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Association of Habitual Snoring with Carotid Intima-Media Thickness in Young Males

Sarah C. Jacoby

A Thesis Submitted to the Graduate Faculty of

JAMES MADISON UNIVERSITY

In

Partial Fulfillment of the Requirements

for the degree of

Master of Science

Department of Kinesiology

May 2013

Acknowledgements

I would first like to thank Dr. Trent A. Hargens for his mentorship and for being my Committee Chair. I could have not have successfully completed my thesis project without his guidance and support. This thesis project has been a challenging but extremely beneficial and rewarding experience. I feel very fortunate to have worked with such a knowledgeable and well-respected researcher in the field of sleep apnea.

I would also like to thank Dr. Mikel K. Todd and Dr. Elizabeth S. Edwards for serving as members of my thesis committee. Their time and expertise contributed greatly to the quality of our research and writing.

Finally, I would like to thank Niki Hafner, Brooke Shafer, Justin Faller, Ali Fracasso, Courtney Sutton, Taylor Wenos, and Laura Shupe for their support and encouragement throughout this process. We have shared many enjoyable experiences together in the classroom, in the office, and outside of school. I will always look back on my two years spent here at JMU fondly.

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ABSTRACT

Purpose The primary aims of this study are to: 1) compare carotid intima media thickness (CIMT) of young males who are habitual snorers compared to those who do not snore; 2) to compare CIMT between snorers and non-snorers to the intima media thickness (IMT) of the brachial artery, the control, that is not exposed to the vibrations from snoring; and 3) to determine the risk for developing obstructive sleep apnea (OSA) in snorers compared to non-snorers through the use of validated questionnaires. *Methods* Subjects were classified as snorers (n=17) or non-snorers (n=6) according to the prescreening questionnaire. Height, weight, neck and waist circumferences, body composition, physical activity level, heart rate variability and accelerometer data were collected on each subject. Brachial and carotid IMT was imaged via ultrasonography; IMT measurements were compared between groups using brachial IMT as the control to compare CIMT between groups. *Results* No significant group differences were noted for any study variable, however a correlation was found between CIMT and physical activity (PA) (expressed as MET-min/wk) ($r=0.65$, $p = .001$). After controlling for PA, mean CIMT was greater in snorers compared to non-snorers (0.367 mm vs. 0.310 mm respectively; $p=.014$). Brachial IMT did not differ between groups. Questionnaire data showed that 58.8% of snorers were at high risk for developing OSA compared to 0% for non-snorers. *Conclusion* Results suggest that the vibrations from snoring may contribute to vascular remodeling in the carotid artery, and may be an early mechanism contributing to the development of endothelial dysfunction, and early subclinical sign of CVD risk in those at high risk for OSA.

CHAPTER I

Introduction

Obstructive sleep apnea (OSA) is a common sleep disorder, characterized by interrupted ventilation during sleep combined with excessive daytime sleepiness [1]. OSA affects an estimated 15 million adult Americans and is considered the most prevalent form of sleep-disordered breathing [1, 2]. Despite the high prevalence of OSA, it is estimated that as many as 80-90% of those 15 million individuals with OSA remain undiagnosed [1, 3-5]. OSA is defined as the intermittent cessation of breathing caused by complete (apnea) or partial (hypopnea) collapse of the airway due to an imbalance of forces in the pharynx during sleep [6]. Apnea is the complete cessation of airflow for ≥ 10 seconds due to complete collapse of the airway and hypopnea is a ≥ 50% reduction in airflow for ≥ 10 seconds or ≥ 30% reduction in airflow accompanied by a ≥ 4% decrease in oxygen saturation due to partial collapse of the airway [1]. The reduced airflow and decreased oxygen saturation create conditions of hypoxemia and hypercapnia as ventilation becomes inadequate to meet the metabolic demands of the body. Hypoxemic and hypercapnic conditions stimulate the peripheral chemoreceptors located in the carotid bodies of the internal carotid arteries to transmit signals to the central nervous system to increase sympathetic nervous system (SNS) activity. As a result, the interrupted breathing event is terminated and normal ventilation is reestablished [7]. The individual cycles between sleep and wakefulness as the SNS is repeatedly stimulated, resulting in the persistence of a pattern of hypoxemia and hypercapnia [7]. The inability to sleep

continuously and restoratively throughout the night often results in excessive daytime sleepiness, a key symptom of OSA [6].

The pathophysiology of OSA is complex; a variety of factors predispose to upper airway narrowing and collapse during sleep within an individual [8]. Factors predisposing one to OSA include mechanical properties and collapsibility of the upper airway, ability to reflexively activate pharyngeal muscles, respiratory control system stability, decreased lung volume, and the predisposition to awakening during sleep [1, 9]. The upper airspace region of the pharynx, the pharyngeal airway, is composed of muscular and soft tissues in contrast to other regions of the respiratory tract such as the trachea and bronchi that are surrounded by more rigid, cartilaginous support [9]. The plasticity of the pharyngeal airway allows for essential, non-respiratory functions such as speaking, chewing, and swallowing [9]. However, its collapsibility presents a potential problem for the essential respiratory function of the upper airway because the passage must remain open during breathing to allow for sufficient ventilation and gas exchange [9]. Sleep, especially rapid eye movement (REM) sleep, causes reductions in muscular tone of the pharyngeal airway, resulting in narrowing of the airway and increased resistance to airflow [10]. These changes contribute significantly to hypoventilation and increased arterial Pco_2 in normal sleeping subjects [11].

The primary abnormality in patients with OSA is an anatomically small pharyngeal airway resulting from obesity and/or bone and soft tissue structures [1]. Obesity causes dynamic loading of the airway due to the weight of fatty tissue in the neck, resulting in minor collapse [12]. When awake, OSA patients experience increased airflow resistance and greater intrapharyngeal negative pressure during inspiration [1]. In

response to the negative pressure, mechanoreceptors, located primarily in the larynx, increase activity of pharyngeal dilatory muscles, thereby maintaining airway patency while awake [1]. In individuals with already anatomically narrow airways, the suppressant effects of sleep on pharyngeal muscle tone predisposes them to airflow limitations (snoring and hypopneas) and complete airway closures (obstructive apneas) [9]. During sleep, the reflex pharyngeal muscle activity that drives this neuromuscular compensation is reduced or lost, leading to pharyngeal narrowing and intermittent complete collapse [1]. Resulting apneas or hypopneas stimulate effort to breath and ultimately arouse the individual from sleep to terminate the apneic event [1]. Thus, an upper airway that requires reflex driven muscle activation to remain open during the wakefulness may be at risk for collapse during sleep [1].

