127.7 (9.4)
Diastolic blood pressure, mm Hg	79.7 (2.5)	81.2 (7.2)	79.3 (8.1)
Glucose, mg/dL	84.5 (9.8)	86.2 (6.4)	92.5 (11.9)[b]
Total cholesterol, mg/dL	191.6 (32.9)	211.2 (38.5)	217.9 (33.1)[b]
HDL-C, mg/dL	49.5 (8.1)	50.0 (12.4)	49.5 (15.4)
LDL-C, mg/dL	118.9 (31.7)	139.0 (34.5)	140.7 (28.0)[b]
Triglycerides, mg/dL	116.1 (30.5)	110.8 (31.1)	131.9 (64.3)
AHI, events/h	5.0 (2.3)	19.9 (5.4)[c]	64.4 (22.9)[d,e]
Desaturation index, events/h	8.9 (9.9)	22.9 (15.6)	20.8 (22.3)[b]
Arousals index, events/h	10.2 (3.8)	20.6 (7.1)[c]	36.0 (17.8)[d,e]
SaO_2<90%, %TST	2.0 (1.7)	10.3 (9.6)[f]	29.5 (23.9)[d,e]
Mean nighttime SaO_2, %	94.7 (1.7)	93.3 (1.5)	91.3 (3.8)d
Total sleep time, min	367.9 (38.9)	355.0 (57.9)	369.0 (79.6)
Sleep efficiency	82.2 (6.2)	82.7 (8.8)	81.1 (11.2)
Epworth score	7.7 (1.7)	10.3 (3.4)[f]	10.6 (4.5)[d]

Abbreviations: AHI, apnea-hypopnea index; BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; SAHS, sleep apnea-hypopnea syndrome; SaO_2, arterial oxygen saturation; SaO_2<90%, percentage of time with SaO_2<90%; TST, total sleep time.
[a]Data are shown as mean (SD) or number of patients (%) for continuous and categorical variables, respectively.
[b]*P*<.05, controls vs patients with severe SAHS (Mann-Whitney test).
[c]*P*<.01, controls vs patients with mild-to-moderate SAHS (Mann-Whitney test).
[d]*P*<.01, controls vs patients with severe SAHS (Mann-Whitney test).
[e]*P*<.01, patients with mild-to-moderate SAHS vs patients with severe SAHS (Mann-Whitney test).
[f]*P*<.05, controls vs patients with mild-moderate SAHS (Mann-Whitney test).

≥30. Of the 42 patients, 15 were diagnosed as having mild or moderate SAHS and 27 as having severe SAHS. The validated Spanish version of the ESS[19] was used to obtain a subjective quantification of the level of sleepiness. At 8:00 AM, at the end of polysomnography, a sample of venous blood was obtained for analysis, including concentrations of NTproBNP and high-sensitivity CRP (hsCRP).

In this prospective study, patients with severe SAHS and daytime symptoms received effective nCPAP with the Solo CPAP System (Respironics, Carlsbad, California, USA) for 6 months following overnight polysomnographic titration. Titration began at a pressure of 4 cm H_2O and once respiratory events appeared the pressure was increased by 1 cm H_2O every 2-5 minutes until an optimal pressure was reached to normalize the events and improve sleep architecture, assessed during the rapid eye movement phase and with the patient in a supine position. After 2-3 hours the pressure was steadily reduced until the minimum effective pressure was reached. The patients received detailed instructions on the use of nCPAP and the period of use was confirmed using a timer.

During the treatment period, contact was maintained with the patients in order to resolve potential problems or undesirable effects (particularly leaks). It was confirmed that an appropriate mask was employed, and confirmation was obtained on a monthly basis that there were no changes in BMI or smoking habit and that no new clinical signs had appeared. Treatment adherence was considered acceptable if the patient used nCPAP for at least 4 hours per night and on at least 75% of the nights during the study period. After 6 months, the ESS score was assessed again and a new blood sample was obtained under the same conditions as the first sample.

