The impact of obstructive sleep apnea syndrome on renin and aldosterone

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Abstract. – OBJECTIVE: Obstructive Sleep Apnoea Syndrome (OSAS) is a respiratory disorder characterized by recurrent airflow obstruction caused by total or partial collapse of the upper airway. OSAS is an established independent factor of cardiovascular risk together with other risk factors such as smoking and increased lipids. The aim of our study was to measure serum levels of aldosterone and renin in OSAS patients that did not suffer from arterial hypertension and compare them to matched healthy subjects in order to reveal the impact of chronic intermittent hypoxia on the renin-angiotensin-aldosterone system.

PATIENTS AND METHODS: The patients that enrolled in this study were 19 OSAS patients who had undergone overnight polysomnography and had an Apnoea Hypopnoea Index (AHI) greater than 10 events/hour. They were compared to 20 healthy non-OSAS closely matched controls. Serum aldosterone and direct renin concentration were measured by radioimmunoassay.

RESULTS: Aldosterone concentration follows a diurnal variation; therefore, all blood samples were obtained at the same time (6 AM). There were no significant differences in serum aldosterone levels between the two studied groups of OSAS patients and the healthy subjects group (140.6 pg/ml ± 25.2 vs. 133.2 pg/ml ± 18.5 with p = 0.223). Similar were the results for the renin levels (25.0 ± 6.9 vs. 24.9 ± 4.4 with p = 0.360).

CONCLUSIONS: Our study suggests that patients with OSAS, but without existing hypertension have aldosterone and renin levels similar to healthy subjects. According to our findings a direct connection between OSAS and the development of arterial hypertension may not be established via sympathetic system activation.

Key Words: OSAS, RAAS, Renin, Aldosterone, sympathetic system activation.

Abbreviations


Introduction

Obstructive Sleep Apnoea Syndrome (OSAS) is a highly prevalent respiratory disorder in adults in developed countries. It is characterized by recurrent airflow obstruction caused by total or partial collapse of the upper airway. In OSAS, the upper airway obstruction is usually caused by abnormal anatomy or abnormal control of the muscles that maintain the patency of the upper airway. These disruptions of breathing result in intermittent hypoxia and hypercapnia and surges of sympathetic activation¹.

Obstructive Sleep Apnoea Syndrome is an established independent factor of cardiovascular risk together with other risk factors such as smoking and increased lipids². Moreover, it is supposed to play an important role in arterial hypertension, although the relating mechanisms still remain unclear. It is known the sympathetic activity of patients with OSAS is increased and can lead to arterial hypertension, oxidative stress, chronic inflammation and endothelial dysfunction³. Moreover, chronic intermittent hypoxia is also present in OSAS patients, which may also cause endothelial dysfunction and production of pro-inflammatory cytokines⁴,⁵.
The Autonomic Nervous System (ANS) acts as a motor system undertaking a large number of specialised tasks, stimulatory and inhibitory, in a wide range of target organs. The ANS is divided into the sympathetic system, that forms the major part of the ANS and the parasympathetic system that originates from the brain stem and supplies the seventh, ninth and tenth cranial nerves.

Sleep onset is associated with marked changes in the function of the respiratory system and the heart. Depending on the stage of sleep, different patterns of haemodynamic and autonomic responses are observed. During NREM sleep there is a decrease in heart rate, and systolic blood pressure falls as well. These changes, are supposed to occur because of changes in autonomic activity. However, data on autonomic function during sleep in humans are still limited owing to methodological problems. Therefore, the investigation of the autonomic system hyperreactivity can only be done indirectly.

The renin-angiotensin-aldosterone system (RAAS) regulates extracellular fluid volume and blood pressure. The RAAS protects against loss of salt and water, but may contribute to excess sodium retention that could lead to arterial hypertension. This system may be potentiated by increased sympathetic system activity, increased insulin levels, and increased leptin levels, factors that are frequently present in OSAS.

Sleep apnoea patients have been shown to have higher angiotensin II concentrations compared to healthy subjects. Patients with arterial hypertension and high risk for OSAS have lower plasma renin activity and greater aldosterone excretion compared to subjects at low risk for OSAS. However, in this study polysomnography was not performed in all the subjects, but the presence of OSAS was only based on questionnaires.