Large population studies have helped to establish numerous demographic correlates and risk factors for OSA [5, 13]. OSA is more prevalent in males than females, with a patient case ratio of 8:1 [4]. Sex differences in the diagnosis of OSA are attributed to differences in clinical presentation and differences in pathophysiology [14]. It is also thought that sex differences in prevalence may be explained by the likelihood that women are under diagnosed with OSA due different expression of clinical symptoms [16]. Men usually present with typical symptoms such as snoring or daytime sleepiness, while women are more likely to present atypical symptoms of insomnia or fatigue or have concomitant clinical depression or hypothyroidism [15]. Atypical symptoms in women compared to men can cause diversion from the OSA diagnosis. Mechanisms that are thought to play a role in the prevalence of OSA diagnosis between sexes include differences in obesity, upper airway anatomy, breathing control, hormones, and aging

[17]. The prevalence of OSA increases with age; with a 2 to 3 fold higher prevalence in older individuals (>65 years old) compared with those in middle age (30-64 years old) [4]. As with many other chronic diseases, OSA risk increases with age due to physiological mechanism related to aging and due to increased risk for developing CVD with age. OSA and CVD are strongly associated together, for example, atrial fibrillation and congestive heart failure are considered risk factors for developing OSA; risk for both of these diseases increases with age [18].

Established risk factors for OSA include habitual snoring, overweight and obesity, central body fat distribution, large neck girth, and craniofacial and upper airway abnormalities [4]. There is a graded increase in OSA prevalence with increasing body mass index, neck circumference, and waist to hip ratio [4]. Craniofacial and upper airway structure play an important role in the occurrence of OSA, as these contribute to the anatomy and potential narrowing of the upper airway [4, 9]. In addition, suspected risk factors for OSA include genetics, smoking, menopause, alcohol use before sleep, and nighttime nasal congestion [4]. Several studies have shown an increased risk of OSA in families of patients with the breathing disorder [4]. This association may suggest that risk factors come from similar lifestyles or may indicate a genetic basis to OSA [5]. Smoking is a possible risk factor; however, this has only been reported in few studies. Possible mechanisms for the role of smoking include airway inflammation, smoking related diseases, and declining blood nicotine levels on sleep stability [4]. Hormonal differences are believed to account for the sex difference in OSA prevalence. Because sex hormone levels change dramatically with menarche, pregnancy, and menopause, it is plausible that these changes modify the risk of OSA [4]. Experimental studies show an acute effect of

alcohol on apnea and hypopnea frequency but the effect of long-term alcohol use on the development or progression of OSA is unknown [5]. Nasal congestion at night, whether due to allergies, acute upper respiratory tract infection, or anatomy, has been linked to snoring and OSA in both experimental and epidemiological studies [5]. OSA patients also exhibit higher prevalence of hypertension, type II diabetes, cardiovascular disease, and stroke compared to those without OSA [1, 18-24].

The physiological mechanisms linking OSA to other chronic diseases are complex and inadequately understood. However, autonomic dysregulation and endothelial dysfunction provide key explanations for the association between OSA and chronic disease [1, 25-32]. The first mechanism listed above, autonomic dysregulation, has many mechanisms are thought to contribute to it; these include hyperleptinemia, increased sympathetic activity, systemic inflammation, oxidative stress, and impaired baroreflex [12]. OSA-related oxygen desaturation and accompanying apnea/hypopnea events promote degenerative remodeling of the arterial wall and may be an early sign of subclinical atherosclerosis [20, 22, 33-35]. The extent of hypertrophy damage in the carotid artery caused by remodeling appears to correlate with frequency and intensity of hypoxemia [34]. Measurement of the thickness of the intima-media layer of the carotid artery (CIMT), through ultrasound imaging, has been demonstrated to be a valid and dependable non-invasive marker of subclinical atherosclerosis [36, 37]. CIMT thickness indicates a higher level of CVD risk by indicating the presence of early atherosclerosis and wall hypertrophy [34].

A second plausible mechanism is autonomic dysregulation induced by hypoxemia. Severity of OSA measured by the frequency and intensity of hypoxemia

appears to be significant in determining development of atherosclerosis independent of traditional CVD risk factors [36-42]. Heart rate variability (HRV) is a measure of autonomic effect on the heart; OSA patients have been shown to have varying levels of sympathetic stimulation corresponding to O_2 levels that cause respective changes in heart rate [43]. Autonomic function impairment indicates severity of hypoxemia in OSA patients and is associated with HRV patterns [43]. Hypoxemia causes increased sympathetic stimulation due to chemoreceptor activation; this leads to vasoconstriction and increased arterial pressure that contribute to carotid wall damage [43, 44]. Previous studies based on muscle sympathetic nerve activity (MSNA) have shown increased sympathetic activity during sleep apneic events as well as a sustained increase in MSNA during daytime awake rest [7]. Another study focusing on the sympathetic system found patients with moderate to severe OSA to have a higher mean heart rate and reduced heart rate variability. Patients with reduced total variance in heart rate are commonly found to have some form of heart failure or dilated cardiomyopathy [45]. These authors suggest that the repetitive sympathetic activation and blood pressure surges that occur in response to apneic episodes during sleep might cause an impairment of baroreflex and other cardiovascular reflexes that carry over, even into daytime wakefulness [45].

Previous studies have suggested that snoring and OSA may be important risk factors in the development of carotid atherosclerosis and stroke [46, 47]. A more recent study investigated whether snoring is an independent risk factor for developing carotid atherosclerotic plaque [22]. Heavy snoring was objectively measured and found to be a significant risk factor for the presence of atherosclerosis compared to participants who did not snore [22]. The relationship remained after adjustments were made for other

atherosclerotic risk factors and was independent of other measures of sleep-disordered breathing and hypoxia [22]. One study found that middle-aged patients with severe OSA, but without overt cardiovascular diseases, demonstrated early signs of atherosclerosis by means of increased arterial stiffness and carotid remodeling [20]. The experimental and control groups presented no differences in classical cardiovascular risk factors, the vascular abnormalities are independently associated with OSA [20].

Atherosclerosis involves several interrelated processes including endothelial dysfunction, inflammation, oxidative stress, vascular smooth cell activation, platelet activation, and thrombosis. OSA is also associated with the majority of these factors; venous [48] and arterial [26, 30] endothelial dysfunction, systemic inflammation [13], increased levels of plasma VEGF (vascular endothelial growth factor) [25], reactive oxygen species [49], soluble adhesion molecules [50], coagulation factors [51], and endothelin-1 [32] have all been reported in patients with OSA. Each of these factors contributes to the development of atherosclerosis and CVD by extension in OSA patients and may help to explain the link between OSA and CVD.