Analysis of NTproBNP and hsCRP

All patients went to bed at 11:00 PM and were woken at 8:00 AM. Samples of peripheral venous blood were obtained at 8:00 AM—after completion of the polysomnography recording and before the patient got out of bed—in standard collection tubes (Li- and NH^+_4 heparin) and stored at –80°C prior to analysis. Concentrations of NTproBNP were determined by immunoassay using the Elecsys proBNP kit for the Roche Elecsys 2010 analyzer (Roche Diagnostics, Mannheim, Germany). This electrochemoluminescence immunoassay contains monoclonal antibodies that recognize epitopes in the N-terminal region (1-76) of proBNP (1-108), using a 2-step sandwich technique involving streptavidin-coated microparticles that allows concentrations of at least 5.0 pg/mL to be detected. The coefficient of variation for within-run precision was 1.8% and the conversion factor, pg/mL × 0.118 = pmol/L.

The plasma concentrations of hsCRP were measured by latex microparticle-enhanced immunoturbidimetry (Tina-quant) using a Roche/Hitachi 717 automatic analyzer (Boehringer-Mannheim, Mannheim, Germany), allowing a detection limit of 0.003 mg/dL. The within-run and between-run coefficients of variation were 1.3% and 5.7%, respectively. Comparison of hsCRP determination using this method with the results obtained by nephelometry showed a correlation coefficient of $r = 0.996$. The conversion factor was mg/dL × 0.01 = g/L.

Statistical Analysis

Quantitative variables were expressed as means (SD) and qualitative data as percentages. Differences between groups were analyzed by Mann-Whitney test and differences in proportions were analyzed by χ^2 test. The normal distribution of different variables was assessed by Kolmogorov-Smirnov test. The concentrations of NTproBNP and hsCRP between the 3 groups (control, mild-to-moderate SAHS, and severe SAHS) were compared by analysis of variance (ANOVA) with Bonferroni correction for multiple comparisons. Correlations were assessed using the Pearson correlation coefficient. To assess the relative strength of the association between the concentrations

