The impact of obstructive sleep apnea on endothelial function during weight loss in an obese pediatric population

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Abstract

Background: Childhood obesity is an increasing problem with substantial comorbidities such as obstructive sleep apnea (OSA) and increased cardiovascular morbidity. Endothelial dysfunction is an underlying mechanism related to both obesity and OSA.

Research Question: To investigate the effect of weight loss on endothelial function and OSA in obese children and to determine whether a change in endothelial function can be linked to an improvement in OSA.

Methods: Obese children between 8-18 years of age were recruited while entering a 12-month inpatient weight loss program. Patients were followed at 3 study visits: baseline, after 10 months of weight loss, and 6 months after ending the program (18 months). Anthropometry and endothelial function were determined (EndoPAT) at all study visits. At baseline, sleep screening with a portable device (ApneaLink) was performed. This was repeated after 10 months if OSA was diagnosed at baseline.

Results: At baseline, 130 children were included, of which 87 had OSA (67%). Seventy-two patients attended the follow-up visit at 10 months, and 28 patients attended the follow-up visit at 18 months. The BMI z-score decreased after 10 months (from 2.7 (1.4-3.4) to 1.7 (0.5-2.7); $p<0.001$) and remained stable at 18 months. Endothelial function improved significantly after weight loss, evidenced by a shorter time to peak response (TPR) and higher reactive hyperemia index ($p=0.02$ and $p<0.001$), and remained improved after 18 months ($p<0.001$ and $p=0.007$). After 10 months of weight loss, 10 patients had residual OSA. These patients had a higher TPR at 10 months (225 (75-285)s) than those without OSA (135 (45-225)s) and patients with a normalized sleep study (105 (45-285)s; $p=0.02$). Linear mixed models showed that more severe OSA was associated with a worse TPR at baseline and less improvement after weight loss.

Conclusion: Weight loss improves endothelial function in an obese pediatric population. However, even after weight loss, endothelial function improved less in the presence of OSA.

Key words

Weight loss, obesity, obstructive sleep apnea, sleep-disordered breathing, endothelium, adolescents, pediatrics

Abbreviation list

BCM: Body composition monitor

BMI: Body mass index

BIC: Schwarz's Bayesian Criterion

En% : Total energy intake

MaxD: Maximal dilatation after occlusion measured by Endopat

oAHI: Obstructive apnea-hypopnea index

ODI: Oxygen desaturation index

OSA: Obstructive sleep apnea

PWA: Pulse wave amplitude

RHI : Reactive hyperemia index

TPR : Time to peak response

WHR: Waist-to-hip ratio

1.1 Introduction

According to the World Health Organization[1], 340 million children and adolescents worldwide were overweight or obese in 2016. In Belgium, 19% of children and adolescents between 2 and 17 years of age are overweight, and approximately 5.8% of them are obese[2]. This makes childhood obesity one of the most serious public health challenges of the 21st century, since obesity in childhood increases the risk of obesity at an adult age[3] and is associated with several cardiovascular risk factors[4]. Obesity is also a well-known risk factor for developing obstructive sleep apnea (OSA). OSA is characterized by intermittent cycles of upper-airway collapse associated with hypoxia and arousals during sleep. In the general pediatric population the prevalence of OSA is estimated to be approximately 2-3%[5]; however, OSA can be present in 13 to 60% of obese children[6]. Similar to obesity, OSA is independently associated with important morbidity, mostly involving the cardiovascular and metabolic systems[7,8]. Several studies have shown increased inflammation and activation of the autonomic nervous system in pediatric OSA[9-11]. These processes could lead to impaired endothelial function, which is an early marker for cardiovascular risk. Indeed, studies have shown that endothelial dysfunction is more frequent in normal weight children with OSA than in healthy peers[12]. The interaction between obesity and OSA could augment endothelial dysfunction supporting the concept that both conditions can amplify long-term cardiovascular risk[10,13].

Our group previously showed that a residential weight loss program improves endothelial function in obese children[14] as well as OSA[15,16]. However, how OSA influences endothelial function during weight loss has not yet been investigated. Although endothelial dysfunction has long-term negative consequences, it is still reversible in children, making it an important factor to investigate. Therefore, the goal of this study was to investigate the effect of weight loss on endothelial function and OSA in an obese pediatric population and to determine whether a change in endothelial function can be linked to an improvement in OSA.