The pathophysiological mechanism linking snoring, OSA, and stroke are still unclear and remain widely unexplored. Carotid atherosclerosis is a leading cause of stroke and is a complex disease involving several highly interrelated processes, including endothelial dysfunction, inflammation, oxidative stress, vascular smooth muscle cell activation, platelet activation, and thrombosis [35]. Recent studies have indicated that OSA is also associated with many of these processes [1, 7, 13, 24].

In the absence of coexisting OSA, the prevailing view of snoring is that there is little evidence to support a pathological role in development of vascular disease [52, 53].

However, a recent study done by Lee, et al. (2008) identified heavy snoring, in the absence of hypoxemia, as an independent risk factor for carotid atherosclerosis. Vibration-induced endothelial dysfunction in rabbits provides a plausible pathophysiological mechanism linking snoring and localized carotid atherosclerosis [28, 54]. Endothelial dysfunction plays a pivotal role in the early pathogenesis of atherosclerosis and can be detected through CIMT measurement in patients with sleep-disordered breathing [28]. Self-reported snoring has been shown to be significantly associated with increased CIMT measures, independent of traditional CVD risk factors [23]. Studies to date have used middle aged and older males as participants; the extent to which CIMT remodeling occurs in young men is unknown. Identifying snoring in young men as an early marker of subclinical atherosclerosis could have substantial public health implications for the management and early detection in the cascade of events from habitual snorer, to OSA diagnosis, to overt CVD development [22].

Purpose

The primary aims of this study are to: 1) compare carotid intima media thickness (CIMT) of young males who are habitual snorers compared to those who do not snore; 2) to compare CIMT between snorers and non-snorers to IMT of the brachial artery, the control, that is not exposed to the vibrations from snoring; and 3) to determine the risk for developing obstructive sleep apnea (OSA) in snorers compared to non-snorers through the use of validated questionnaires.

Hypothesis

Young males who habitually snore will have higher CIMT measurements compared to age-matched non-snorers. The relationship between CIMT and brachial IMT in snorers will demonstrate carotid endothelial dysfunction compared to non-snorers. Finally, validated questionnaires will indicate a greater risk for developing OSA in snorers compared to non-snorers.

Limitations

- The study included only those individuals classified as overweight-to-obese according to body mass index (BMI) classifications (overweight = BMI > 25 kg/m^2)
- The snoring group consisted of college-aged males only, aged 18-26, with varying physical activity levels.
- The potential effect of alcohol was not controlled for, however, anecdotally subjects who mentioned alcohol affecting their snoring were asked to abstain for the night they wore the accelerometer.

Delimitations

- Subjects must be non-smokers, free from any known form of CVD, pulmonary, or metabolic disease.
- Subjects currently using medications that may influence the hemostatic regulations under investigation will be excluded from the study.

Significance of the Study

Snoring occurs commonly among the general population, it is estimated that as many as 47.7% of men and 33.6% of women snore [23]. Several studies have determined plausible mechanisms for snoring being an important, self-reported risk factor for atherosclerosis and eventually CVD [22, 23, 28, 54]. The vibrations to the carotid artery caused by snoring have been shown to be associated with endothelial dysfunction. In addition, habitual snorers have been found to have greater CIMT measures independent of other traditional CVD risk factors compared to non-snorers. This association has been observed in older and middle-aged males but no studies have been done on young males to date. This study will be the first to determine a potential association between snoring and CIMT; if a link can be established between young male habitual snorers and increased CIMT measurements then snoring could become a significant, self-diagnosable, subclinical risk factor for atherosclerosis and CVD.

Definition of Terms

1. Apnea: Complete cessation of airflow for ≥ 10 seconds.
2. Hypopnea: ≥ 50% reduction in airflow for ≥ 10 seconds or ≥ 30% reduction in airflow accompanied by a ≥ 4% decrease in oxygen saturation.
3. Obstructive Sleep Apnea (OSA): A common sleep disorder characterized by repeated complete or partial upper airway obstructions during sleep which causes loud snoring, oxygen desaturation, exaggerated sympathetic nervous system response, and frequent arousals to re-establish breathing.

CHAPTER II

Methodology

Participants

Twenty-three (23) males from the James Madison University (JMU) and Harrisonburg, Virginia communities were recruited for this study. Subjects were non-smokers, free from any known CVD, pulmonary, or metabolic disease. The use of medications that may influence the hemostatic parameters under investigation were cause for exclusion from the study. All subjects were classified as overweight-to-obese according to body mass index (BMI) classifications (overweight = BMI > 25 kg/m^2) [55]. Subjects were asked to report to the Human Performance Laboratory (HPL) on two consecutive days, for a total of two hours (one hour each visit). All subjects were given signed consent forms to read and sign that provided a comprehensive description of the study, the risks and benefits associated with the study, and explained how confidentiality would be maintained. All procedures were approved by the JMU Institutional Review Board prior to initiation of this study.

Visit 1

Questionnaires

A prescreening questionnaire was used to determine if volunteers met the subject criteria and it was used to classify subjects as 'snorers' or 'non-snorers'. After consent was obtained, subjects were asked to complete a health history questionnaire providing basic health information and identifying any potential exclusion criteria. Subjects were then asked to complete three additional questionnaires: the Berlin Questionnaire (BQ), STOP

(**S**creening **T**ool for **O**bstructive **S**leep apnea) questionnaire, and Epworth Sleepiness Scale to assess their risk factors for OSA, level of snoring, and level of daytime sleepiness respectively. To account for the impact of habitual physical activity, the International Physical Activity Questionnaire (IPAQ-SF) short form was used to assess current physical activity level in all subjects.

Body Composition

Upon completion of all the questionnaires, subjects had their body weight measured using a physician's scale and recorded to the nearest 0.1 kg, and height measured and recorded to the nearest 0.5cm. Waist and neck circumference measurements were also taken according to the American College of Sports Medicine (ACSM) guidelines using a cloth tape measure with spring-loaded handle, to reduce compression of the skin [55]. Estimates of percent body fat were then made via dual-energy x-ray absorptiometryer (DEXA) (Delphi QDR; Hologic; Boston, MA). The DEXA scan consisted of total body/abdominal adiposity and bone density.

Resting Blood Pressure

Following these tests, the subjects' resting blood pressure was measured according to ACSM procedures [55].

