HIGHLIGHTED TOPIC | Physiology and Pathophysiology of Sleep Apnea

Effects of nasal continuous positive airway pressure and oxygen supplementation on norepinephrine kinetics and cardiovascular responses in obstructive sleep apnea

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Submitted 29 April 2005; accepted in final form 6 September 2005

NORADRENERGIC ACTIVATION, as indicated by augmented sympathetic neural activity, elevated circulating norepinephrine (NE) levels, and elevated urinary NE excretion, is a hallmark of obstructive sleep apnea (OSA) (3, 4, 8). Cardiovascular consequences of this activation include high blood pressure and enhanced cardiac contractility, as well as adrenergic receptor desensitization (8, 10, 16–18). The elevated NE levels in OSA could be the result of an enhanced NE release rate and/or decreased NE clearance rate. We previously reported that OSA patients have an enhanced NE release rate under hypoxic conditions and a tendency toward decreased NE clearance (27).

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Treatment of OSA with nasal continuous positive airway pressure (CPAP) reduces circulating levels of norepinephrine, as well as NE excretion (2, 8, 9, 11). The CPAP-induced reduction in NE is associated with a reduction in blood pressure, cardiac contractility, and restoration of desensitized β-adrenergic receptors (18, 19, 25).

The purpose of this study was to determine whether CPAP reduces NE levels by altering daytime release and/or clearance rate and whether such effects would be related to reductions in blood pressure. Considering prior observations that patients with OSA have a tendency toward diminished NE clearance and increased NE levels and that CPAP reduces NE levels, we hypothesized that CPAP treatment would lead to an increase in NE clearance and/or a decrease in NE release rate.

We compared CPAP to a placebo CPAP treatment and to an oxygen supplementation treatment. Nocturnal oxygen supplementation has been suggested by some as an alternative therapy in the noncompliant or the CPAP-noncompliant OSA patient (14, 21). In some OSA patients who cannot tolerate CPAP and are not candidates for a surgical procedure, supplemental oxygen therapy is associated with a reversal of OSA-related nocturnal hypoxemia but does not affect respiratory disturbance index (RDI) (14). The effect of supplemental oxygen on sympathetic activation in OSA has not been rigorously studied.

METHODS

Patients and screening. Patients were recruited from the community through advertisement and referral from sleep-disorder clinics. Screening criteria included clinical suspicion of OSA as per loud snoring with or without excessive daytime sleepiness. Because apneic patients frequently have hypertension (6), and because antihypertensive treatments have noticeable effects on the sympathetic nervous system (24), all hypertensive patients receiving such treatment were tapered off medication for at least 2 wk before participation in the study. Other inclusion criteria included weight being between 1.0 and 2.0 times the ideal body weight (15) and age between 30 and 65 yr. Patients were excluded if they were receiving medications known to affect sleep. Patients with other major medical disorders, including congestive heart failure, symptomatic obstructive pulmonary disease, history of narcolepsy, prior surgery for treatment of OSA, current alcohol or drug abuse, or psychosis were excluded.

Screening included a history and physical examination and complete blood count, chemistry panel, and electrocardiogram. Potentially eligible candidates underwent an unattended overnight home screening sleep study using the Stardust (Respironics, Marietta, GA) home sleep recording system. Subjects with an apnea-hypopnea index (AHI) >15 were then admitted for 3 nights to the University of California, San Diego (UCSD) General Clinical Research Center (GCRC) Gillin

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Laboratory of Sleep and Chronobiology, where full polysomnography confirmed the diagnosis of OSA (2, 11). Subjects with periodic limb movement >15 on baseline polysomnography were excluded from further participation. The same team of nighttime technicians and daytime technicians performed and scored the polysomnograms.

Sleep was recorded using the Grass Heritage (model PSG36-2, West Warwick, RI) sleep recording system using a standard montague. All records were scored according to Rechtschaffen and Kales criteria (22). Percent time less than 90% arterial oxygen saturation (SaO₂ <90%) was determined and averaged over the nights. Apneas were defined as a decrement in airflow >90% from baseline for a period ≥10 s. Hypopneas were defined as a decrement in airflow ≥50% but <90% from baseline for a period ≥10 s. The number of apneas and hypopneas per hour of sleep were determined to obtain the AHI.