1.2 Material and methods

1.2.1 Study population and study design

Obese children and adolescents aged 8-18 years were consecutively recruited while entering an inpatient weight loss treatment program at the rehabilitation center "Het Zeepreventorium" in De Haan (Belgium). The weight reduction program was extensively discussed elsewhere[17]. Briefly, it consists of a multicomponent treatment with moderate dietary restrictions (10% of total energy intake (En%) consists of proteins, 50-55 En% of carbohydrates (with <10 En% of mono- and disaccharides) and 30-35 En% of fat), increased physical activity (minimum of 10 hours/week), psychological support and medical supervision. Exclusion criteria were neuromuscular or known endocrine diseases, genetic or craniofacial syndromes, acute illness, uncontrolled chronic disease or the use of anti-inflammatory drugs at the moment of the study.

All patients were asked to complete 3 study visits: a baseline visit at the moment of admission in the weight loss program, a second visit after 10 months of weight loss treatment, and a third visit 6 months after ending the weight loss program (18 months of follow-up).

The Ethics Committee of Ghent University Hospital and Antwerp University Hospital approved this study (n° B670201731779), and informed consent was obtained from the patients and their parents or legal guardians before the start of the study.

1.2.2 Anthropometry

Height, weight, waist circumference and waist-to-hip ratio (WHR) were measured using standardized techniques by skilled personnel. Body mass index (BMI) was calculated as weight in kilograms over height in m^2 and was further analyzed as z-scores using the Flemish growth study as a reference population[18]. Overweight and obesity were defined according to the International Obesity Task Force criteria[19].

1.2.3 Blood pressure

Blood pressure was measured at each time point by an automated validated oscillometric device at the nondominant upper arm with an adjusted cuff. Blood pressure was measured three times, and the mean value was used. Corresponding sex-, age- and height-specific percentiles for systolic and diastolic blood pressure were calculated[20].

1.2.4 Body composition measurement

A body composition monitor (BCM) measurement was performed at each time point in the morning after an overnight fast (Fresenius Medical Care, St. Wendel, Germany) with the child lying supine.

Electrodes were attached following the wrist-ankle approach, in a tetrapolar arrangement with two electrodes placed on the hands and two on the feet. To guarantee good contact of the electrodes with the skin, degreasing with diethylether was performed before placement of the electrodes. Age, sex, height, weight and blood pressure were registered by the device before starting the measurement. If the quality calculated by BCM was <75%, the measurement was repeated, and only good quality measurements were used. All guidelines for the use of the BCM were as follows: nonelectrical bed, no cell phones and no electrical devices within 1 meter of the device. The parameter of interest was: fat mass expressed as a percentage.

1.2.5 Peripheral arterial tonometry

Peripheral microvascular endothelial dysfunction was noninvasively measured at the distal phalanx of the index finger with Endo-PAT 2000 (Itamar Medical Ltd, Caesarea, Israel). Measurements were performed at every study visit in the morning after an overnight fast in a temperature controlled room (21-24°C).

Briefly, finger probes were placed at the fingertips of both hands to measure arterial pulse wave amplitudes. After a 5-minute baseline assessment, a manometer cuff was inflated to supra-systolic pressures (≥ 200 mmHg), and the brachial artery of the nondominant arm was occluded. After 5

minutes, the cuff was released, and reactive hyperemia was observed for 5 minutes. Measurements were performed according to manufacturers' guidelines and recommendations in children[21].

Parameters of interest were the reactive hyperemia index (RHI), the mean baseline pulse wave amplitude (PWA), the maximal dilatation after occlusion (MaxD) and the time to peak response (TPR). The augmentation index was assessed as an index for arterial stiffness[22].

1.2.6 Sleep assessment

Sleep assessment was performed using a portable device (ApneaLink Air™, ResMed, Switzerland)[23,24] at baseline and after 10 months only when sleep-disordered breathing was observed during the first investigation. Respiratory airflow was measured by a nasal pressure cannula (detecting -10 hPa to +10 hPa), and the oxygen saturation and pulse rate were obtained by using a pulse oximeter and a pulse sensor (sampling rate of 1 Hz). Respiratory effort was measured by a thorax belt. Tracings were all manually reviewed, and measurements associated with poor pulse tracing or aberrant respiratory signals were excluded from the analysis. The sleep study was repeated if less than 4 hours of good quality signal were obtained.

The obstructive apnea hypopnea index (oAHI) was defined as the number of obstructive apneas and hypopneas per hour of sleep divided by the total hours of sleep. OSA was diagnosed by an oAHI ≥2[25]. The oxygen desaturation index (ODI) was defined as the number of episodes of oxygen desaturation per hour of sleep. An episode of oxygen desaturation consists of a decrease of at least 3% in blood oxygen saturation[26].