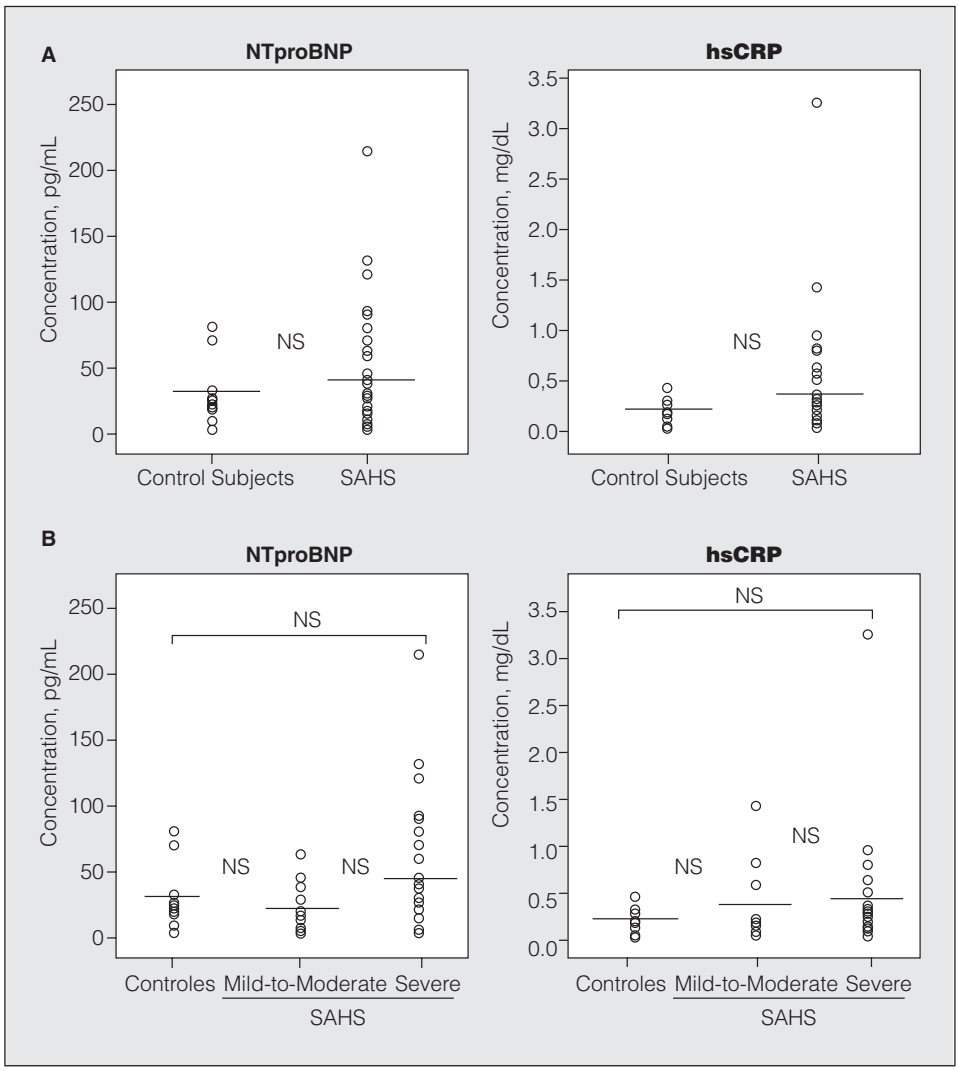

Figura 1. Serum concentrations of N-terminal prohormone brain natriuretic peptide (NTproBNP) and high-sensitivity C-reactive protein (hsCRP). A) Serum concentrations of NTproBNP and hsCRP in control subjects (n=14) and patients with sleep apnea-hypopnea syndrome (SAHS, n=42). B) Serum concentrations of NTproBNP and hsCRP in control subjects (n=14) and patients with mild-to-moderate (n=15) and severe SAHS (n=27). NS indicates not significant. Horizontal lines indicate mean values.

of NTproBNP or hsCRP and the possible contributing factors, multiple regression analysis was performed using all patients with SAHS as a single group. The concentrations of biomarkers before and after treatment with nCPAP were compared with the *t* test for paired data. Statistical analyses were performed using the SPSS statistical package version 14.0 (SPSS Inc, Chicago, Illinois, USA) and statistical significance was established at $P<.05$.

Results

After adjustment for BMI, age, and smoking habit, the mean concentrations of NTproBNP were not significantly different between the 3 groups (ANOVA, $P=.174$). The mean (SD) serum concentrations of NTproBNP were not significantly higher in patients with SAHS (38.28 [43.71] pg/mL) than in control subjects (31.7 [25.1] pg/mL) ($P=.597$, Figure 1A). The titers of NTproBNP were also not significantly higher in patients with severe SAHS (46.50 [51.70] pg/mL) than in control subjects (31.7 [25.1] pg/mL, $P=.321$) or patients with mild-to-moderate SAHS (23.49 [16.34] pg/mL, $P=.103$) (Figure 1B).

There were also no statistically significant differences in hsCRP concentration between the 3 groups (ANOVA, $P=.491$). Concentrations were not significantly higher in patients with SAHS (0.39 [0.56] mg/dL) than in control subjects (0.20 [0.12] mg/dL) ($P=.235$, Figure 1A). The concentrations were also not significantly higher in patients with severe SAHS (0.39 [0.61] mg/dL) than in control subjects (0.20 [0.12] mg/dL, $P=.257$) or patients with mild-to-moderate SAHS (0.37 [0.47] mg/dL, $P=.894$) (Figure 1B).

Table 2 shows the Pearson correlation coefficients between NTproBNP or hsCRP concentration and age, BMI, systolic and diastolic blood pressure, metabolic variables, polysomnographic variables, and ESS scores for patients with SAHS. The concentrations of NTproBNP were positively correlated with age, serum glucose concentration, AHI, and the percentage of time with an

arterial oxygen saturation (SaO_2) below 90% (r=0.37, P<.01) (Figure 2), and they were negatively correlated with mean nighttime SaO_2. Interestingly, the concentrations of hsCRP were not correlated with any of the variables analyzed.