Most of the previous studies on the subject face methodological problems or lack sufficient statistical power. In a past study aldosterone levels were found to be lower in the group patients than in the group of controls. In contrary, in another study, blood pressure and levels of angiotensin II and aldosterone were compared in the groups of OSAS patients and controls, however the OSAS patients had been already diagnosed with arterial hypertension and they were more obese. Another study reported that OSAS and hyperaldosteronism were both highly prevalent in patients with resistant hypertension.

The aim of our study was to measure serum levels of aldosterone and renin in OSAS patients that did not suffer from arterial hypertension and compare them to matched healthy subjects in order to reveal the impact of chronic intermittent hypoxia and ANS activation on the renin-angiotensin-aldosterone system.

Patients and Methods

Patients
The study was approved by our Institutional Ethics Committee and informed written consent was obtained from all the participants. The patients that enrolled in this study were 19 OSAS patients that had undergone overnight polysomnography and had an AHI greater than 10 events/hour. They were compared to 20 healthy non-OSAS controls that were also studied with polysomnography to rule out sleep apnoea. Sleep apnoea patients were closely matched to the controls regarding their age, gender and Body Mass Index (BMI). The selection of controls was performed in individuals that had already performed full polysomnography and a total number of 240 patients were screened to find matched controls. All subjects in both groups were otherwise healthy, and did not take any medications. A full medical history and sleep history was obtained from each patient. Thus, subjects with arterial hypertension, cardiovascular or cerebrovascular disease and diabetes were not included in the study. Each patient had also completed Epworth Sleepiness Scale questionnaire.

Polysomnography
An overnight full polysomnography was performed in all participants to determine the presence and severity of OSAS (Profusion PSG, Compumedics, Australia). An obstructive apnoea/hypopnoea was defined as an event that lasts for > 10sec and is characterized by a transient reduction in (hypopnoea), or complete cessation of (apnoea) breathing. According to the American Academy of Sleep Medicine (AASM) criteria from 2007, a hypopnoea can be defined as a decrease from baseline in the amplitude of a valid measure of breathing during sleep that either reaches >50% with an oxygen desaturation of 3% or an arousal, or alternatively a 30% reduction with 4% oxygen desaturation. The degree of severity was defined on the basis of the
number of apnoeas and hypopnoeas occurring per hour of sleep (Apnoea-Hypopnoea Index – AHI) and the severity of daytime symptoms.

**Sample Collection and Measurements**

The blood samples for the measurements were obtained at 6 AM, when the patient was awakened in the sleep laboratory, in a supine position before any physical activity. Routine laboratory measurements were performed in the biochemistry laboratory. The remaining blood samples were centrifuged and frozen at –80°C for further analysis. Each participant was asked to collect 24h urine starting from the previous day of the polysomnography. Therefore, when the overnight study was finished we also collected the 24h urine collection. Urine biochemistry tests were performed in the biochemistry laboratory.

Serum aldosterone concentration (pg/ml) was measured by radioactive immunoassay, ACTIVE® aldosterone RIA (Immunootech, Czech Republic/BeckmanCoulter). Sensitivity of the method was 7.64 pg/ml and the intra- and inter-assay coefficients of variation average 4.5% and 9.8% respectively. Urine aldosterone was measured using the same kit. Sensitivity of the method was 7.64 pg/ml and the intra- and inter-assay coefficients of variation average 7.4% and 6.4% respectively in the urine samples. Plasma renin was measured by ACTIVE® renin IRMA (Immunootech, Praha, Czech Republic/BeckmanCoulter). Sensitivity of the method was 0.7 pg/ml and the intra- and inter-assay coefficients of variation average 1.63% and 2.6% respectively.

**Statistical Analysis**

Results are reported as mean ± SEM. Continuous variables were compared between groups using one-way analysis of variance (ANOVA). Post-hoc analyses were performed where indicated. Statistical significance was defined as $p < 0.05$. IBM SPSS Statistics v. 20 (SPSS Inc., Chicago, IL, USA) was used for all the statistics.