1.2.7 Statistical analysis

All statistical analyses were performed using SPSS 26.0 (SPSS, Chigaco, Illinois, USA). Normality was tested by the Kolmogorov-Smirnov test and bar graph. Normally distributed data are summarized as the mean ± standard deviation, and skewed data are summarized as the median and range (minimum – maximum). To compare two groups, independent-samples Student's t tests and Mann-Whitney U

tests were used. To compare three groups, one-way ANOVA or independent sample Kruskal-Wallis tests were used. To compare data at different points in time, a repeated-measures ANOVA with LSD post hoc tests was performed or a Friedman test or a Wilcoxon signed rank test as a nonparametric alternative. Correlations were computed using Pearson or Spearman correlation analysis. To investigate a possible association between endothelial dysfunction, obesity and OSA over time, a linear mixed model analysis was used. Schwarz's Bayesian Criterion (BIC) was used to find the model with the best fit. For all analyses, $p \leq 0.05$ was considered statistically significant.

1.3 Results

1.3.1 Baseline characteristics

A total of 130 patients were included with a mean weight of 103.2 ± 24.7 kg and a mean BMI of 36.7 ± 0.5 kg/m², which corresponds to a BMI z-score of 2.7 ± 0.03. The mean age at baseline was 14 ± 0.19 years, and 41.5% of patients were boys. Of these patients, 118 were Caucasian (84 North European, 23 North African and 2 East European), 1 was Asian and 11 were of African descent.

Based on sleep screening at enrollment, 43 patients (33%) did not have OSA, and 87 patients (67%) were diagnosed with OSA. All sleep tracings were of sufficient quality, and no assessments were excluded. The characteristics of patients with and without OSA are compared in Table 1. Waist circumference and WHR significantly differed between the groups ($p<0.001$), as did all sleep variables. There were significantly more boys in the OSA group than in the non-OSA group ($p<0.001$). No differences in endothelial function could be found at baseline.

At baseline, correlations between endothelial function and other variables were investigated. PWA correlated with waist circumference ($r=0.27$, $p=0.003$) and systolic blood pressure ($r=0.23$, $p=0.011$). The augmentation index correlated with diastolic blood pressure ($r=-0.36$, $p<0.001$), WHR ($r=-0.21$, $p=0.026$) and desaturation nadir ($r=0.24$, $p=0.009$). However, the correlation between the augmentation index and desaturation nadir did not persist after correction for WHR ($p=0.06$). RHI correlated with diastolic blood pressure ($r=-0.24$, $p=0.009$) and mean saturation ($r=-0.25$, $p=0.008$). TPR correlated with diastolic blood pressure ($r=0.24$, $p=0.009$). All correlation coefficients were <0.4, indicating only a weak correlation between endothelial function and the other variables.

1.3.2 Follow-up data

Only those patients who received an Endo-PAT measurement after 10 months were included for further analysis. Of the 130 patients included at baseline, 72 patients fulfilled these criteria. Drop-out

was attributable to premature completion of the weight loss program in 52 of the patients, while 6 patients were excluded based on the absence of an Endo-PAT measurement or sleep assessment.

The patients who were not included for further analysis (58 patients) did not differ in sex (p=0.2), BMI z-score (p=0.2), age (p=0.7), oAHI (p=0.9) or TPR (p=0.2) compared to the 72 patients who were included. After 18 months, only 28 patients participated in the follow-up visit, corresponding to a drop-out rate of 61%. Children who dropped out of the study after the first follow-up visit were similar regarding all baseline characteristics, sleep variables and endothelial function compared to the other patients.

The patient characteristics of these 72 patients at the different study visits are shown in Table 2.

Throughout time, there was a significant improvement in obesity-related parameters. The fat percentage decreased from a mean of 43.2 ± 6.4% at enrollment to a mean of 32.5 ± 8.3% at 10 months ($p<0.001$). After 18 months, there was an increase in fat mass to 36.3 ±7.6% ($p<0.001$). The median absolute decrease in BMI z-score between baseline and 10 months was 0.9 (range = 0.1-1.9). BMI z-score and waist circumference did not change significantly between 10 and 18 months. Despite improvements in weight loss parameters after 10 months, only 23% of boys and 15% of girls reached a normal weight (35% remained obese and 42% overweight for boys; 35% obese and 50% overweight for girls).

Blood pressure decreased significantly from 123/69 (± 11/7) mmHg at enrollment to 116/63 (± 9/6) mmHg after 10 months of weight loss treatment ($p<0.001$) but increased again 6 months after ending the program to reach values that were similar to baseline values (128/69 (± 17/9); $p<0.001$).

At baseline, OSA was initially diagnosed in 47 of these 72 patients (65.3%). Ten of the patients had residual OSA at the repeated sleep screening at visit 2, which corresponds with a treatment success of 79%. The oAHI decreased significantly between the two sleep screenings ($p<0.001$), from a median of 2.74 per hour (range = 0.43-12.65) to 1.55 per hour (range = 0.00-4.66). The ODI also decreased

significantly (p<0.001), from a median of 1.8 per hour (range = 0.1-11.2) to 0.9 per hour (range = 0.0-5.8).