Actigraph Sleep Assessment

At the end of the first visit, subjects were instructed on proper setup and usage of the Actigraph accelerometer (GT-3X plus; Actigraph; Pensacola, FL), to wear while sleeping for one night outside the lab. This small, lightweight device was used to measure and store data about their movement during sleep. The accelerometer was worn on the subjects' non-dominant wrist during sleep. Subjects were instructed to wear the

Actigraph at night and record the times they went to bed and the time they got out of bed on a log sheet. Variables measured during sleep included sleep/wake activity, sleep onset, sleep latency, amount of sleep and sleep efficiency. Actilife 6 Data Analysis Software provided sleep-scoring values using several validated scoring algorithms.

Visit 2

Ultrasonography

Subjects were asked to return to the HPL one day after the first visit to return the Actigraph device for data download and analysis. Subjects then underwent measurements of carotid intima media thickness (CIMT) and brachial artery wall thickness via ultrasonography (DC-6; Mindray; Mahwah, NJ). For the CIMT and brachial measurements, subjects were brought to a quiet room to lie supine for ten minutes. After this time, a portable ultrasound machine and gel were used to obtain and save ultrasound images of the carotid and brachial arteries for later analysis. For the CIMT measurement, the participant was instructed to lie in a supine position with the head slightly extended and turned in the opposite direction of the carotid artery being studied. For the brachial measurement, the participant was asked to lay their arm supine on the table while images were saved.

Statistical Analysis

All variables were compared using independent sample t-tests, to assess group differences of study variables. Pearson correlations were calculated to identify potential relationships between CIMT, brachial artery wall thickness, and other study variables. Analysis of covariance (ANCOVA) was then employed to examine group differences with CIMT or brachial artery wall thickness as the dependent variable, and any

significant correlation as the covariate. For example, group difference in CIMT was used as the dependent variable and the significant correlation between CIMT and PA was used as the covariate. An alpha level of $p<0.05$ was set as the *a priori* statistical significance.

Chapter III

Manuscript

Association of Habitual Snoring with Carotid Intima-Media Thickness in Young Men

Abstract

Purpose The primary aims of this study are to: 1) compare carotid intima media thickness (CIMT) of young males who are habitual snorers compared to those who do not snore; 2) to compare CIMT between snorers and non-snorers to the intima media thickness (IMT) of the brachial artery, the control, that is not exposed to the vibrations from snoring; and 3) to determine the risk for developing obstructive sleep apnea (OSA) in snorers compared to non-snorers through the use of validated questionnaires. *Methods* Subjects were classified as snorers (n=17) or non-snorers (n=6) according to the prescreening questionnaire. Height, weight, neck and waist circumferences, body composition, physical activity level, heart rate variability and accelerometer data were collected on each subject. Brachial and carotid IMT was imaged via ultrasonography; IMT measurements were compared between groups using brachial IMT as the control to compare CIMT between groups. *Results* No significant group differences were noted for any study variable, however a correlation was found between CIMT and physical activity (PA) (expressed as MET-min/wk) ($r=0.65$, $p = .001$). After controlling for PA, mean CIMT was greater in snorers compared to non-snorers (0.367 mm vs. 0.310 mm respectively; $p=.014$). Brachial IMT did not differ between groups. Questionnaire data showed that 58.8% of snorers were at high risk for developing OSA compared to 0% for non-snorers. *Conclusion* Results suggest that the vibrations from snoring may contribute

to vascular remodeling in the carotid artery, and may be an early mechanism contributing to the development of endothelial dysfunction, and early subclinical sign of CVD risk in those at high risk for OSA.

Introduction

Obstructive sleep apnea (OSA) is a common sleep disorder that is characterized by interrupted ventilation during sleep combined with excessive daytime sleepiness [1]. OSA affects an estimated 15 million adult Americans and is considered the most prevalent form of sleep-disordered breathing [1, 2]. Despite the high prevalence of OSA, it is estimated that as many as 80-90% of those 15 million individuals with OSA remain undiagnosed [1, 3-5]. OSA is defined as the intermittent cessation of breathing caused by complete (apnea) or partial (hypopnea) collapse of the airway due to an imbalance of forces in the pharynx during sleep [6]. Apnea is defined as the complete cessation of airflow for ≥ 10 seconds due to complete collapse of the airway and hypopnea is defined as ≥ 50% reduction in airflow for ≥ 10 seconds or ≥ 30% reduction in airflow accompanied by a ≥ 4% decrease in oxygen saturation due to partial collapse of the airway [1]. The reduced airflow and decreased oxygen saturation create conditions of hypoxemia and hypercapnia as ventilation becomes inadequate to meet the metabolic demands of the body. Hypoxemic and hypercapnic conditions stimulate the peripheral chemoreceptors located in the carotid bodies of the internal carotid arteries to transmit signals to the central nervous system to increase sympathetic nervous system (SNS) activity. As a result, the interrupted breathing event is terminated and normal ventilation is reestablished [7]. The individual cycles between sleep and wakefulness as the SNS is

repeatedly stimulated and, as a result, the pattern of hypoxemia and hypercapnia persists [7]. The inability to sleep continuously and restoratively throughout the night often results in excessive daytime sleepiness, a key symptom of OSA [6].

Several risk factors for developing OSA have been determined; factors include habitual snoring, obesity (defined by a body mass index [BMI] > 28 kg/m^2), age ≥ 40 years, male gender, enlarged neck size, hypertension, and upper airway abnormalities [8]. Structural abnormalities can contribute to reductions in airway patency and include narrow nasal cavity, macroglossia (large tongue), retrognathia (posterior positioning of the mandible), micrognathia (undersized jaw), and excessive pharyngeal soft tissue. [2, 4, 8-11]. Other potential risk factors for OSA include smoking, alcohol consumption, post-menopausal hormonal status, and African American ethnicity [8]. However, further research is required regarding these potential risk factors, as they do not currently have sufficient evidence supporting their contribution to the development of OSA.

Numerous physiological consequences of decreased airflow and oxygen saturation affect not only the ability to sleep through the night but also day-to-day activities. Daily activities are affected by tiredness during wakeful hours, impaired cognitive function, and motor vehicle and occupational accidents [12]. OSA patients also exhibit higher prevalence of hypertension, type II diabetes, cardiovascular disease, and stroke [1, 9, 10, 13-17].

The physiological mechanisms linking OSA to other chronic diseases are complex and inadequately understood. One proposed mechanism linking OSA to cardiovascular disease (CVD) is dysfunction of the vascular endothelium [1, 18-25]. OSA-related oxygen desaturation and accompanying apnea/hypopnea events promote degenerative

remodeling of the arterial wall and may be an early sign of subclinical atherosclerosis [10, 15, 26-28]. The extent of hypertrophy damage in the carotid artery caused by remodeling appears to correlate with frequency and intensity of hypoxemia [27]. Measurement of the thickness of the intima-media layer of the carotid artery (CIMT), through ultrasound imaging, has been demonstrated to be a valid and dependable non-invasive marker of subclinical atherosclerosis [29, 30]. CIMT thickness indicates a higher level of CVD risk by indicating the presence of early atherosclerosis and wall hypertrophy [27].

