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Influence of treatment on leptin levels in patients with obstructive sleep apnoea

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Influence of treatment on leptin levels in patients with obstructive sleep apnoea. B.M. Sanner, P. Kollhosser, N. Buechner, W. Zidek, M. Tepel. ©ERS Journals Ltd 2004.

ABSTRACT: Obstructive sleep apnoea syndrome (OSAS) is a common disorder in obesity. Leptin, an adipocyte-derived signalling factor, plays an important role in metabolic control. There is growing evidence that leptin regulation is altered in OSAS. Therefore, the aim of this study was to test the hypothesis that effective treatment will influence leptin levels in OSAS patients.

Serum leptin levels were determined in 86 consecutive patients (aged 57.5±11.0 yrs) with polysomnographically verified OSAS. In addition, leptin levels were reassessed and treatment efficacy was evaluated by polysomnography after 6 months of therapy.

Patients were treated with continuous or bilevel positive airway pressure, a mandibular advancement device or conservatively, depending on the clinical symptoms. Mean serum leptin levels did not change with treatment in the whole study group (7.3±5.0 *versus* 7.5±4.8 ng·mL^{-1}), however, leptin levels decreased in effectively treated patients (8.5±5.0 *versus* 7.4±5.1 ng·mL^{-1}) while they increased in ineffectively treated patients (5.0±4.0 *versus* 7.7±4.1 ng·mL^{-1}). Furthermore, not only was there a significant and independent correlation between the change in leptin levels with treatment and the change in body mass index, but also with the change in apnoea/hypopnoea index.

Effective treatment of sleep-disordered breathing may have significant effects on leptin levels in obstructive sleep apnoea syndrome patients. Changes in leptin levels are related to changes in apnoea/hypopnoea index in obstructive sleep apnoea syndrome patients.

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Obstructive sleep apnoea syndrome (OSAS) is a common disorder with an estimated prevalence of at least 2–4% among middle-aged adults [1]. Recently, there has been growing interest in potential relationships between OSAS and leptin regulation. Leptin is an adipocyte-derived signalling factor, the circulating levels of which reflect energy stores, and seems to play an important role in metabolic control, reproduction and neuroendocrine signalling [2]. Plasma leptin levels have been found to be closely correlated with the size of the adipose tissue depot [3]. There is increasing evidence that in obesity, a common disorder in OSAS patients [4], the regulation of leptin secretion can be disturbed by several mechanisms. Specifically, polymorphisms of the β_3 receptor have been identified, which may result in an impaired suppression of leptin secretion after β_3 receptor activation and which are more prevalent in obese subjects [5].

One study published recently was able to demonstrate that sleep apnoeic males have higher plasma leptin levels than non-apnoeic age- and body mass index (BMI)-matched obese males [6]. Furthermore, leptin levels decreased significantly following treatment of OSAS [7], even without a significant change of body weight [8].

Until now, the association between OSAS and leptin homeostasis has not been fully understood. Therefore, the aim of this study was to test the hypothesis that effective treatment will influence leptin levels in OSAS patients.

Methods

A total of 100 consecutive patients referred to the current authors' sleep laboratory for snoring, suspected OSAS or excessive daytime sleepiness were selected for the study. There were no shift workers in the population.

Males and females were eligible for the study if they were ⩾21 yrs old and had polysomnographically verified OSAS with an apnoea/hypopnoea index (AHI) of more than or equal to five per hour of sleep. Use of sedatives and muscle relaxants, or refusal to participate in the study, were exclusion criteria.

Weight was taken in light clothing and during the fasting state with an electronic scale. None of the subjects reported significant strenuous physical activity within a period of 72 h before testing.

Blood was drawn at 08:00 h after an overnight fast at baseline and at 6 months after study initiation in the morning after polysomnography in the sleep laboratory. Serum leptin levels were determined by the radioimmunoassay for human leptin (Mediagnost, Tübingen, Germany). The leptin levels were measured blind to the severity of OSAS.