In addition, BMI was positively correlated with AHI (r=0.60, P=.0001), arousals index (r=0.46, P=.001), and the proportion of time with an SaO_2 below 90% (r=0.37, P=.008), and negatively correlated with mean nighttime SaO_2 (r=–0.37, P=.0001).

The Pearson correlation coefficients between the concentrations of NTproBNP or hsCRP and age, blood pressure, metabolic variables, polysomnographic variables, and ESS score in the control subjects showed that the concentrations of hsCRP were not correlated with any of the variables considered and the concentration of NTproBNP was negatively correlated only with BMI (P=.0006).

The Pearson correlation coefficients between the same variables were calculated for all subjects (Table 3). There was a statistically significant correlation between the concentration of hsCRP and the proportion of time with an SaO_2 below 90% (r=0.28, P=.03). A similar correlation was observed for the concentration of NTproBNP (r=0.34, P=.012), which was also negatively correlated with the mean nighttime SaO_2 (r=–0.31, P=.01).

To identify independent predictors of variations in the concentrations of NTproBNP and hsCRP in patients with SAHS, stepwise multiple linear regression was carried out and the following independent variables were included in the model: age, BMI, smoking habit, blood pressure, metabolic variables, AHI, arousals index, desaturation index, percentage of time with an SaO_2 below 90%, mean nighttime SaO_2, and ESS score. The strongest predictor

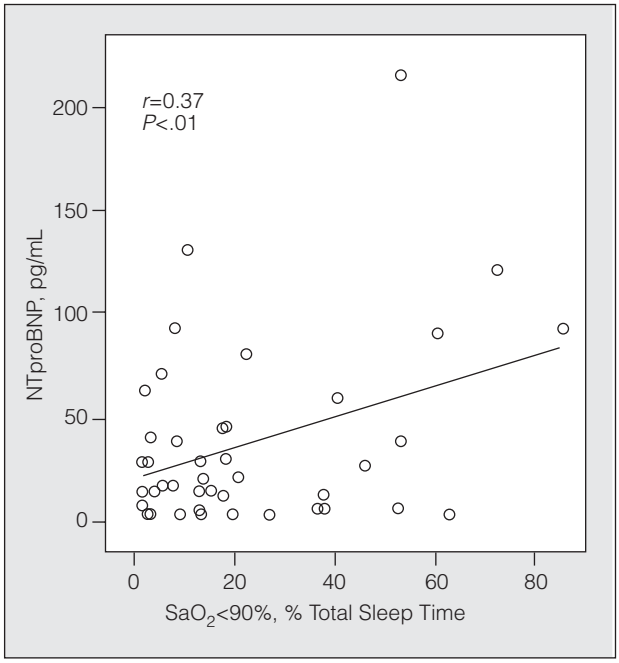

Figure 2. Correlation between the serum concentrations of N-terminal prohormone brain natriuretic peptide (NTproBNP) and the percentage of time with an arterial oxygen saturation (SaO_2) less than 90% in patients with sleep apnea-hypopnea syndrome (n=42).

TABLE 2
Correlation Coefficients Between Concentrations of NTproBNP or hsCRP and Age, BMI, Blood Pressure, Metabolic Variables, Polysomnographic Variables, and Epworth Score in Patients With SAHS (n=42)

	NTproBNP	*P*	hsCRP	*P*
Age	0.31	.02[a]	0.25	.05
BMI	0.13	.19	–0.05	.37
Systolic blood pressure	–0.18	.12	–0.16	.15
Diastolic blood pressure	0.25	.05	0.07	.32
Glucose	0.28	.04[a]	0.18	.13
Total cholesterol	–0.12	.37	–0.12	.24
HDL-C	0.06	.33	–0.25	.05
LDL-C	–0.14	.30	–0.02	.44
Triglycerides	0.06	.35	–0.02	.43
AHI	0.29	.03	0.03	.12
Desaturation index	0.08	.58	0.03	.84
Arousals index	0.12	.22	–0.02	.44
SaO_2<90%	0.37	.009[a]	0.24	.06
Mean nighttime SaO_2	–0.36	.01[a]	–0.14	.18
Epworth score	0.04	.41	0.03	.33