Concerning endothelial function, the RHI increased significantly after 10 months of treatment (p<0.001) and remained improved after 18 months (p=0.007) (1.40 (range = 0.79-2.89); 1.57 (range = -0.01-3.58) and 1.83 (range = 0.78-3.87), respectively). TPR shortened from 195 seconds at baseline (range = 45-285) to 135 seconds (range = 45-285) after 10 and 18 months (p=0.02 and p<0.001).

1.3.3 Relationship between OSA and endothelial function

The 47 patients who were diagnosed with OSA at baseline were further divided into a residual OSA and a normalized OSA group based on the second sleep screening after 10 months. Table 3 compares the baseline characteristics of patients who never had OSA, patients with residual OSA and those with a normalized sleep study. WHR and waist circumference were significantly lower in the non-OSA group than in both the normalized OSA (p= 0.007 and 0.003) and residual OSA groups (p=0.001 and 0.04) (Table 3). When comparing the variables between these groups after 10 months of treatment, a significantly lower RHI was found in the residual OSA group than in the normalized OSA group (p=0.04). The TPR was significantly longer in the residual OSA group than in the other groups (p=0.015). Regarding sleep variables, only the oAHI (p<0.001) and ODI (p=0.009) were significantly different between groups. These characteristics are presented in Table 4.

1.3.4 Linear mixed model

A linear mixed model was fitted with a random intercept and slope. The linear mixed model (Table 5) included time as a fixed factor and the oAHI and WHR as covariates, including their possible interactions. Only significant interaction terms were kept, and interaction terms were removed in order of complexity and for interactions with a similar number of terms, starting with the least significant interaction (first removal of terms with a p-value of 0.9, subsequently those with a p-value of 0.8, until only significant terms remained). Age and height did not contribute to TPR and were therefore not included in the final model. The final model showed that more severe OSA at baseline

was associated with a worse TPR at baseline (p=0.05), and a significant oAHI-time interaction was found (p=0.02). Thus, the model showed that the presence of OSA influenced endothelial function over time and that a higher oAHI was associated with less improvement in TPR after weight loss. Figure 1 shows the mean TPR evolution after 10 months of weight loss for patients with different OSA severities.

1.4 Discussion

In this longitudinal study we confirmed the effects of a long-term residential weight loss program on anthropometric parameters as well as on endothelial dysfunction and OSA. These beneficial effects for waist circumference, BMI z-score and endothelial function parameters remained significantly improved 6 months after ending the weight loss program, whereas we noticed a reversal of the beneficial effects for the fat percentage and systolic and diastolic blood pressure. We also showed an improvement in OSA in 79% of patients during a weight loss program in an obese pediatric population. Interestingly, patients with residual sleep apnea had worse endothelial function after 10 months of weight loss therapy than patients with normalized sleep function. Furthermore, OSA leads to less improvement in endothelial function over time. We thus provide evidence for the important impact of obstructive sleep apnea on endothelial function.

At baseline, very few significant differences were found between pediatric obese children with and without sleep apnea. Children with OSA had a significantly higher waist circumference and WHR, indicating a more central type of obesity. Our research group has consistently found more central obesity in obese children with OSA[10,27], which has also been confirmed by other studies[28,29].

Significant weight loss after treatment was observed in our population. However, despite this intense program with substantial weight loss, only a minority of the children (23% of boys and 15% of girls) reached a normal weight. Furthermore, 6 months after ending the weight loss program, a significant increase in obesity-related parameters was seen, although parameters mostly remained significantly different from baseline values. The findings underline the difficulty these patients encounter in maintaining their weight loss after leaving the rehabilitation center and continuing treatment at home on their own. Interestingly, the WHR continued to decrease after ending the program, meaning that even though there was weight regain, this did not necessarily lead to central obesity.

Obese children exhibit impaired endothelial function at both the macrovascular[30] and microvascular levels[31]. However, microvascular endothelial dysfunction appears to precede macrovascular

dysfunction[32], making it an important factor to study in a pediatric population. Indeed, the PAT technology used in this study investigated endothelial function at the microvascular level. An improvement in microvascular endothelial function after weight loss was seen in our population, as RHI and TPR improved between baseline and 10 months. This has also previously been proven by Bruyndoncxk et al[14]. In this cohort, we additionally followed the patients 6 months after ending the weight loss program. Eighteen months after the start of the study, both parameters of endothelial function remained stable even though there was weight regain. While endothelial function stabilized at these beneficial levels until the end of the study, blood pressure (both systolic and diastolic) rose again over the additional 6 months, returning to baseline values. Further research is necessary to confirm these long-term beneficial effects on endothelial function and to investigate the underlying mechanisms herein. The elevation of blood pressure after weight regain is especially important in children with OSA, as sleep apnea is an independent risk factor for hypertension[33].