Polysomnography

All patients underwent overnight polysomnography (Somnostar 4100; SensorMedics Co., Yorba Linda, CA, USA)

according to widely accepted methods [9]. The entire recording was supervised by a technician.

Polysomnography records were scored in 30-s periods for sleep, breathing and oxygenation. According to the commonly used clinical criteria [10], a breathing event during objectively measured sleep was defined as abnormal if either a complete cessation of airflow lasting $\geqslant$10 s took place (apnoea), or a reduction in respiratory airflow of $\geqslant$50% of the airflow lasting longer than 10 s associated with either an arousal or a desaturation of >3% could be discerned (hypopnoea). Obstructive apnoea was defined as absence of airflow in the presence of chest-wall motion. The average number of episodes of apnoea and hypopnoea per hour of sleep (AHI) was calculated. OSAS was diagnosed when the AHI was $\geqslant 5 \cdot h^{-1}$, associated with typical clinical features. Sleep was staged manually using the methods of RECHTSCHAFFEN and KALES [11].

A further sleep study was subsequently performed to determine the continuous positive airway pressure (CPAP) required to abolish snoring, apnoeas and oxygen desaturation in those patients who were eligible for CPAP treatment.

Patients who refused CPAP therapy were offered a mandibular advancement device (for details see [12]) or a "conservative treatment" (sleep hygiene and dietetic measures).

Follow-up

Blood samples were taken the morning after baseline polysomnography. During follow-up, all patients were seen by their home physicians after 1 and 3 months. Patients were asked to continue taking all prescribed medication for cardiac and other conditions that they had been receiving at baseline. Furthermore, the patients were advised to retain their usual behaviours, including diet and daily activity.

After 6 months, the patients were re-evaluated by polysomnography and blood samples were taken again the following morning. Compliance with prescribed CPAP or bilevel therapy was based on the actual hours of CPAP or bilevel usage as registered by an integrated hour meter. The study protocol was approved by the local ethics committee. Written informed consent was obtained from all patients before entry into the study.

Statistical analyses

Results are presented as means±SD. All p-values reported are two-tailed. Inter-group differences were analysed for significance with the paired t-test, with Bonferroni's correction for multiple comparisons.

The relationships between the leptin values and parameters indicative of the severity of OSAS were first explored by bivariate regression analysis. To determine the independent association of leptin values with OSAS in the presence of other significant risk factors, stepwise multiple logistic regression analysis was performed in a second step. The multivariate statistical model was built in steps and was designed to select only factors that correlated with the serum leptin values at a level of significance of $\leqslant$0.05 for the final multiple regression model.

Results

From the 100 patients initially selected, one refused to participate and OSAS could not be verified by polysomnography in 13. Thus, the results are based on the remaining 86 patients (69 males, 17 females). The mean age of the study group was 57.5±11.0 yrs (range 30–96) and the mean BMI was 31.2±5.6 $kg \cdot m^{-2}$ (19.0–52.9). All patients had a polysomnographically verified OSAS with a mean AHI of 28.0±19.5·h of $sleep^{-1}$ (5.3–89.6), a minimum oxygen saturation during sleep of 81.5±9.4% (51.3–86.9), a mean oxygen saturation during sleep of 92.8±3.7% (77.2–98.0) and a disturbed sleep structure.

Forty-eight of the patients (55.8%) had a known or newly diagnosed systemic hypertension, 25 patients had diabetes mellitus (29.1%), 10 patients had chronic obstructive pulmonary disease (11.6%) and 20 patients had coronary artery disease (23.3%). Hypercholesterolaemia (total cholesterol $\geqslant 5.2\ mmol \cdot L^{-1}$) was present in 72 (83.7%) and hypertriglyceridaemia (triglyceride level $\geqslant 1.8\ mmol \cdot L^{-1}$) was found in 41 patients (47.7%). Forty-three of the hypertensive patients were treated with antihypertensive agents (27 with angiotensin converting enzyme inhibitors, 24 with calcium channel blockers and 20 with a β-blocking agent), 13 patients were treated with a diuretic agent, seven with theophylline, 13 with a nitrate, one with digitalis, five with insulin and three with an oral hypoglycaemic agent.