A second plausible mechanism is autonomic dysregulation induced by hypoxemia. Many mechanisms are thought to contribute to autonomic dysregulation including hyperleptinemia, increased sympathetic activity, systemic inflammation, oxidative stress, and impaired baroreflex [31]. Severity of OSA measured by the frequency and intensity of hypoxemia appears to be significant in determining development of atherosclerosis independent of traditional CVD risk factors [29, 30, 32-36]. Hypoxemia causes increased sympathetic stimulation due to chemoreceptor activation; this leads to vasoconstriction and increased arterial pressure that contribute to carotid wall damage [37, 38]. OSA leads to overall autonomic dysfunction; this has been demonstrated through heart rate variability (HRV), muscle sympathetic nerve measurement and catecholamine levels in patients [37].

In the absence of coexisting OSA, the prevailing view of snoring is that there is little evidence to support a pathological role in development of vascular disease [39, 40]. However, a recent study done by Lee, et al. (2008) identified that heavy snoring in the absence of hypoxemia is an independent risk factor for carotid atherosclerosis. Vibration-

induced endothelial dysfunction in rabbits provides a plausible pathophysiological mechanism linking snoring and localized carotid atherosclerosis [21, 41]. Endothelial dysfunction plays a pivotal role in the early pathogenesis of atherosclerosis and can be detected through CIMT measurement in patients with sleep-disordered breathing [21]. Self-reported snoring has been shown to be significantly associated with increased CIMT measures, independent of traditional CVD risk factors [9]. Studies to date have used middle aged and older males as participants; the extent to which CIMT remodeling occurs in young men is unknown. Identifying snoring in young men as an early marker of subclinical atherosclerosis could have substantial public health implications for the management and early detection in the cascade of events from habitual snorer, to OSA diagnosis, to overt CVD development [10].

The purposes of this study are to first, compare CIMT measurements of young males who are habitual snorers compared to those who do not snore. Second, to compare IMT measures of vessel health in the brachial artery, not exposed to vibrations caused by snoring, to CIMT measures. Brachial IMT will serve as control so that CIMT between groups can be compared. And third, to determine the likelihood of the presence of OSA through the use of validated questionnaires.

Methods

Participants

Twenty-three (23) males from the James Madison University (JMU) and Harrisonburg, Virginia communities were recruited for this study. Subjects were non-smokers, free from any known CVD, pulmonary, or metabolic disease. The use of

medications that may influence the hemostatic parameters under investigation were cause for exclusion from the study. All subjects were classified as overweight-to-obese according to body mass index (BMI) classifications (overweight = BMI > 25 kg/m^2) [42]. Subjects were asked to report to the Human Performance Laboratory (HPL) on two separate occasions, on consecutive days, for a total of two hours (one hour each visit). All subjects were given signed consent forms to read and sign that provided a comprehensive description of the study, the risks and benefits associated with the study, and explained how confidentiality would be maintained. All procedures were approved by the JMU Institutional Review Board prior to initiation of this study.

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Body Composition

Upon completion of all the questionnaires, subjects had their body weight measured using a physician's scale and recorded to the nearest 0.1 kg, and height measured and recorded to the nearest 0.5cm. Waist and neck circumference measurements were also taken according to the American College of Sports Medicine (ACSM) guidelines using a cloth tape measure with spring-loaded handle, to reduce compression of the skin [42]. Estimates of percent body fat were then made via dual-energy x-ray absorptiometryer (DEXA) (Delphi QDR; Hologic; Boston, MA).

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Ultrasonography

Subjects were asked to return to the HPL one day after the first visit to return the Actigraph device for data download and analysis. Subjects then underwent measurements of carotid intima media thickness (CIMT) and brachial artery wall thickness via ultrasonography (DC-6; Mindray; Mahwah, NJ). For the CIMT and brachial measurements, subjects were brought to a quiet room to lie supine for ten minutes. After this time, a portable ultrasound machine and gel were used to obtain and save ultrasound images of the carotid and brachial arteries for later analysis. For the CIMT measurement, the participant was instructed to lie in a supine position with the head slightly extended and turned in the opposite direction of the carotid artery being studied. For the brachial measurement, the participant was asked to lay their arm supine on the table while images were saved. Brachial IMT served as the control measurement to compare CIMT to determine if there was an effect from the vibrations of snoring.

Statistical Analysis

All variables were compared using independent sample t-tests, to assess group differences of study variables. Pearson correlations were calculated to identify potential relationships between CIMT, brachial artery wall thickness and other study variables. Analysis of covariance (ANCOVA) was then employed to examine group differences with CIMT or brachial artery wall thickness as the dependent variable, and any significant correlation as the covariate. For example, group difference in CIMT was used as the dependent variable and the significant correlation between CIMT and PA was used as the covariate. An alpha level of $P<0.05$ was set as the *a priori* statistical significance.

Results

Twenty-three subjects were separated into two groups categorized by snorers and non-snorers. Characteristics of the two groups are presented in Table 1. The snorers and non-snorers groups did not differ in measures of age, body composition, cardiovascular health, physical activity, or sleep questionnaire results except for the STOP questionnaire. Of the snorers, ten out of seventeen (58.8%) were considered at 'high risk' for OSA according to the STOP questionnaire compared to 0% of the non-snorers were at high risk.

CIMT

CIMT measurements did not differ between groups ($p = .093$). However, a significant positive correlation was noted between CIMT and PA, as expressed by MET-min per week (Figure 2). After controlling for PA, CIMT was greater in the snorers vs. the non-snorers (Figure 3). Brachial artery wall thickness measurements did not differ between groups, and no correlation was noted with PA.

Discussion

The present study aimed to assess the association between self-reported snoring and subclinical carotid atherosclerosis in young males. The primary finding of this study is an increased CIMT in snorers compared to non-snorers after controlling for the effects of physical activity in young, male, overweight subjects without any known form of CVD. Between groups, brachial artery measurements were not different; suggesting that endothelium dysfunction of the carotid artery may be caused by proximity to vibrations due to snoring. It is important to note that a CIMT greater than the 75th percentile for

one's age is considered 'increased risk' for CVD [43]. No subject in the current study met this criterion. This is not surprising given the young age of our subjects. However, our findings may indicate self-reported snoring in young males to be an early potential marker for long-term development of OSA and CVD.