Abbreviations: AHI, apnea-hypopnea index; BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; hsCRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol; NTproBNP, N-terminal prohormone brain natriuretic peptide; SAHS, sleep apnea-hypopnea syndrome; SaO_2, arterial oxygen saturation; SaO_2<90%, percentage of time with SaO_2<90%.
[a]Statistically significant.

TABLE 3
Correlation Coefficients Between Concentrations of NTproBNP or hsCRP and Age, BMI, Blood Pressure, Metabolic Variables, Polysomnographic Variables, and Epworth Score in All Study Subjects

	NTproBNP	*P*	hsCRP	*P*
Age	0.32	.01[a]	0.24	.06
BMI	0.04	.76	–0.03	.80
Systolic blood pressure	–0.09	.47	–0.16	.23
Diastolic blood pressure	0.22	.10	0.07	.60
Glucose	0.24	.07	0.17	.20
Total cholesterol	–0.05	.71	–0.04	.72
HDL-C	0.04	.73	–0.24	.07
LDL-C	–0.05	.66	0.04	.74
Triglycerides	0.07	.60	–0.01	.90
AHI	0.26	.05	0.12	.37
Desaturation index	0.07	.58	0.07	.57
Arousals index	0.14	.27	0.07	.60
SaO_2<90%	0.34	.01[a]	0.28	.03[a]
Mean nighttime SaO_2	–0.31	.01[a]	–0.19	.14
Epworth score	0.08	.53	0.12	.36

Abbreviations: AHI, apnea-hypopnea index; BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; hsCRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol; NTproBNP, N-terminal prohormone brain natriuretic peptide; SAHS, sleep apnea-hypopnea syndrome; SaO_2, arterial oxygen saturation; SaO_2<90%, percentage of time with SaO_2<90%.
[a]Statistically significant

of variation in the concentration of NTproBNP was the proportion of time with SaO_2 below 90% (*P*=.017), which accounted for 37% of the variance.

Interestingly, there were no strong predictors of variation in the concentration of hsCRP. In another model in which the proportion of time with SaO_2 below 90% was excluded, the most powerful predictor of the plasma concentration of NTproBNP was the mean nighttime SaO_2 (*P*=.021), which accounted for 36% of the variance. Again, there were no predictors of variation in the concentration of hsCRP in this model (Table 4).

Finally, the same variables were analyzed as predictors of NTproBNP and hsCRP concentration in all groups considered together (n=56) via linear regression analysis. The proportion of time with SaO_2 below 90% was identified as the strongest predictor of NTproBNP concentration (*P*=.012), accounting for 34% of the variance, and also of the concentration of hsCRP (*P*=.036), in this case accounting for 28% of the variance.

Effects of Treatment With nCPAP

In the patients with severe SAHS, BMI did not change significantly and no new infectious or cardiovascular diseases were detected during the 6 months of treatment with nCPAP. The mean pressure of nCPAP was 7.76 (1.57) cm H_2O and the mean duration of nighttime use was 6.30 (1.33) hours. The AHI during polysomnographic titration of nCPAP changed significantly from 64.4 (22.9) to 6.8 (1.1) (*P*<.001). The ESS score also fell, from 10.6 (4.5) before treatment to 6.3 (1.4) after treatment (*P*<.001). Six patients were excluded from the study as their mean nighttime use of nCPAP was less than 4 hours (remaining group, n=21). Treatment with nCPAP did not lead to a statistically significant reduction in the concentration of NTproBNP (51.86 [12.42] pg/mL before treatment and 43.44 [11.71] pg/mL after treatment, *P*=.53) or hsCRP (0.41 [0.50] mg/dL before treatment and 0.22 [0.03] mg/dL after treatment, *P*=.21) (Figure 3).