No significant difference in endothelial function could be found at baseline between the obese children with and without sleep apnea. This stands in contrast to several studies that found a cross-sectional link between OSA and endothelial function in normal-weight children[34-36]. Bhattacharjee et al. investigated the interaction between obesity and OSA in prepubertal children and found that obesity and sleep apnea can independently increase the risk of endothelial dysfunction and that both interact to increase this effect[13]. It is possible that the direct effect of obesity overrules the independent effect of OSA on endothelial function in our population, as we did not compare the subjects to a normal-weight control population. It should also be noted that the population in the study by Bhattacharjee et al. was, regardless of weight, generally younger, more racially diverse and had more severe OSA than our population. After weight loss, we were able to demonstrate the detrimental effects of OSA on endothelial function. To the best of our knowledge, this is the first longitudinal study in an obese pediatric population to investigate the relationship between endothelial function and OSA. No differences in endothelial function were found at baseline between patients with residual OSA and those without OSA. After 10 months of weight loss, endothelial function ameliorated; however, this

improvement was significantly less pronounced in patients with residual OSA, as evidenced by a longer TPR and lower RHI. This was further supported by our linear mixed model, which showed less improvement in endothelial function in patients with OSA than in patients without OSA. Additionally, this effect seems to be dependent on the severity of OSA. This indicates that the resolution of OSA is an important factor in the improvement of endothelial function and should be considered when treating an obese child. Our results support the evidence that OSA makes an important contribution to the vascular damage seen in these patients[37]. Therefore, children with residual OSA after weight loss should be further treated for sleep apnea to avoid serious cardiovascular complications later in life. Gozal et al. demonstrated that endothelial function improves after adenotonsillectomy in normal-weight children with OSA[12]. However, we and others have shown that adenotonsillectomy is not the preferred first-line treatment for OSA in an obese child[11,15,38], and weight loss therapy should be considered first in obese children without adenotonsillar hypertrophy. In phase 2 of the NANOS study it was shown that adenotonsillectomy can help improve the severity of OSA in children with obesity[39]. The best treatment strategy for children with OSA and obesity remains complex and should be considered on an individual basis. However, in view of the high prevalence of OSA in obese children and because OSA can exacerbate the comorbidities of obesity in these children, an optimal treatment strategy should simultaneously target OSA and obesity. Other therapies in addition to weight loss therapy can be considered in obese children with OSA to separately tackle endothelial dysfunction. Several studies have shown positive effects of nutritional supplementation with n-3 fatty acids and antioxidants in this context[40-43]. Studies in adults have shown that even extensive weight loss and a related reduction in OSA lead to only a modest or even a nonsignificant reduction in blood pressure [44,45]. This could indicate that long-lasting structural cardiovascular changes are less sensitive to a nonpharmacological intervention, such as weight loss, in adult patients. Therefore, it is of utmost importance to treat cardiovascular comorbidities at an early stage when these comorbidities are still reversible.

A few limitations of our study should be considered. First, a portable screening device was used for the diagnosis of sleep-disordered breathing instead of the gold standard polysomnography, as this was not available in the rehabilitation center. As a result, we were not able to assess sleep architecture or identify arousals. However, the study by Lesser et al. showed the Apnealink device to be a sensitive and specific screening tool for the evaluation of OSA in obese children[46]. ApneaLink equipment was also attached and monitored overnight by skilled personnel. Second, we had an overall, substantial drop-out rate after 18 months of 61%. Obesity research and treatment are highly prone to compliance and motivational issues, as drop-out before the first follow-up visit was associated with full discontinuation of obesity treatment in 90% of cases. Third, as the majority of our population was Caucasian, our results might not be applicable to other patient populations.

In conclusion, both obesity and obstructive sleep apnea are important, independent predictors of endothelial function in a pediatric population. Although weight loss significantly improves endothelial function in obese children, it is less likely to improve in the presence of persistent OSA.

1.5 Acknowledgments

Special thanks to all colleagues from 'Het Zeepreventorium' and to Eddy Basslé specifically for helping us with the practical side of this study and providing us with a venue in which we could conduct all measurements. We also thank all patients and their parents for their participation in this study.

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1.7 Author contributions

All authors certify that they have participated in the study and take responsibility for the content. The study was designed by LB, BDW, KVH, SV and AVE. Measurements were performed by MY, EV and AVE. Technical support was provided by HH and ADG. The database was made by SJ, EM, MY, EV and AVE and statistical analysis was performed by SJ, EM, SV and AVE. Data interpretation was done by SJ, EM, SV and AVE. All authors were involved in the writing of the manuscript.