Snoring is a well-established, independent risk factor for obstructive sleep apnea, but few studies have assessed the association between self-reported snoring and risk for OSA [8]. To evaluate this association, we used two separate questionnaires, the STOP Questionnaire and Epworth Sleepiness Scale (ESS) to assess level of risk for OSA and daytime sleepiness respectively. A high percentage of the snorers were at high risk for developing OSA according to the STOP Questionnaire; in comparison, zero percent of the non-snorers were indicated to be at high risk for OSA according the STOP Questionnaire. Therefore, based on results from the STOP questionnaire, young males who snore are at a greater risk for developing OSA compared to non-snorers. However, scores between groups did not differ on the ESS questionnaire indicating the risk for OSA was not different between snorers and non-snorers based on levels of excessive daytime sleepiness. Given our young, student population, it is not surprising that both groups indicated feeling drowsy during the day. It is likely that this daytime sleepiness is reflected by a student lifestyle of voluntarily restricted sleep, rather than the presence of OSA.

Many studies have investigated endothelial dysfunction as a plausible pathological mechanism linking OSA and CVD [19-26]. However, fewer studies have looked at the association between snoring and endothelial dysfunction [10, 21, 41] and no studies to date have specifically measured CIMT in young subjects who snore. In

response to this, the present study found young male habitual snorers had significantly greater CIMT compared with non-snorers after adjusting for physical activity, suggesting endothelial dysfunction of the carotid artery. We believe this finding can be explained by a previously proposed mechanism, suggesting that energy from snoring vibrations is transmitted to the carotid artery resulting in vessel wall intima media damage remodeling [21]. Our finding that brachial measurements were not significantly different between groups supports this mechanism interpretation.

Our study is one of many to determine a significant correlation between physical activity (PA) and carotid arterial wall thickness [44]. When combined, previous research using large sample sizes (>28,000 subjects) has found an inverse relationship between self-reported PA and CIMT [45-49]. Higher PA levels earlier in life are associated with attenuated increases in CIMT measured at three and six years later [45, 49]. However, a study done by Yamada et al. [50] using a small sample size (n=149) found no relationship between CIMT and PA level assessed using a non-specific questionnaire. It is important to note that self-reported PA questionnaires provide subjective data and are therefore limited by a weak correlation with direct measures of physical fitness [51]. Questionnaires may only provide meaningful data in large cohorts and with specifically designed instruments to measure PA; this may explain the inconsistent finding regarding the correlation [44].

Several potential mechanisms responsible for changes in arterial wall thickness in response to exercise training have been proposed [44]. Systemic shear stress plays an important role in the regulation of large artery remodeling [52] and the formation of carotid atherosclerotic plaques has been linked to the presence of low mean shear rate

[53]. Exercise is associated with an increase in systemic shear stress; this perhaps explaining the exercise training induced decrease in artery wall thickness [44]. High systemic sheer stress, such as what is found during exercise, helps to prevent carotid atherosclerotic plaques from forming in the arteries. Another potential mechanism is oxidative stress; when the production of reactive oxygen species (ROS) exceeds the efficiency of antioxidant defenses it causes endothelial dysfunction that contributes to the development of atherosclerosis [54, 55]. Exercise training is associated with antioxidant effects and therefore may lower oxidative stress and is associated with decreased arterial wall IMT [44, 56].

Finally, inflammation is also a potential mechanism to explain the correlation between CIMT and PA given the importance of the inflammatory process in the development of atherosclerosis [44]. Exercise training is associated with anti-inflammatory effects; therefore it is possible that exercise training may alter arterial wall thickness [44]. A study done by Goto et al. [57] found that the effect on arterial wall thickness is dependant on exercise training intensity. Mild and moderate intensity exercise did not show increased serum levels of pro-inflammatory markers, however, high intensity exercise did demonstrate a significant increase in serum pro-inflammatory markers and free radical formation [57]. Although the study did not measure CIMT it is reasonable to speculate that high intensity exercise would contribute to increased IMT measurements. Our study found a significant, positive correlation between PA and CIMT. Of the 23 total subjects, 11 were classified as having a high level of physical activity according to the IPAQ-SF. Given the large number of subjects with a high level of

physical activity, our fining supports the inflammatory mechanism leading to increased arterial wall IMT.

This is the first study to determine an association between self-reported snoring and subclinical atherosclerosis in young males. However, a relatively small sample size was used and the extent to which these findings can be applied to females is unknown. A strength of the study was that outside factors that could affect CIMT such as PA, medication use, smoking, and other chronic diseases that could impact study variables were controlled for through strict exclusion criteria and statistical control. Future research should compare CIMT between varying levels of severity of snoring using a larger sample size to determine a stronger association. Additionally, overnight polysomnography with 12-lead EKG could be used to better determine the likelihood of OSA and detect heart rate variability patterns. Overall, the present study established self-reported snoring as an early potential sign in the cascade of events from habitual snorer, to high risk for the development of OSA, to overt cardiovascular disease development. Self-reported snoring is inexpensive and minimally time consuming thus, may be helpful in early identification of those at increased risk for the development of OSA and CVD.

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Table 1. Subject Characteristics

	Snorers (n=17)	Non-Snorers (n=6)
Age, yr.	20.2 (1.9)	20.0 (2.0)
BMI, kg/m^2	29.1 (2.9)	27.8 (2.7)
Waist Circumference, cm	93.1 (11.3)	88.4 (5.6)
Neck Circumference, cm	39.8 (1.8)	38.8 (1.1)
Total Body Fat %	20.5 (7.4)	17.2 (3.2)
Trunk Fat %	19.4 (8.9)	15.6 (3.1)
Resting HR, BPM	67.2 (7.9)	63.0 (4.6)
Systolic BP, mmHg	121.4 (6.0)	119 (5.8)
Diastolic BP, mmHg	82.6 (6.6)	80.3 (2.9)
Brachial Artery, mm	.330 (.060)	.330 (.040)
IPAQ-SF,MET-min/wk	3328.5 (3038.2)	3675.8 (2417.5)
Epworth Sleepiness Scale	10.2 (3.2)	9.5 (3.7)
Sleep Latency, min.	5.43 (5.7)	2.00 (3.9)
Sleep Efficiency, %	85.5 (12.1)	87.2 (7.7)
Total Sleep Time, min.	325.8 (69.7)	386.4 (42.0)
# of Awakenings per hour of sleep	.412 (.245)	.617 (.630)

Values are means with SD in parentheses; *n*, no. of subjects; BMI, body mass index; HR, heart rate; and BPM, beats per minute.