Discussion

The main finding of this study is that the baseline concentrations of NTproBNP and hsCRP do not differ significantly between patients with SAHS when grouped according to AHI. Our results also show that there is no clear association with inflammatory markers such as CRP once the presence of undiagnosed silent myocardial infarction and other heart disease has been excluded and, furthermore, that known risk factors affecting the secretion of the biomarkers have a disproportionate influence on the association with SAHS. In patients with SAHS, however, the concentrations of NTproBNP are related to the severity of oxyhemoglobin desaturation, independently of potential confounding factors such as age, BMI, smoking, blood pressure, blood glucose levels, lipid concentrations, and other polysomnographic variables. Although this relationship is indicative of the noxious hemodynamic effects of nighttime oxygen desaturation, treatment with nCPAP did not cause a significant reduction in NTproBNP concentration in these patients, making it difficult to establish a causal relationship between SAHS and cardiovascular risk.

Previous studies have found that SAHS is associated with left ventricular dysfunction[20-22] and inflammatory processes,[14,15] but the presence of moderate-to-severe obesity and possible confounding factors may have influenced the results obtained. Obesity also increases the probability of developing risk factors for heart disease, especially hypertension, hypercholesterolemia, and diabetes.

TABLE 4
Stepwise Multiple Regression Analysis of Independent Predictors of Serum Concentrations of NTproBNP and hsCRP in Patients With SAHS

Predictors	NTproBNP			hsCRP		
	β	*r*	*P*	β	*r*	*P*
Age	0.232	0.241	.128	0.250	0.250	.110
BMI	–0.002	–0.002	.991	–0.051	–0.051	.748
Smoking habit	–0.029	–0.031	.848	–0.073	–0.073	.645
Systolic blood pressure	–0.181	–0.194	.223	0.176	0.176	.265
Diastolic blood pressure	0.194	0.205	.199	–0.120	–0.120	.451
Glucose	0.181	0.185	.247	–0.246	–0.246	.116
Total cholesterol	–0.022	–0.023	.886	–0.022	–0.023	.886
HDL-C	0.052	0.056	.730	–0.028	–0.028	.860
LDL-C	–0.054	–0.057	.721	–0.054	–0.057	.721
Triglycerides	–0.084	–0.090	.575	–0.084	–0.090	.575
AHI	0.112	0.097	.545	0.026	0.026	.868
Desaturation index	–0.134	–0.118	.462	–0.023	–0.023	.887
Arousals index	–0.298	–0.236	.138	0.032	0.032	.842
SaO_2<90%	–0.366	–0.366	.017	0.239	0.239	.127
Mean nighttime SaO_2	–0.124	–0.055	.733	–0.142	–0.142	.368
Epworth score	–0.022	–0.023	.885	0.066	0.066	.680

Abbreviations: AHI, apnea-hypopnea index; BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; SAHS, sleep apnea-hypopnea syndrome; SaO_2, arterial oxygen saturation; SaO_2<90%, percentage of time with SaO_2<90%.