Depending on the clinical symptoms, the polysomnographical result and the acceptance of therapy, seven patients were treated conservatively, 11 patients received a mandibular advancement device and 68 patients were treated with continuous or bilevel positive airway pressure.

Polysomnography

Control polysomnography after 6 months of treatment documented improvement of OSAS in the whole study group, while BMI did not change during follow-up (table 1).

Table 1. – Effect of therapy on body mass index (BMI) and polysomnographic data in the whole study group

	Before treatment	After treatment	Statistical significance
BMI $kg \cdot m^{-2}$ #	31.2±5.6	31.2±5.6	NS
Total sleep time min	386.4±57.2	387.4±52.1	NS
Sleep efficiency %	87.4±10.1	89.5±9.5	NS
Stage wake %	9.1±8.9	7.1±7.8	NS
Sleep stages 1+2 %	66.0±13.6	64.9±11.1	NS
Sleep stages 3+4 %	13.7±10.4	14.4±7.5	NS
Sleep stage REM %	10.8±6.3	12.7±5.9	<0.05
AHI $n \cdot h^{-1}$	28.0±19.5	5.2±8.2	<0.001
Minimum nocturnal Sa,O_2 %	81.5±9.4	87.2±10.5	<0.001
Mean nocturnal Sa,O_2 %	92.7±3.7	94.1±2.2	<0.001

Data are presented as means±SD. #: weight (kg) divided by height^2 (m^2). REM: rapid eye movement; AHI: apnoea/hypopnoea index; Sa,O_2: arterial oxygen saturation; NS: nonsignificant.

Objectively measured compliance documented an average use of CPAP or bilevel devices of 5.2±1.4 h·night^{-1} (1.1–7.6). CPAP compliance in the 2–3 nights immediately preceding the serum leptin measurement was not different from the overall compliance. All patients treated with the mandibular advancement device tolerated this treatment well and reported daily use of the appliance.

Leptin levels

In the whole study group, mean serum leptin levels did not change with treatment (7.3±5.0 *versus* 7.5±4.8 ng·mL^{-1}, p=0.65). Treatment was defined effective if the remaining AHI was <5·h^{-1}. According to this definition, 59 patients (one patient with conservative treatment, three patients with a mandibular advancement device, 53 patients with CPAP and two patients with bilevel positive airway pressure therapy) were treated effectively. Their AHI decreased from 29.4±20.3 to 1.6±1.3·h^{-1} (p<0.001). In this subgroup there was also a decrease in serum leptin levels (8.5±5.0 *versus* 7.4±5.1, p<0.05). In contrast, in the ineffectively treated group (n=26), serum leptin levels increased (5.0±4.0 *versus* 7.7±4.1, p=0.01), although the AHI decreased significantly (24.8±17.7 *versus* 13.7±10.6, p<0.01).

There was no significant correlation between the change in leptin levels and objectively measured compliance with CPAP or bilevel devices (r=0.06, p=0.61).

To clarify the relationship between OSAS and serum leptin levels, correlation analyses were performed. There were significant associations between baseline leptin levels and BMI (r=0.59, p<0.001) (fig. 1), minimum oxygen saturation during sleep (r=-0.27, p=0.01) and mean oxygen saturation during sleep (r=-0.22; p=0.04), indicating that more severe OSAS is associated with higher leptin levels, but not with age, AHI or time during sleep with an oxygen saturation <90%. In a multiple stepwise linear regression analysis (R^2=0.37), BMI proved to be the only parameter significantly and independently associated with baseline leptin levels (β=0.61, p<0.001). None of the other possible risk factors tested (mean and minimum oxygen saturation during sleep, AHI, time during sleep with an oxygen saturation <90%, age and sex) was included in the stepwise multiple logistic regression model because the level of significance was >0.05.