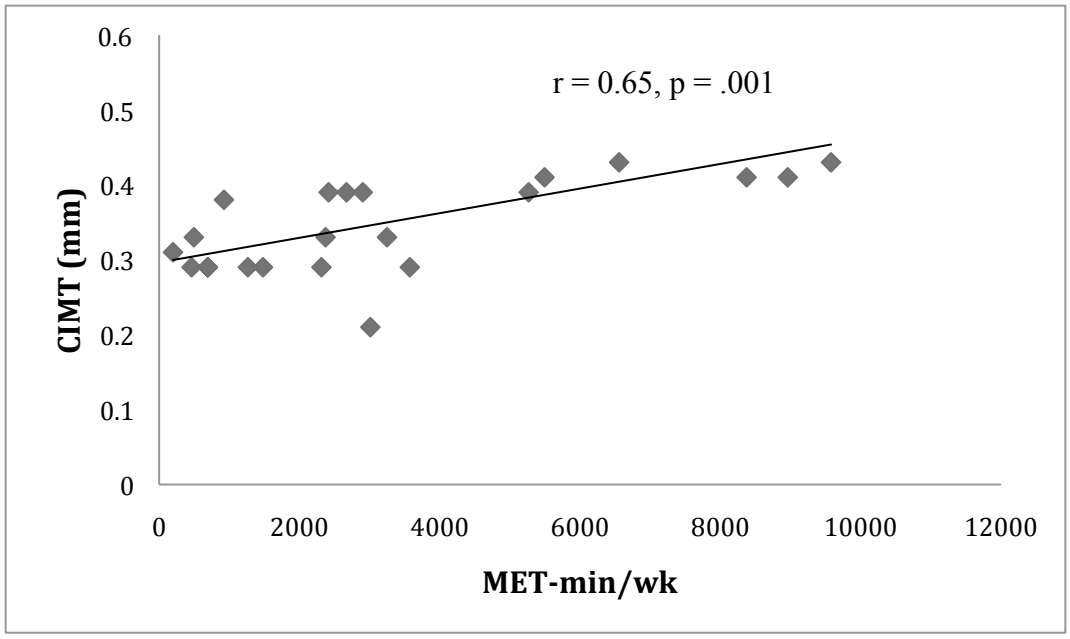

Figure 1. MET-min/wk. and CIMT Correlation. **N**ote that MET-min/wk. was significantly correlated with CIMT (*P = .001).

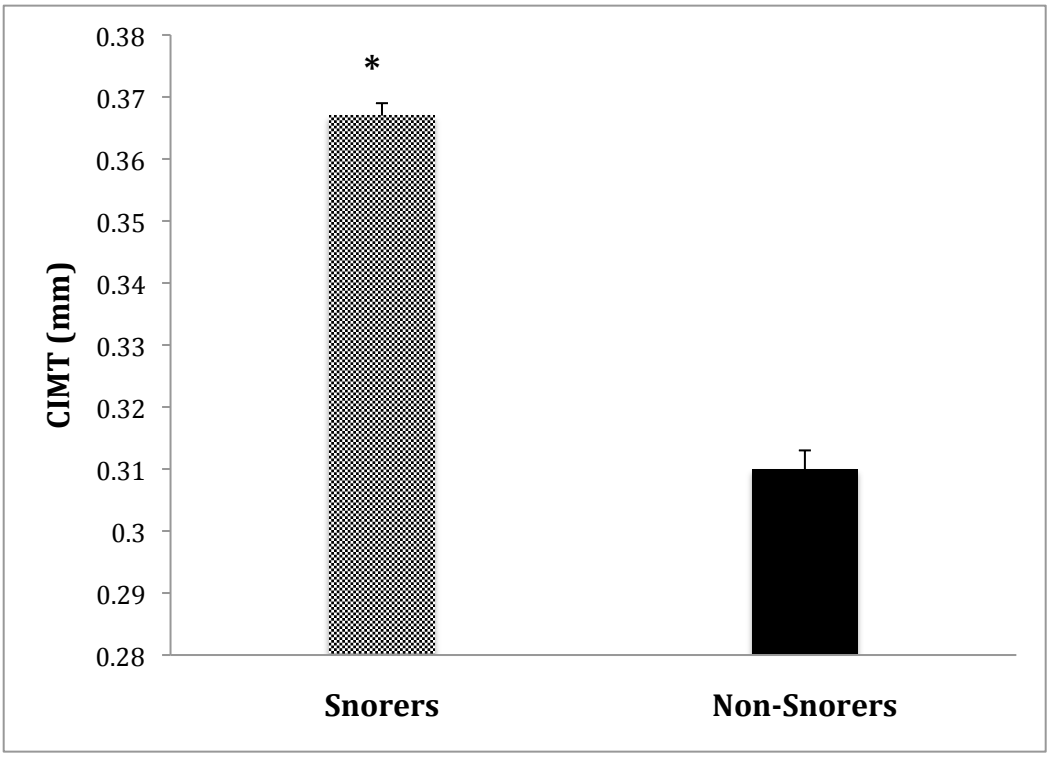

Figure 2. CIMT: carotid intima media thickness. Adjusted means after controlling for MET-min/wk, note that CIMT measures were significantly greater in snorers compared to non-snorers (*P = .014). Data represent mean values +1 SEE.

Appendix A

Berlin Questionnaire

Subject ID_____ Date Completed ____/____/____

Height (cm) _____Weight (kg) _____ Age _____

Please choose the correct response to each question.

Category 1

1. **Do You Snore?**
   - ☐ a. Yes
   - ☐ b. No
   - ☐ c. Don't know

If you snore:

2. **Your snoring is:**
   - ☐ a. Slightly louder than breathing
   - ☐ b. As loud as talking
   - ☐ c. Louder than talking
   - ☐ d. Very loud – can be heard in adjacent rooms

3. **How often do you snore?**
   - ☐ a. Nearly every day
   - ☐ b. 3-4 times a week
   - ☐ c. 1-2 times a week
   - ☐ d. 1-2 times a month
   - ☐ e. Never or nearly never

4. **Has your snoring every bothered other people?**
   - ☐ a. Yes
   - ☐ b. No
   - ☐ c. Don't Know

5. **Has anyone noticed that you quit Breathing during your sleep?**
   - ☐ a. Nearly every day
   - ☐ b. 3-4 times a week
   - ☐ c. 1-2 times a week
   - ☐ d. 1-2 times a month

☐ e. Never or nearly never

Category 2

6. **How often do you feel tired or fatigued after you sleep?**
   ☐ a. Nearly every day
   ☐ b. 3-4 times a week
   ☐ c. 1-2 times a week
   ☐ d. 1-2 times a month
   ☐ e. Never or nearly never

7. **During your waking time, do you feel tired, fatigued or not up to par?**
   ☐ a. Nearly every day
   ☐ b. 3-4 times a week
   ☐ c. 1-2 times a week
   ☐ d. 1-2 times a month
   ☐ e. Never or nearly never

8. **Have you ever nodded off or fallen asleep while driving a vehicle?**
   ☐ a. Yes
   ☐ b. No

If yes:

9. **How often does this occur?**
   ☐ a. Nearly every day
   ☐ b. 3-4 times a week
   ☐ c. 1-2 times a week
   ☐ d. 1-2 times a month
   ☐ e. Never or nearly never

Category 3

10. **Do you have high blood pressure?**
    ☐ a. Yes
    ☐ b. No
    ☐ c. Don't Know

Appendix B

Epworth Sleepiness Scale

Subject ID_____ Date Completed ____/____/____

This questionnaire asks you to indicate the chances of you becoming drowsy during hours of the day that you are not in bed sleeping. "How likely are you to doze off or fall asleep in the following situations?"