1.8 Funding

This research was made possible with financial support from the 'Research Foundation Flanders (Fonds voor Wetenschappelijk Onderzoek)', FWO-TBM project number 150179.

1.9 Figure legends

Figure 1: Mixed model analysis of TPR over time in function of OSA severity. The figure represents the average evolution of the TPR over 10 months weight loss of patients with different oAHI profiles.

Table 1: Baseline characteristics of patients with and without OSA.

	Non-OSA	OSA	p
N	43	87	
Sex (male/female)	8/35	46/41	<0.001 [1]
Age (years)	15 (10 - 19)	14 (8 - 19)	0.4 [2]
Height z-score	0.45 ± 1.1	0.75 ± 1.1	0.2 [3]
BMI z score	2.6 (1.4 - 3.7)	2.7 (1.8 - 3.6)	0.2 [2]
Fat percentage (%)	43.36 ±5.41	44.32 ±5.96	0.4 [3]
Waist (cm)	107.0 (88.3 - 141.0)	117.0 (87.0 – 165.0)	0.001 [2]
Waist-to-hip ratio	0.93 ±0.05	0.98 ±0.06	<0.001 [3]
Systolic BP (mmHg)	122 ±12.32	123.55 ±10.62	0.5 [3]
Systolic BP percentile	87 (12 – 99)	88 (22-99)	0.8 [2]
Diastolic BP (mmHg)	67.63 ±7.19	69.87 ±8.17	0.1 [3]
Diastolic BP percentile	56 ± 24	62 ± 23	0.2 [3]
oAHI (/hour)	1.33 (0.43 - 1.99)	3.65 (2.01 - 56.86)	<0.001 [2]
SaO_2 (%)	97 (95 - 98)	96 (93 - 98)	<0.001 [2]
SaO_2 nadir (%)	93 (83 - 96)	90 (72 - 94)	<0.001 [2]
ODI (/hour)	0.8 (0.1 - 3.6)	2.9 (0.3 - 47.3)	<0.001 [2]
MaxD	1.21 (0.96 - 2.51)	1.24 (0.90 - 2.39)	0.5 [2]
Augmentation index (%)	-9.34 (-33.09 - 4.41)	-12.27 (-41.25 - 7.30)	0.1 [2]
RHI	1.42 (1.00 - 2.41)	1.40 (0.79 - 4.01)	0.4 [2]
TPR (s)	165 (45 - 285)	195 (45 - 285)	0.2 [2]
PWA	602.60 (71.54 - 1203.71)	560.53 (90.98 - 1361.99)	0.8 [2]

Results are presented as mean ± standard deviation or median (minimum –maximum).

BP, blood pressure; oAHI, obstructive apnea hypopnea index; ODI, oxygen desaturation index; SaO_2, mean saturation; SaO_2nadir, saturation nadir; MaxD, maximum dilatation after occlusion; RHI, reactive hyperemia index; TPR, time to peak response; PWA, mean baseline pulse wave amplitude

[1] Chi²

[2] Mann-Whitney U

[3] Independent-samples T test

(footnote: one patient had recently turned 19 years at the moment of the baseline measurements, the patient still fulfilled the inclusion criteria (<18 years) at the moment of inclusion).

Table 2: Patient characteristics at baseline, after 10 months and after 18 months follow-up

	Baseline	10 months	18 months	p
N	72	72	28	
Sex (male/female)	26/46	26/46	12/16	0.9 [4]
Age (years)	14.9 (8.4 – 19)	15.7 (9.2 – 19.9)	16.0 (9.8 – 19.6)	
Height z-score	0.41 ± 1.04	0.41 ± 1.02	0.37 ± 1.00	0.001 [2,#,£]
BMI z-score	2.7 (1.4 – 3.4)	1.7 (0.5 – 2.7)	1.75 (0.4-2.9)	<0.001 [1,$,#]
Fat percentage (%)	43.3 ±6.4	32.5 ±8.3	36.3 ±7.6	<0.001 [2,$,#,£]
Waist circumference (cm)	112.5 (87 – 150)	92 (75 – 124.8)	84 (69.7-119)	<0.001 [1,$,#]
Waist-to-hip ratio	0.96 ±0.06	0.92 ±0.07	0.87 ±0.07	<0.001 [2,#,£]
Systolic BP (mmHg)	123.36 ±10.68	115.54 ±8.88	127.93 ±17.14	<0.001 [2,$,£]
Systolic BP percentile	90 (38 – 99)	67 (8 – 98)	94 (13 – 99)	<0.001 [1,$,£]
Diastolic BP (mmHg)	69.46 ±7.19	63.36 ±6.19	68.75 ±9.28	<0.001 [2,$,£]
Diastolic BP percentile	65 (10 – 99)	40 (5 – 88)	63 (9 – 99)	<0.001 [1,$,£]
oAHI (/hour)	2.74 (0.43 – 12.65)	1.55 (0.00 – 4.66)	/	<0.001 [3]
SaO_2 (%)	97 (94 – 98)	97 (95 – 98)	/	0.02 [3]
SaO_2 nadir (%)	91 (72-96)	92 (72-95)	/	0.04 [3]
ODI (/hour)	1.8 (0.1 – 11.2)	0.9 (0.0 – 5.8)	/	<0.001 [3]
MaxD	1.24 (0.96 – 2.51)	1.29 (0.95 – 3.59)	1.71 (0.84 – 3.98)	0.2 [1]
Augmentation Index (%)	-10.46 (-40.35 – 7.30)	-8.42 (-51.54 – 15.69)	-9.80 (-27.33 – 13.88)	0.9 [1]
RHI	1.40 (0.79 – 2.89)	1.58 (1.14 – 3.58)	1.83 (0.78 – 3.87)	0.01 [1,$,#]
TPR (s)	195 (45 – 285)	135 (45 – 285)	135 (45 – 225)	0.002 [1,$,#]
PWA	563.67 (71.54 – 1352.05)	541.26 (52.58 – 1103.26)	246.65 (45.41 – 1527.36)	0.3 [1]