Fifty men and women completed screening and were admitted to the study. All patients gave signed consent to the protocol, which was approved by the UCSD Human Subjects Institutional Review Board.

**Treatment design and randomization.** Randomization took place on the second night of admission. Patients were randomized to a 2-wk therapeutic trial of nasal CPAP (n = 17), placebo CPAP (n = 17), placebo CPAP or nocturnal oxygen (n = 17). In the therapeutic CPAP group, conventional manual overnight CPAP titration was performed in increasing steps of 1–2 cmH₂O until unequivocal obstructive apneas or hypopneas were controlled in the second or third rapid eye movement sleep period. All patients randomized to CPAP had an effective titration as defined by an AHI <10. Patients randomized to placebo CPAP or supplemental oxygen underwent a mock titration. On the third night of admission, patients slept with their assigned treatment.

Equipment for the three treatment arms was similar and consisted of a CPAP generator (Aria LX CPAP System, Respiromics, Murrysville, PA), CPAP mask (Profile Light, Respironics), tubing, heated humidifier (Fisher and Pykel HC199, Auckland, New Zealand), and oxygen concentrator (Alliance, Healthdyne Technologies model 505, Marietta, GA). The concentrator could be switched to produce room air. The supplemental gas (room air or oxygen) was introduced into the CPAP generator (Aria LX CPAP System, Respironics, Murrysville, PA), CPAP mask (Profile Light, Respironics), tubing, heated humidifier (Fisher and Pykel HC199, Auckland, New Zealand), and oxygen concentrator delivering oxygen at 3 l/min (inspired oxygen fraction of 32–34% at the mask). The morning after the third night, patients were instructed and shown how to use the CPAP mask and tubing system. With this system, the oxygen concentrator (Alliance, Healthdyne Technologies model 505, Marietta, GA), CPAP mask (Profile Light, Respironics), tubing, heated humidifier (Fisher and Pykel HC199, Auckland, New Zealand), and oxygen concentrator delivering oxygen at 3 l/min (inspired oxygen fraction of 32–34% at the mask).

The rate of NE release was calculated by the formula:

\[
\text{NE release rate (ng/min)} = \frac{\text{3H-NE infused}}{\text{minute}} \times \frac{\text{3H-NE/liter plasma}}{}
\]

Volume of distribution was calculated from the half-life and clearance by the equation: volume = clearance/rate constant of decay (26). The plasma NE half-lives were derived by using a standard curve-fitting program assuming biexponential decay of the [3H]NE.

**Data analysis.** Data were analyzed by two-way (group × time) ANOVA and hierarchical multivariable regression. Separate ANOVAs were run to test whether treatment led to a change in NE clearance and to test whether treatment led to a change in NE release rate. Distribution of all variables was evaluated by Levene’s test of equality of variance; parametric tests were used only in case of normal distribution. Initial relationships among variables were explored by Pearson correlations. Significance level was set at \( P < 0.05 \) for two-sided tests.

### Table 1. Subject characteristics and treatment compliance data

<table>
<thead>
<tr>
<th></th>
<th>CPAP</th>
<th>Oxygen</th>
<th>Placebo CPAP</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>17</td>
<td>17</td>
<td>16</td>
</tr>
<tr>
<td>Men/women</td>
<td>15/2</td>
<td>13/4</td>
<td>13/3</td>
</tr>
<tr>
<td>Normotensive/hypertensive</td>
<td>9/8</td>
<td>12/5</td>
<td>12/4</td>
</tr>
<tr>
<td>Age, yr</td>
<td>47.6 ± 2.58</td>
<td>43.9 ± 2.49</td>
<td>49 ± 2.56</td>
</tr>
<tr>
<td>Body mass index</td>
<td>31.7 ± 1.4</td>
<td>30.7 ± 1.2</td>
<td>32.2 ± 1.7</td>
</tr>
<tr>
<td>Compliance, h/night</td>
<td>6.78 ± 0.381</td>
<td>6.56 ± 0.316</td>
<td>5.98 ± 0.345</td>
</tr>
</tbody>
</table>

Values are means ± SE. CPAP, continuous positive airway pressure. Nightly compliance was averaged over 2 wk; no significant differences among groups.
Because NE clearance and release rate are not independent variables, these ANOVA tests were alpha adjusted. Data analyses were performed using the SPSS 12.0 programming package (Chicago, IL). All variables are presented as means ± SE.