In a second step, the relationship between the improvement of nocturnal respiration and the change of leptin levels was examined. There was a significant correlation between the change in leptin levels and the change in the BMI (r=0.25, p=0.02), the change in the AHI (r=0.33, p=0.002) (fig. 2) and the change in the mean oxygen saturation during sleep (r=-0.23, p=0.04), but not with the minimum oxygen saturation during sleep.

In addition, multiple stepwise linear regression analysis was performed with the change of leptin levels as the dependent variable, and indicators of the change of severity of OSAS with treatment and change of BMI as independent variables. The regression analysis disclosed that both the change in the AHI (β=0.36, p=0.001) and the change in the BMI (β=0.27, p=0.01) were independently correlated with the change in leptin levels (R^2=0.18).

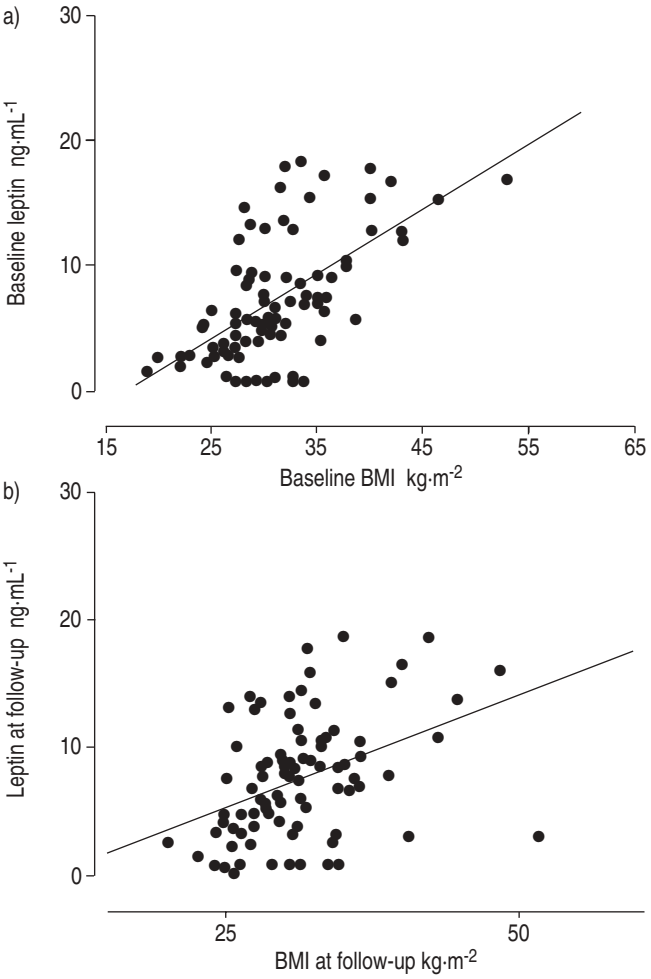

Fig. 1.–Body mass index (BMI) plotted against leptin levels a) at baseline (r=0.59, p<0.001) and b) at follow-up after 6 months (r=0.41, p<0.001).

Discussion

The most important finding in this study is that there is a relationship between the change in leptin levels and the degree of improvement of nocturnal respiration, as expressed by the AHI, independent of the change in BMI. Patients who are treated effectively with a resulting AHI of <5·h^{-1} show a decrease in their leptin levels, while leptin levels increase in ineffectively treated OSAS patients. This may indicate that OSAS contributes, at least in part, to elevated leptin levels, independent of obesity. Another important result of the present study is the fact that a ~50% reduction in the AHI in