Results are presented as mean ± standard deviation or median (minimum –maximum).

BP, blood pressure; oAHI, obstructive apnea hypopnea index; ODI, oxygen desaturation index; SaO_2, mean oxygen saturation; SaO_2nadir, saturation nadir; MaxD, maximum dilatation; RHI, reactive hyperemia index; TPR, time to peak response; PWA, pulse wave amplitude;

[1] Friedman test

[2] Repeated measures ANOVA

[3] Wilcoxon Signed-Rank Test

[4] Chi^2

$: significant difference between baseline and 10 months follow-up

#: significant difference between baseline and 18 months follow-up

£: significant difference between 10 months follow-up en 18 months follow-up

Table 3: Baseline patient characteristics of patients without OSA, with a normalized sleep study and residual OSA

	Non OSA	Normalized OSA	Residual OSA	p
N	25	37	10	
Sex (male/female)	4/21	23/14	2/8	0.002 [3,#]
Age (years)	14.4 ± 2.1	14.3 ± 2.3	13.7 ± 2.6	0.6 [2]
Height z-score	0.09 ± 0.98	0.67 ± 1.06	0.25 ± 0.96	0.08 [2]
BMI Z-score	2.5 (1.4-3.3)	2.7 (1.8-3.4)	2.7 (2.3-3.2)	0.3 [1]
Fat percentage (%)	42.8 ± 5.9	43.4 ± 7.4	43.5 ± 3.4	0.9 [2]
Waist circumference (cm)	106 (88-134)	117 (87-144)	119 (95-150)	0.001 [2,$,#]
Waist-to-hip ratio	0.92 ± 0.05	0.97 ± 0.06	1.00 ± 0.07	0.001 [2,$,#]
Systolic BP (mmHg)	121 ± 11	125 ± 10	125 ± 14	0.4 [2]
Systolic BP percentile	84 (51-99)	90 (38-99)	87 (55-99)	0.4 [1]
Diastolic BP (mmHg)	67 ± 6	71 ± 7	70 ± 10	0.1 [2]
Diastolic BP percentile	59 (10-94)	69 (26-99)	59 (20-99)	0.3 [1]
oAHI (/hour)	1.6 (0.4-1.9)	3.46 (2.13 – 12.65)	4.58 (2.49 – 7.24)	<0.001 [1,$,#]
SaO_2 (%)	97 (95-98)	96 (94 – 98)	96.5 (94 – 98)	0.03 [1,$]
SaO_2 nadir (%)	93 (83-96)	91 (72 – 93)	89.5 (84 – 93)	<0.001 [1,$,#]
ODI (/hour)	0.8 (0.1-2.2)	2.5 (0.3 – 11.2)	3.55 (0.8 – 6.3)	<0.001 [1,$,#]
MaxD	1.23 (0.96-2.51)	1.24 (1.04-1.98)	1.33 (1.01-2.39)	0.6 [1]
Augmentation index (%)	-8.12 (-27.65-4.41)	-12.79 (-40.35 – 7.30)	-12.53 (-34.23 - -6.20)	0.2 [1]
RHI	1.42 (1.00-2.32)	1.40 (1.04 – 2.21)	1.22 (0.79 – 2.89)	0.2 [1]
TPR (s)	165 (45-285)	165 (75 – 285)	210 (105 -255)	0.4 [1]
PWA	610 (88-1089)	575 (245-1509)	515 (186-1184)	0.8 [1]

Results are presented as mean ± standard deviation or median (minimum –maximum).