RESULTS

Pretreatment. Table 1 presents subject characteristics and treatment compliance data. Body mass index (BMI) was computed as the ratio of body weight in kilograms divided by the square of height in meters (kg/m²). Before treatment, subjects randomized to the three treatment groups were similar in age, BMI, gender distribution, and hypertension distribution. Compliance with treatment was based on the average number of hours of use per night during the 2-wk treatment period. There was no significant difference among the three groups in treatment compliance (F = 1.693, P = 0.195).

Before treatment, among all patients, blood pressure was related to both day and night NE excretion rates (P < 0.05) (Table 2). AHI was related to NE release rate (P < 0.01) and day and night NE excretion rates (P < 0.05). SaO₂ < 90% correlated with NE release rate (P < 0.01), supine plasma NE levels (P < 0.01), and day and night NE excretion rates (P < 0.01).

Posttreatment. Table 3 presents the effects of treatment on AHI, SaO₂ < 90%, blood pressure, and heart rate. There were no significant pretreatment differences among the groups for any of these variables. AHI was significantly reduced in the CPAP condition (F = 28.9, P < 0.001) but not in the oxygen or placebo CPAP conditions. SaO₂ < 90% was significantly reduced in the CPAP condition and the oxygen conditions (F values >6.3, P < 0.05) but not in the placebo CPAP condition. Systolic (F = 7.8, P = 0.013) and diastolic (F = 6.1, P = 0.026) blood pressure and heart rate (F = 7.7, P ≤ 0.014) were decreased in response to CPAP but not the oxygen or placebo CPAP.

Two weeks of CPAP led to a significant increase in NE clearance (F = 9.53, P ≤ 0.01) (Table 4 and Fig. 1). The enhanced clearance of NE was due to an expanded volume of distribution (F = 7.7, P ≤ 0.017), not to a shortened half-life (P = 0.06) (Table 4). NE clearance, volume of distribution, and half-life were unchanged after either oxygen or placebo CPAP. Supine plasma NE levels were reduced after CPAP (F = 7.9, P ≤ 0.018) but unchanged after either oxygen or placebo CPAP. Daytime NE excretion was reduced after 2 wk of CPAP treatment (F = 20.8, P < 0.001) and oxygen treatment (F = 9.9, P < 0.01) but unchanged after placebo CPAP. Nighttime NE excretion was reduced after CPAP treatment only (F = 5.6, P < 0.05). NE release rate was unchanged with any treatment (Fig. 1).

In an effort to better understand the role of NE in blood pressure and heart rate responses to treatment, we conducted a series of multiple regression analyses examining possible predictors of posttreatment levels of systolic and diastolic blood pressure and heart rate. Dependent variables were entered into the regression in blocks as follows: block 1: age, BMI, gender, and diagnosis of hypertension; block 2: pretreatment AHI, SaO₂ < 90%, and the respective pretreatment blood pressure or heart rate; block 3: pretreatment NE clearance, NE release rate, supine plasma NE, and daytime and nighttime NE excretion; block 4: posttreatment AHI, SaO₂ < 90%; block 5: posttreatment NE clearance, NE release rate, supine plasma NE, and daytime and nighttime NE excretion. Posttreatment systolic blood pressure was predicted by pretreatment systolic blood pressure (β = 0.450, P = 0.005), posttreatment NE clearance (β = –0.836, P = 0.001), and posttreatment NE release rate (β =
Posttreatment heart rate was predicted by pretreatment heart rate (\( r^2 = 0.717, P = 0.005 \)), yielding the full regression model of \( r^2 = 0.687, F = 16.1, P < 0.001 \), with all other predictor variables dropping out as not significant. Posttreatment diastolic blood pressure was predicted by pretreatment diastolic blood pressure (full regression model: \( r^2 = 0.284, F = 10.5, P < 0.01 \)). Posttreatment heart rate was predicted by pretreatment heart rate (\( \beta = 0.706, P = 0.000 \)), pretreatment NE clearance (\( \beta = 0.253, P = 0.008 \)), and pretreatment \( S_{\text{O}_2} < 90\% \) (\( \beta = 0.293, P = 0.004 \)), yielding the full regression model of \( r^2 = 0.788, F = 35.7, P < 0.001 \).