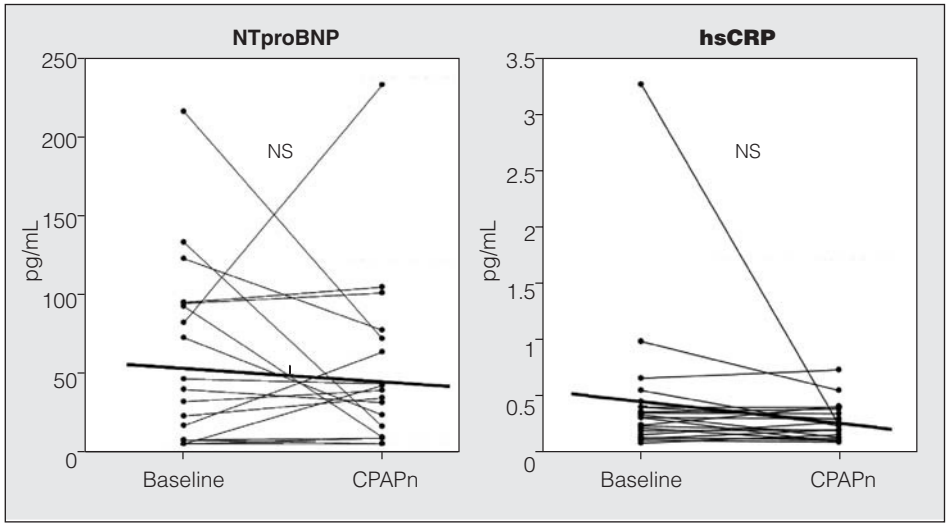

Figure 3. Effect of nasal continuous positive airway pressure (nCPAP) on the concentrations of N-terminal prohormone brain natriuretic peptide (NTproBNP) and high-sensitivity C-reactive protein (hsCRP) in patients with severe sleep apnea-hypopnea syndrome (n=21) treated for 6 months. There were no significant differences between the concentrations of NTproBNP and hsCRP before and after treatment. NS indicates not significant. Horizontal lines indicate mean values.

Left ventricular dysfunction may occur in the context of coronary heart disease, hypertension, valvular heart disease, primary myocardial heart disease, or diabetes mellitus. It has therefore been suggested that SAHS could also be an independent cause of this condition. Various studies have found that asymptomatic diastolic dysfunction is common in patients with severe SAHS,[5,6,20] but there are difficulties associated with the use of echocardiography (currently the gold standard for diagnosis of the condition), due to the obesity of the patients and the limited availability of the technique in primary health care centers. If diastolic dysfunction is left untreated and progresses, the risk of death is high. In addition, although diastolic heart failure is common, it is difficult to diagnose and associated with SAHS. Determination of the concentration of NTproBNP may improve the ability of clinicians to detect heart disease in this clinical context. BNP and proBNP are specifically secreted by the ventricles in response to volume expansion and pressure overload, but also in myocardial hypoxia. We found that the concentrations of NTproBNP did not differ significantly according to the severity of SAHS in patients classified according to AHI. This observation confirms the findings of Svatikova et al[23] and Tasci et al[24] when known factors that could alter cardiac secretion of NTproBNP were excluded. However, the extent of nighttime hypoxia seems to be the most powerful predictor of variations in the concentration of NTproBNP in patients with SAHS. These results may be consistent with the notion that acute myocardial hypoxia, even in the absence of ventricular dysfunction, activates cardiac *BNP* gene expression and increases the concentrations of BNP and proBNP, as reported by Goetze et al.[25,26]

There is also evidence of a correlation between NTproBNP and blood glucose concentration, which may be more a characteristic of obesity[27] than of SAHS itself; it may also be linked to age,[28] since this can affect the secretion of proBNP by influencing the characteristics of the ventricular myocardium.

The concentrations of NTproBNP in control subjects who were overweight or obese displayed a significant negative correlation with BMI, confirming the previously reported association between high BMI and low BNP concentration in healthy obese individuals.[29] We should therefore consider whether the cutoff for elevated serum concentrations of NTproBNP is considerably lower in obese or overweight individuals, leading to underestimation of the predictive value of this biomarker.

The results of our study contrast with the findings of Shamsuzzaman et al,[14] Yokoe et al,[15] and Can et al,[30] who reported that CRP concentrations are independently associated with SAHS severity. One possible explanation for the lack of statistically significant differences in our study is that the patients and control subjects used in previous studies[16,31] were far more obese than those considered here. Recently, Ryan et al[32] reported that CRP concentration is independently associated with obesity. Also, the impact of nCPAP on the serum concentration of CRP is still not clear.[33]

The hypoxia due to apnea may also have been less severe in our patients than in those of previous studies.[34] Most studies did not provide detailed information on comorbidity and treatment, which could have affected circulating concentrations of CRP. Unlike previous authors, we found no statistically significant correlation between BMI, blood pressure, metabolic variables, or polysomnographic variables and the concentration of CRP. In addition, stepwise multiple regression analysis revealed that these variables were not associated with increased CRP concentration in patients with SAHS, though the size of our sample may have been too small to allow detection of subtle differences in this biomarker. The correlations observed when continuous variables were considered in all subjects differed from those for patients with SAHS, although the correlation coefficients indicated a wide dispersion in the data obtained.

