Supplement contents

S1. Twenty-four-hour blood sampling

The blood was drawn during the fourth day and night in the sleep laboratory at baseline and was repeated in sleep apnea patients at the end of both CPAP and sham-CPAP treatment

periods (for more details see supplement). Twenty-four-hour blood sampling was performed serially, every 60 minutes for the assessment of blood IL-6 and TNFR1 levels, leptin and adiponectin. During the sleep periods, blood samples were obtained from an adjacent room through a perforation in the wall by connecting external tubing to the indwelling catheter. Single blood samples for measurement of fasting blood glucose and insulin levels were drawn in the morning following the overnight sleep recording.

S2. Assaying

Blood collected from the indwelling catheter was collected in EDTA-containing tube and refrigerated until centrifugation (within 3 hours). Blood was stored in a -80 C freezer until assay. Concentrations of IL-6, TNFR1 were measured every 60 minutes, leptin and adiponectin were measured every 120 minutes throughout the 24-hour period, CRP was measured twice (morning and evening) while single blood samples for the measurement of fasting blood glucose and insulin were drawn the morning following the overnight sleep recording. All samples were processed in the same manner. Plasma TNFR1, IL-6 and hsCRP concentrations were measured by ELISA (R&D Systems, Minneapolis, MN). The intra- and inter-assay coefficients of variation (CVs) were from 4.4 and 6.1 for TNFR1, from 7.4 and 7.8 for IL-6 and from 5.5 and 11.6 for hsCRP respectively. The lower detection limits for TNFR1, IL-6 and hsCRP were 0.043 pg/ml, 0.094 pg/ml and 0.124 mg/l respectively. Adiponectin was measured by a commercially available radioimmunoassay. The intra- and inter-assay CVs were 4.0 and 8.1 respectively, and the minimal detection limit was 1 ng/ml. Insulin was determined with a double antibody method using reagents obtained from Millipore Co. (Billerica, MA). The sensitivity of this assay was 2 μU/ml and the inter- and intra-assay CVs were less than 10%.

S3. CPAP and sham-CPAP usage

All patients with sleep apnea underwent consecutively CPAP and sham-CPAP treatment in a random counterbalanced order. The optimal nasal CPAP pressure was determined during a full night polysomnographic study as the pressure necessary to abolish all respiratory events and snoring, secondary arousals and episodes of SaO_2 desaturation during REM sleep and in the supine position. During the sham-CPAP phase, patients were given an identical machine (S8 Escape, ResMed, USA) but the pressure derived was set at the level of 1 cm H_2O when used with a modified mask. The mask (Ultra Mirage II Nasal mask) was modified by adding an airflow restrictor and by increasing the amount of exhalation ports. Moreover the airflow through the exhalation port and the operating noise in the sham CPAP was virtually identical to those of active CPAP which allowed a successful replication. To ensure CPAP adherence, CPAP usage was monitored closely on a daily basis by calculating the time the patient was breathing through the machine and not just the time the machine was on. A respiratory therapist visited the home of each patient regularly to provide us information regarding CPAP usage (number of hours used daily, pressure settings and mask leakage) and reinforce adherence. The respiratory therapist continued to visit patients following the same protocol during the sham-CPAP phase. As regular users of CPAP were considered those who used the apparatus for at least 4 hours each night, five or more nights a week.

S4. CPAP compliance: 'low' versus 'high' adherence to therapy

We further created two groups according to our group's median of CPAP daily use in order to better explore the effect of CPAP use on the variables of interest. The 'low adherence' group used the CPAP less than 6 hours while the 'high adherence' group used the CPAP equally or more than 6 hours per night. In both groups respiratory variables (AHI and minimum SaO_2) improved significantly compared to baseline and sham-CPAP. No significant change in BMI,

CRP, IL-6, TNFR1, HOMA index, leptin and adiponectin was observed either in the ‘low’ or ‘high’ CPAP adherence group. Epworth Sleepiness scale total score improved significantly while average MSLT sleep latency was practically unchanged in both groups. In terms of the PVT variables, number of lapses and median RT tended to improve after CPAP compared to baseline in the ‘high adherence’ group, while number of lapses and median RT tended to increase and increased significantly in the ‘low adherence’ group respectively.

S5. Table S5. Order effect by ANOVA for 2x2 crossover study

	p
BMI (kg/m^2)	0.21
AHI	0.34
Minimum O_2 Saturation	0.98
Log hsCRP (ng/ml)	0.48
log IL-6 (pg/ml)	0.08
TNFR1 (pg/ml)	0.84
logLeptin (ng/ml)	0.44
Adiponectin (ng/ml)	0.66
HOMA	0.48
Epworth Sleepiness Scale	0.44
MSLT (min)	0.43
PVT	
median RT (msec)	0.73
lapses	0.81