**DISCUSSION**

The impetus for this study arose from observations that CPAP treatment successfully reduces plasma and urine NE in OSA. We wondered whether a mechanism of this effect was CPAP-induced alterations in NE clearance or release. We observed that in patients treated with CPAP, in addition to reductions in circulating NE and NE excretion, daytime NE clearance was increased. We did not observe a change in daytime NE release rate, although prior studies have shown that OSA patients have increased sympathetic nerve activity in the daytime and that CPAP leads to a reduction in sympathetic neural activity during sleep. Our measures help specify where CPAP induces changes in daytime sympathetic nerve activity. There was a clear reduction in urine NE with CPAP. NE in the urine comes from the blood and from NE released by sympathetic nerves in the kidney. Our findings suggest that CPAP causes a reduction in renal sympathetic neuronal activity, which might diminish sodium retention and renin release. CPAP is reported to diminish daytime muscle sympathetic nerve activity to the leg (23), so there appears to be localized changes in nerve activity to the kidney and some muscle vasculature, although we did not find an overall decrease in NE release rate. CPAP also led to an increase in the volume of distribution for NE. This could be due to more active diffusion of NE out of the bloodstream, enhanced reversible transport of NE by uptake-1 and uptake-2 mechanisms, or enhanced binding of NE by plasma proteins and cellular structures. Our study does not indicate which of these potential mechanisms predominates, but we have previously found a suggestion of enhanced binding to receptors after CPAP (25).

We compared CPAP against another treatment for OSA, namely oxygen supplementation. Although both treatments successfully improved oxygen saturation, only CPAP reduced AHI. Only CPAP significantly lowered daytime blood pressure (\( r^2 = 0.293, F = 16.1, P < 0.001 \)), yielding the full regression model of \( r^2 = 0.706, F = 10.5, P < 0.01 \)). Posttreatment diastolic blood pressure was predicted by pretreatment diastolic blood pressure (full regression model: \( r^2 = 0.253, F = 0.008 \)), with all other predictor variables dropping out as not significant. Posttreatment diastolic blood pressure was predicted by pretreatment diastolic blood pressure (full regression model: \( r^2 = 0.293, P = 0.004 \)), yielding the full regression model of \( r^2 = 0.788, F = 35.7, P < 0.001 \).

**Table 4. Treatment effects on norepinephrine kinetics, plasma levels, and urinary excretion**

<table>
<thead>
<tr>
<th></th>
<th>Pre</th>
<th>Post</th>
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<tbody>
<tr>
<td>NE clearance, liters/min</td>
<td>3.17±0.28</td>
<td>3.89±0.24</td>
</tr>
<tr>
<td>NE release to blood, ng/min</td>
<td>907±135</td>
<td>924±81</td>
</tr>
<tr>
<td>Plasma NE, pg/ml</td>
<td>278±29</td>
<td>229±19</td>
</tr>
<tr>
<td>Volume of distribution, liter</td>
<td>7.3±0.86</td>
<td>11.6±1.5</td>
</tr>
<tr>
<td>Short half-life, min</td>
<td>1.73±0.21</td>
<td>2.17±0.25</td>
</tr>
<tr>
<td>Long half-life, min</td>
<td>80±14</td>
<td>101±13</td>
</tr>
<tr>
<td>Supine plasma NE, pg/ml</td>
<td>282±30</td>
<td>211±21</td>
</tr>
<tr>
<td>NE excretion, day, μg/h</td>
<td>2.88±0.32</td>
<td>1.80±0.25</td>
</tr>
<tr>
<td>NE excretion, night, μg/h</td>
<td>1.35±0.22</td>
<td>1.02±0.13</td>
</tr>
</tbody>
</table>