**Results**

Demographic data of the studied groups of the OSAS patients and controls are illustrated in Table I. The OSAS group consists of 19 patients with moderate to severe OSAS, whereas the control group consists of 20 healthy subjects. There was a close matching of the studied subjects regarding important factors that may affect the AHI and aldosterone and renin measurements, such as the age, Body Mass Index, systolic and diastolic pressure and heart rate. Patients with OSAS had an AHI of 30.5 events/hour ± 5.0, with significant nocturnal desaturation of 6% ± 2, whereas the lowest $SaO_2$ of 77.6% ± 2.1 on average. OSAS patients that participated in the study had an Epworth Sleepiness Scale (ESS) score ≥ 10, thus showing a remarkable daytime sleepiness, which is a main symptom in OSAS. The main characteristics of the sleep studies in both groups are shown in Table II.

Moreover, typical biochemistry measurements were performed for all the subjects of the study. These results are illustrated in Table III. There is no statistically significant difference between the two groups, which is important because of the impact of renin and aldosterone on the regulation of $K^+$ and $Na^+$ as well as water retention (Table III).

Aldosterone concentration follows a diurnal variation; therefore, all blood samples were ob-

| Table I. Characteristics of OSAS patients and healthy controls of the study. |
|-------------------------------|-------------------------------|-------------------------------|-------------------------------|
| Demographics                  | Moderate to severe OSAS (n=19) | Healthy controls (n=20)       | Significance ($p < 0.05$)    |
| Males/Females                 | 14/5                          | 14/6                          | NS                            |
| Age (years)                   | 58 ± 5                        | 50 ± 3                        | NS                            |
| Height (cm)                   | 172 ± 2                       | 174 ± 2                       | NS                            |
| Weight (kg)                   | 92 ± 4                        | 84 ± 4                        | NS                            |
| Body Mass Index (BMI) (kg/m²) | 30.8 ± 1.2                    | 27.6 ± 1.1                    | NS                            |
| Systolic BP (mmHg)            | 128 ± 2                       | 117 ± 2                       | NS                            |
| Diastolic BP (mmHg)           | 80 ± 2                        | 75 ± 1                        | NS                            |
| Heart rate awake (bpm)        | 81 ± 3                        | 76 ± 2                        | NS                            |
| $SpO_2$ awake (%)             | 95.6 ± 0.5                    | 96.8 ± 0.3                    | NS                            |

Values are mean ± SEM (Standard Error of the Mean).
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Table II. Sleep parameters from the overnight full polysomnography that was performed both in patients and controls.

<table>
<thead>
<tr>
<th>Sleep parameters</th>
<th>Moderate to severe OSAS (n = 19)</th>
<th>Healthy controls (n = 20)</th>
<th>Significance (p values)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AHI (events/hour)</td>
<td>30.5 ± 5.0</td>
<td>3.7 ± 0.6</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Lowest SaO₂ (%)</td>
<td>77.6 ± 2.1</td>
<td>88.6 ± 1.1</td>
<td>= 0.034</td>
</tr>
<tr>
<td>Time below 90% (min)</td>
<td>64.6 ± 20.2</td>
<td>1.4 ± 0.7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Average SaO₂ desaturation (number)</td>
<td>6 ± 2</td>
<td>3.4 ± 0.4</td>
<td>= 0.006</td>
</tr>
<tr>
<td>ESS score</td>
<td>14 ± 2</td>
<td>5 ± 3</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Table III. Routine laboratory examinations in OSAS patients and healthy controls.

<table>
<thead>
<tr>
<th>Laboratory examination</th>
<th>Moderate to severe OSAS (n = 19)</th>
<th>Healthy controls (n = 20)</th>
<th>Significance (p values)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Haematocrit (%)</td>
<td>42.8 ± 1.0</td>
<td>43.8 ± 0.9</td>
<td>NS</td>
</tr>
<tr>
<td>K⁺ (mmol/l)</td>
<td>4.5 ± 0.07</td>
<td>4.8 ± 0.09</td>
<td>NS</td>
</tr>
<tr>
<td>Na⁺ (mmol/l)</td>
<td>141.0 ± 0.4</td>
<td>139.3 ± 0.4</td>
<td>NS</td>
</tr>
<tr>
<td>Urea (mg/dl)</td>
<td>39 ± 5.0</td>
<td>35 ± 1.4</td>
<td>NS</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>0.88 ± 0.05</td>
<td>0.93 ± 0.03</td>
<td>NS</td>
</tr>
<tr>
<td>SGOT (U/l)</td>
<td>24 ± 3</td>
<td>21 ± 1</td>
<td>NS</td>
</tr>
<tr>
<td>SGPT (U/l)</td>
<td>30 ± 3</td>
<td>24 ± 2</td>
<td>NS</td>
</tr>
</tbody>
</table>