Systolic BP, systolic blood pressure; diastolic BP, diastolic blood pressure; oAHI, obstructive apnea hypopnea index; ODI, oxygen desaturation index; SaO2, saturation; SaO2nadir, saturation nadir; Percentage of time with SaO2 <90%, percentage of time with saturation below 90%; MaxD, maximum dilatation; AI, augmentation index; RHI, reactive hyperemia index; Tmax, time to maximum dilatation; PWA, pulse wave amplitude; hs-CRP, high sensitive C-reactive protein

[1] Independent Sample Kruskal-Wallis Test

[2] One-Way ANOVA

[3] Chi²

$: significant difference between non-OSA and normalized OSA

#: significant difference between non-OSA and residual OSA

£: significant difference between normalized OSA and residual OSA

Table 4: Follow-up patient characteristics after 10 months weight loss treatment of patients without OSA, with a normalized sleep study and residual OSA

	Non OSA	Normalized OSA	Residual OSA	p
N	25	37	10	
Age (years)	15.4 ± 2.2	15.1 ± 2.1	14.7 ± 2.7	0.7 [2]
Height z-score	0.11 ± 0.98	0.66 ± 1.04	0.25 ± 0.98	0.1 [2]
BMI Z-score	1.5 (0.7 – 2.7)	1.7 (0.5 – 2.5)	1.9 (1.1 – 2.5)	0.4 [1]
Fat percentage (%)	33.16 ±7.11	32.78 ±9.91	29.54 ±2.93	0.5[2]
Waist circumference (cm)	90 (78 – 124)	93 (75 – 124.8)	99.5 (75 – 116)	0.3 [1]
Waist-to-hip ratio	0.90 ±0.05	0.92 ±0.07	0.96 ±0.09	0.058 [1,#]
Systolic BP (mmHg)	115 ±9	116 ± 8	118 ± 11	0.6 [2]
Systolic BP percentile	63 (18-98)	67 (8-98)	71 (9-96)	0.7 [1]
Diastolic BP (mmHg)	64 ±6	63 ± 7	61 ± 6	0.6 [2]
Diastolic BP percentile	39 (12-88)	41 (5-88)	32 (5-68)	0.7 [1]
oAHI (/hour)	/	1.35 (0.00 – 1.98)	2.70 (2.00 – 4.66)	<0.001 [3]
SaO_2 (%)	/	97 (95 – 98)	97 (96 – 98)	0.8 [3]
SaO_2 nadir (%)	/	92 (72 – 95)	91 (87 – 95)	0.7 [3]
ODI (/hour)	/	0.7 (0.0 - 2.8)	1.8 (0.0 - 5.8)	0.005 [3]
MaxD	1.20 (0.95 - 3.59)	1.30 (1.00 - 3.36)	1.31 (1.06 - 2.70)	0.4 [1]
Augmentation index (%)	-7.74 (-17.73 - 15.69)	-9.11 (-51.54 - 5.23)	-7.20 (-14.01 - 5.09)	0.6 [1]
RHI	1.49 (1.21 - 2.86)	1.63 (1.14 - 3.58)	1.48 (1.18 - 2.32)	0.05 [1,£]
TPR (s)	135 (45 - 255)	105 (45 - 285)	225 (75 - 285)	0.015 [1,#,£]
PWA	544 (50-936)	534 (46-978)	687 (145-1189)	0.3 [1]

Results are presented as mean ± standard deviation or median (minimum –maximum).
Systolic BP, systolic blood pressure; diastolic BP, diastolic blood pressure; oAHI, obstructive apnea hypopnea index; ODI, oxygen desaturation index; SaO2, saturation; SaO2nadir, saturation nadir; Percentage of time with SaO2 <90%, percentage of time with saturation below 90%; MaxD, maximum dilatation; AI, augmentation index; RHI, reactive hyperemia index; Tmax, time to maximum dilatation; PWA, pulse wave amplitude; hs-CRP, high sensitive C-reactive protein

[1] Independent Sample Kruskal-Wallis Test

[2] One-Way ANOVA

[3] Mann-Whitney U

$: significant difference between non-OSA and normalized OSA

#: significant difference between non-OSA and residual OSA

£: significant difference between normalized OSA and residual OSA

Table 5: Mixed model analysis of TPR over time in function of OSA severity (BIC= 1260.5).

Parameter	Estimation ± standard deviation	p
Intercept	28.25 ± 111.79	0.8
oAHI	21.18 ± 10.75	0.05
WHR	95.68 ± 120.42	0.4
Time	-42.06 ± 161.99	0.8
Time*oAHI	-25.74 ± 11.20	0.02
Time*WHR	129.50 ± 173.13	0.5