Values are means ± SE. *Increase with CPAP, \( P = 0.014 \); †increase with CPAP, \( P = 0.017 \); ‡decrease with CPAP, \( P = 0.018 \); §reduction with CPAP and \( O_2, P = 0.000 \); ‖reduction with CPAP, \( P = 0.031 \).
sure; its effect was about twice as large as the nonsignificant decrease seen with oxygen. Both CPAP and oxygen lowered daytime urine NE; the effect of CPAP was about twice as large as oxygen. CPAP also lowered nighttime urine NE, but oxygen failed to do so. This is not surprising, because oxygen failed to alter the number of nighttime apneas, and sympathetic nerve activity increases markedly after apneic episodes (25).

We previously reported that CPAP reduces daytime ambulatory mean arterial blood pressure to the same extent as placebo CPAP but that CPAP leads to a much greater decrease in nighttime ambulatory mean arterial blood pressure compared with placebo CPAP (5). This previous study was of 1-wk duration as opposed to 2-wk duration herein. Studies by Becker et al. (1) and Pepperrell et al. (20) were of longer duration and found blood pressure to decrease with CPAP. It is likely that the beneficial effects of CPAP on blood pressure are more evident with longer treatment.

OSA is associated with hypertension. The proposed mechanism for the association is sympathetic activation (3, 4, 8). Intermittent hypoxia causes hypertension in rats, but chronic hypoxia does not usually cause hypertension in humans (7). On the other hand, recovery from individual episodes of apnea increases sympathetic nerve activity and blood pressure. Our study suggests that apneic episodes are more important than hypoxia in increasing plasma and urine NE and blood pressure. We also examined possible predictors of posttreatment blood pressure and heart rate. We found that posttreatment systolic blood pressure was best predicted by a combination of pretreatment systolic blood pressure and posttreatment NE clearance and release rates (e.g., a lower posttreatment blood pressure was associated with a lower pretreatment blood pressure, a higher posttreatment NE clearance, and a lower posttreatment NE release rate). We are not aware of evidence for a direct link between a change in NE clearance and cardiovascular consequences. Although increased clearance decreases plasma NE levels, the decrease is too small to have direct measurable consequences. An indirect link between NE clearance and cardiovascular consequences is that NE clearance is useful in calculating NE release rate. NE release rate did not change with CPAP therapy despite the decrease in plasma NE levels. In contrast, urine NE levels did decrease with CPAP. An unchanged NE release rate and a diminished urine NE level after CPAP imply that CPAP decreases renal sympathetic nerve activity. Decreased renal sympathetic nerve activity might decrease sodium retention and renin, with lower blood pressure. Our randomization of apneic patients to treatment was successful in that there were no significant pretreatment differences among the three groups in terms of apnea severity, age, weight, or blood pressure. Before treatment, among all patients, apnea severity, as indicated by AHI and SA_{30}, <90%, was related to higher plasma NE levels and higher NE excretion, confirming prior observations of increasing noradrenergic activation with increasing apnea severity (3, 4, 8).

In summary, 2 wk of CPAP treatment for OSA resulted in increased NE clearance rate. Posttreatment systolic blood pressure was related to pretreatment systolic blood pressure and posttreatment NE clearance and NE release rate. CPAP lowered urinary NE and blood pressure more effectively than oxygen supplementation, suggesting that apneic episodes might be more important than hypoxia in determining sympathetic activity and blood pressure.

GRANTS

This work was supported by grants HL-073355, HL-40102, and HL-57265 from the National Heart, Lung, and Blood Institute and the UCSF General Clinical Research Center (MO1RR-00827).

REFERENCES


J Appl Physiol • VOL 100 • JANUARY 2006 • www.jap.org