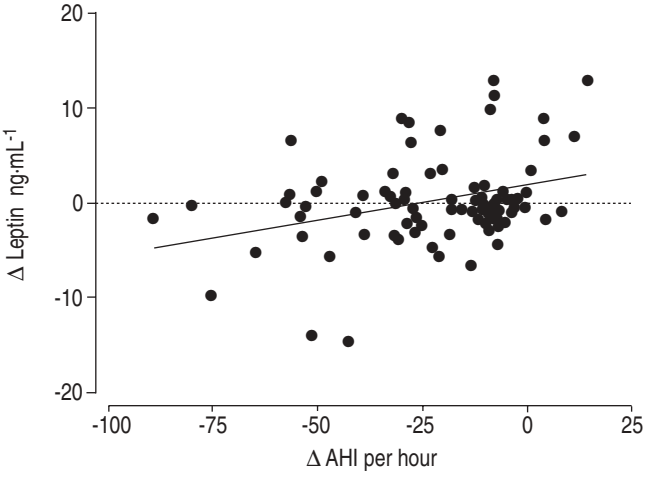

Fig. 2.–Change of leptin levels plotted against change of apnoea/hypopnoea index (AHI) (r=0.33, p=0.002).

the ineffectively treated group did not result in a decrease in leptin levels but even in a significant increase. This is in accordance with a recently published study examining the influence of effective and ineffective treatment of obstructive sleep apnoea on blood pressure [13], emphasising the importance of highly effective treatment.

Obesity is closely associated with increased morbidity and mortality. Most obese humans have increased leptin levels, indicating that obesity is a leptin-resistant state [3]. Interestingly, patients with OSAS have even higher leptin levels than age- and BMI-matched controls [6].

The measurement of respiratory function in the ob/ob mouse has shown that profound obesity is associated with impaired respiratory mechanics and depressed respiratory control, particularly during sleep, indicating that leptin may act on respiratory control mechanisms [14]. In accordance, hyperleptinaemia is associated with hypercapnic respiratory failure in human obesity [15]. In addition, the administration of leptin attenuated rapid breathing patterns and diminished lung compliance, suggesting that the administration of leptin in these leptin-deficient mice reduced hypoventilation [16]. Recently, PHILLIPS *et al*. [17] showed that OSAS is associated with resistance to the weight-reducing effects of leptin. As yet, the mechanisms that result in elevated leptin levels in OSAS patients are not understood.

Treatment of OSAS, for example with CPAP or with a mandibular advancement device, can normalise breathing during the night and reduce leptin levels [7, 8, 18]. The present study indicates that changes in leptin levels are related to changes in BMI and in AHI. In support of this finding, the study of CHIN *et al.* [8] indicated that the correction of breathing by CPAP reduced visceral fat. The results of the present study show that the decrease of leptin levels is highest in patients with a large reduction in AHI. This effect was independent of a change in the BMI. How can these findings be explained?

A study published recently could demonstrate that the magnitude of the decrease in leptin levels after CPAP treatment was significantly correlated with cardiac sympathetic function measured before CPAP treatment [19]. Therefore, it can be speculated that the physiological negative feedback control of leptin secretion by the sympathetic nervous system improves with treatment of OSAS. However, the mechanisms responsible are unclear. Furthermore, treatment of OSAS, especially with CPAP, may be associated with changes in haemodynamics with increases in lung volume or in abdominal pressure and perhaps visceral blood flow. This may change blood flow in the body and could, in turn, have an effect on the leptin clearance, thereby reducing leptin levels [8].

Lastly, it may be speculated that, besides the known pathways of leptin regulation, tissue and/or arterial oxygen tension may interfere with the feedback control of leptin secretion. Although the current authors are not aware of sound data indicating an effect of tissue oxygenation on leptin secretion, the current data are compatible with such a new type of regulatory control of serum leptin levels. Direct effects of hypoxia at high altitude on leptin levels have already been proposed [20]. Furthermore, there are some indirect hints of a relationship between tissue oxygenation and leptin secretion [21]. Therefore, it seems worthwhile to further examine the potential relationship between leptin secretion and oxygen supply to tissue.