Obstructive sleep apnoea syndrome is considered as an independent risk factor for cardiovascular disease and is strongly associated with arterial hypertension, ischemic heart disease, heart failure and cerebrovascular disease. The Wisconsin Sleep Cohort Study has found a dose-response association between sleep-disordered breathing at baseline and the presence of hypertension four years later that was independent of known confounding factors. More specifically the odds ratio for the presence of hypertension at follow-up 4 years later was 2.89 in patients with sleep apnoea\textsuperscript{15}. OSAS and cardiovascular disease share common risk factors, such as obesity, male gender, smoking and advanced age.

![Figure 1. Aldosterone levels in the two studied groups of 19 patients with moderate to severe OSAS and the 20 matched controls. Measurements were done at 6 AM, when the patient was awakened. Data are shown as mean ± SEM.](image_url)
dosterone secretion as a response to the hypoxic stimulus and this has been supposed to be a possible mechanism of arterial hypertension. Past studies that evaluated renin and aldosterone levels as well as blood pressure in patients with OSAS that received CPAP treatment have been controversial. In a prospective uncontrolled evaluation of OSAS patients, CPAP treatment lead to a decrease in aldosterone concentrations; however, blood pressure was not affected\(^{19}\). In another study, CPAP treatment was shown to cause a reduction both in blood pressure and plasma renin and angiotensin II concentrations, which is in contrary to the previous findings\(^{10}\). Thus, a direct pathogenetic mechanism linking renin and aldosterone production in OSAS patients without any co-morbidities is rather weak.

In our study we investigated patients with OSAS that did not have arterial hypertension or any other co-morbidities, who were newly diagnosed with OSAS. Therefore, they had not received CPAP treatment yet. Moreover, the patient group had moderate to severe OSAS with marked SaO\(_2\) desaturation, thus implying the effect of intermittent hypoxia to autonomic nervous system activation and renin and aldosterone levels. The group of OSAS patients was compared to a closely matched group of healthy controls for the age, BMI and arterial blood pressure. Both groups of patients and controls did not take any medication or antihypertensive drugs and did not smoke.

Our results show that moderate to severe OSAS does not affect renin and aldosterone levels, as they were found to be similar both in the group of OSAS patients and the group of healthy control subjects. Moreover, another implication of our findings is that chronic intermittent hypoxia in OSAS does not seem to lead to alterations of aldosterone production. Therefore, OSAS does not seem to affect the RAAS system by itself, unless other confounding factors are also present. However, this could be a field of future research.

Our study has expanded current knowledge as the participants were OSAS patients and healthy controls without arterial hypertension or other co-existing disease that could alter the levels of renin and aldosterone. Moreover, the patients’ group had moderate to severe OSAS with remarkable nocturnal desaturation, thus the impact that chronic intermittent hypoxia could have on renin and aldosterone secretion could be evident in this group. Furthermore, other important factors, such as smoking habit and obesity, were...
carefully investigated and were not found to be significantly different in both groups. It is important that in our study we have also studied renal function and hormone profile of the patients and controls so as to reach more powerful results. Finally, overnight full polysomnography was performed both in OSAS patients and controls to define or exclude the existence of OSAS.

A limitation of our study is the modest sample size; however both males and females were studied unlike in other studies. Renal function was checked and all OSAS patients and controls did not have renal dysfunction.

It is known that aldosterone may play an important role in patients with arterial hypertension. Despite the fact that OSAS has been correlated with higher levels of aldosterone in OSAS patients with resistant hypertension, the intermittent hypoxia that occurs in OSAS does not seem to affect renin and aldosterone concentrations in normotensive OSAS patients. Although OSAS has certain effects on the autonomic nervous system, the metabolism and the heart function, it was not proven to lead to an increase in renin and aldosterone levels in otherwise healthy OSAS patients. These findings are important for the understanding of the renin and aldosterone regulation and the connection between OSAS and arterial hypertension. According to our findings a direct connection between OSAS and the development of arterial hypertension may not be established via sympathetic system activation.

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Conflict of Interest

The Authors declare that there are no conflicts of interest.

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14) American Academy of Sleep Medicine. The AASM Manual for the Scoring of Sleep and Associated