Ryan et al[35] highlighted the potential role of sleepiness in mediating the inflammatory process in SAHS. However, their data did not allow conclusions to be drawn on whether it was a cause or a consequence of elevated proinflammatory cytokines. In addition, although treatment with nCPAP

had a significant effect on nuclear factor κB-dependent genes (principally tumor necrosis factor α, which is also associated with excessive daytime sleepiness), it had no effect on the other inflammatory markers studied. In that study, patients with SAHS and control subjects were grouped according to AHI, and significant differences were found in the degree of sleepiness in the groups with SAHS. In our sample, the ESS scores were not significantly different between the different groups with SAHS.

Furthermore, in contrast to the findings of Tasci et al[24] and Yokoe et al,[15] we did not observe significant differences in the concentrations of NTproBNP or hsCRP in patients with severe SAHS. However, a later reduction (although not statistically significant) was observed in the concentrations of NTproBNP and hsCRP after treatment with nCPAP for 6 months, a longer period than that employed by Pepperell et al[36]; this difference may be attributable to a reduction in ventricular irritability.[37] The effects of nCPAP on the concentrations of NTproBNP and hsCRP would need to be assessed in a large placebo-controlled trial.

Our findings have implications for the stratification of patients with SAHS according to cardiovascular risk factors. This suggests that the impact of SAHS on cardiovascular disease, as reflected by the concentration of NTproBNP, can vary considerably depending on the degree of nighttime oxyhemoglobin desaturation. In contrast, the absolute values of AHI and their relationship with changes in sleep architecture could have a considerably lesser impact on patients' cardiovascular risk profile as reflected by inflammatory (CRP) and hemodynamic (NTproBNP) stress. In our opinion, the different effect of nighttime oxygen desaturation and the frequency of apnea-hypopnea events on NTproBNP indicates that cardiovascular risk profiles differ according to whether the phenotypic expression of SAHS is characterized predominantly by sleep fragmentation or severe nighttime oxygen desaturation. Differences between our results and those reported previously may also be explained by inadequate characterization of heterogeneous groups of patients with SAHS or alteration of hemodynamic and inflammatory responses as a result of confounding factors.

In conclusion, we have shown that the severity of nighttime hypoxemia in SAHS is correlated with NTproBNP concentrations but not with serum concentrations of hsCRP, and that, although a long-term effect cannot be ruled out, these markers are not reduced significantly by nCPAP in the short term. Consequently, although SAHS could not be considered a principal cardiovascular risk factor, it may exacerbate known cardiovascular changes associated with nighttime hypoxemia, which is a major cause of variations in the plasma concentration of NTproBNP in patients with SAHS.

Our study has certain limitations. Firstly, patients with known cardiovascular disease or diseases and factors that predispose to cardiovascular disease were excluded, making it possible that those patients with SAHS who were most predisposed to present cardiovascular disease and elevated concentrations of these biomarkers were excluded in the process. This possibility would make it more difficult to observe statistically significant differences between the groups. In addition, since the subjects were less obese than those included in previous studies, the oxygen desaturation associated with apnea events would have been less, as indicated by the proportion of time with an SaO_2 below 90%, which was found to be lower in our study. This difference could have resulted in CPAP not bringing about a significant reduction in the concentration of these biomarkers even though the hypoxemia was corrected. Another explanation could be that our patients with SAHS were less symptomatic than those in other studies and that, as a result, the treatment was less effective.

Overall, the most significant limitation of our study is that the application of numerous exclusion criteria led to a small sample size that prevented type-2 errors being avoided. Nevertheless, our results were similar to those of the most recent studies, which differ from earlier studies of CRP, since they also controlled for the major confounding factors.

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