References

1. Young T, Palta M, Dempsey J, Skatrud J, Weber S, Badr W. The occurrence of sleep-disordered breathing among middle-aged adults. *New Engl J Med* 1993; 328: 1230–1235.
2. Auwerx J, Staels B. Leptin. *Lancet* 1998; 351: 737–742.
3. Considine RV, Sinha MK, Heiman ML, *et al.* Serum immunoreactive-leptin concentrations in normal-weight and obese humans. *New Engl J Med* 1996; 334: 292–295.
4. Strollo PJ, Rogers RM. Obstructive sleep apnea. *New Engl J Med* 1996; 334: 99–104.
5. Endo K, Yanagi H, Hirano C, Hamaguchi H, Tsuchiya S, Tomura S. Association of Trp64Arg polymorphism of the beta3-adrenergic receptor gene and no association of Gln223Arg polymorphism of the leptin receptor gene in Japanese schoolchildren with obesity. *Int J Obes Relat Metab Disord* 2000; 24: 443–449.
6. Vgontzas AN, Papanicolaou DA, Bixler EO, *et al.* Sleep apnea and daytime sleepiness and fatigue: relation to visceral obesity, insulin resistance, and hypercytokinemia. *J Clin Endocrinol Metab* 2000; 85: 1151–1158.
7. Saarelainen S, Lahtela J, Kallonen E. Effect of nasal CPAP treatment on insulin sensitivity and plasma leptin. *J Sleep Res* 1997; 6: 146–147.
8. Chin K, Shimizu K, Nakamura T, *et al.* Changes in intra-abdominal visceral fat and serum leptin levels in patients with obstructive sleep apnea syndrome following nasal continuous positive airway pressure therapy. *Circulation* 1999; 100: 706–712.
9. American Thoracic Society – Medical Section of the American Lung Association. Indications and standards for cardiopulmonary sleep studies. *Am Rev Respir Dis* 1989; 139: 559–568.
10. American Academy of Sleep Medicine Task Force. Sleep-related breathing disorders in adults: recommendations for syndrome definition and measurement techniques in clinical research. *Sleep* 1999; 22: 667–689.
11. Rechtschaffen A, Kales A. A manual of standardized terminology, techniques and scoring system for sleep stages of human subjects. Washington, DC, U.S. Government Printing Office, 1968 (NIH publication No. 204).
12. Sanner B, Heise M, Knoben B, *et al.* Magnetic resonance imaging of the pharynx and treatment efficacy of a mandibular advancement device in obstructive sleep apnea syndrome. *Eur Respir J* 2002; 20: 143–150.
13. Becker HF, Jerrentrup A, Ploch T, *et al.* Effect of nasal continuous positive airway pressure treatment on blood pressure in patients with obstructive sleep apnea. *Circulation* 2003; 107: 68–73.
14. O'Donnell CP, Tankersley CG, Polotsky VP, Schwartz AP, Smith PC. Leptin, obesity, and respiratory function. *Respir Physiol* 2000; 119: 163–170.
15. Phipps PR, Starritt E, Caterson I, Grunstein RR. Association of serum leptin with hypoventilation in human obesity. *Thorax* 2002; 57: 75–76.
16. Tankersley CG, O'Donnell C, Daood MJ, *et al.* Leptin attenuates respiratory complications associated with the obese phenotype. *J Appl Physiol* 1998; 85: 2261–2269.
17. Phillips BG, Kato M, Narkiewicz K, Choe I, Somers VK. Increases in leptin levels, sympathetic drive, and weight gain in obstructive sleep apnea. *Am J Physiol Heart Circ Physiol* 2000; 279: H234–H237.
18. Ip MS, Lam KS, Ho C, Tsang KW, Lam W. Serum leptin and vascular risk factors in obstructive sleep apnea. *Chest* 2000; 118: 580–586.
19. Shimizu K, Chin K, Nakamura T, *et al.* Plasma leptin levels and cardiac sympathetic function in patients with obstructive sleep apnoea-hypopnoea syndrome. *Thorax* 2002; 57: 429–434.
20. Tschop M, Strasburger CJ, Hartmann G, Biollaz J, Bartsch P. Raised leptin concentrations at high altitude associated with loss of appetite. *Lancet* 1998; 352: 1119–1120.
21. Mise H, Sagawa N, Matsumoto T, *et al.* Augmented placental production of leptin in preeclampsia: possible involvement of placental hypoxia. *J Clin Endocrinol Metab* 1998; 83: 3225–3229.