Use the following scale and indicate the most appropriate number for each situation.

0 = would never doze
1 = slight chance of dozing
2 = moderate chance of dozing
3 = high chance of dozing

	Situation	**Chance of Dozing**
1.	Sitting and reading	_____
2.	Watching T.V.	_____
3.	Sitting, inactive in a public place (ex. Theatre or meeting)	_____
4.	As a passenger in a car for an hour without a break	_____
5.	Lying down to rest in the afternoon when circumstances permit	_____
6.	Sitting and talking with someone	_____
7.	Sitting quietly after a lunch without alcohol	_____
8.	In a car, while stopped for a few minutes in the traffic	_____

Sum of Scores, items 1-8 (staff use only)

Appendix C

Health History Questionnaire

TODAY'S DATE _____

NAME _____ AGE_____ DATE OF BIRTH

_____

ADDRESS

_____

_____

Street City State

Zip

TELEPHONE: HOME/ CELL _____/ _____ E-MAIL ADDRESS _____

Person to contact in case of an emergency _____ Phone _____
(relationship) _____

Have you ever been diagnosed with cardiovascular disease? YES NO

Have you ever been diagnosed with musculoskeletal disease? YES NO

Have you ever had any heart problems? YES NO

Have you ever had Asthma? YES NO

Have you ever been told by a doctor you should not exercise? YES NO

Are you taking any Prescription (include birth control pills) or Non-Prescription medications? Yes No

For each of your current medications, provide the following information:

MEDICATION Dosage- times/ day Time taken Years on medication

Reason for Taking

_____

_____

_____

_____

_____

ACTIVITY LEVEL EVALUATION

What is your occupational activity level? Sedentary Light Moderate Heavy

Do you currently engage in vigorous physical activity on a regular basis? Yes No

If so, what type(s)? _____ How many days per week?

_____

How much time per day? <15 min 15-30 min 30-45 min >60 min

Do you engage in any recreational or leisure-time physical activities on a regular basis? Yes No

If so, what activities?

_____

On average: How often? _____ times/week; for how long? _____ time/session

Do you ever have an uncomfortable shortness of breath during exercise or when doing activities? Yes No

Do you ever have chest discomfort during exercise? Yes No

Appendix D

Informed Consent

James Madison University
Department of Kinesiology
Informed Consent

Purpose

You are being asked to volunteer for a research project conducted by Dr. Trent Hargens from James Madison University entitled, "Association of habitual snoring with carotid intima-media thickness in young men".

The primary goals of this study is to examine whether the vibrations that come from habitual snoring result in structural changes to the blood vessels in your neck that may lead to early signs of cardiovascular disease, and to relate habitual snoring to risk for obstructive sleep apnea (OSA).

Experimental Procedures

You will be asked to visit the Human Performance Laboratory (Godwin 209) 2 times, 1 – 2 days apart. Your total time commitment for participation in this study will be approximately 2 hours. In addition, you will be asked to wear a device on your wrist for 1 night, while you sleep. This will be done in your own home. Detailed information on each visit is provided below:

Visit 1

Before any test is given, you will be asked to complete a screening form and an informed consent, to insure that you meet the study criteria, that you do not have any factors that would disqualify you from participation. Upon completion of the informed consent, you will be asked to complete a short health history questionnaire providing information about your characteristics and health. You will then be asked complete 3 standardized questionnaires about snoring, the quality of your sleep, daytime sleepiness, and risk for OSA.

You will then have your height, weight, waist circumference and neck circumference measured. After that, your body composition will be analyzed via a Dual-energy x-ray absorptiometer (DEXA). The DEXA scan will allow us to measure your percent body fat and the mineral content and density of your bones. The DEXA is much like an X-ray machine. The DEXA will scan your entire body very slowly; so, you will need to lie on a table without moving for almost 10 minutes, while the DEXA is passed over your entire body. You will feel no discomfort associated with this test.

Following the DEXA scan, we will ask that you wear a heart rate monitor while lying down in a darkened room for 15 minutes to get measures of your heart rate. We will also obtain your resting blood pressure.

At the end of this first visit you will also be instructed on the proper use procedures for wearing an accelerometer to monitor your sleep for 1 night. An accelerometer is a small device that is to be worn on your wrist while in bed. You will also be asked to record the time you go to bed and the time you get up the following morning.

Visit 2

One to two days after Visit 1, you will be asked to return to Godwin 209 to return the accelerometer and to complete the remaining tests for the study. You will be asked to lie down in a quiet, darkened room for 10 minutes, after which an ultrasound probe (with gel) will be placed on your neck and arm to get images of your blood vessels. After this, a blood pressure cuff will be placed on your arm and inflated for a period of 5 minutes. After 5 minutes, the blood pressure cuff will be released we will again collect ultrasound images of the blood vessel in your arm. We will collect images for 2 minutes after we release the blood pressure cuff. If at any point during the 5 minutes that the blood pressure cuff is on your arm that you feel you need to stop the test, the researcher will immediately deflate the cuff.

Risks

There are no risks associated with wearing an accelerometer. Also, there is no risk associated with heart rate, carotid ultrasound, blood pressure, height, weight, and waist and neck circumference measures. Measurements with associated risks include: the DEXA scan and ultrasound blood pressure cuff procedure.

The amount of radiation that you will receive in the DEXA exam is less than the amount you will receive during a transatlantic flight, and is equal to about 1/20 of a chest x-ray.

The risks of the ultrasound blood pressure cuff procedure include discomfort often described as your arm is “falling asleep”; there is a temporary reduction or loss of feeling because the blood flow through the vessel is stopped for five minutes. There is no long term risk associated with this procedure.

Benefits

There is no guarantee that you will get any benefit from taking part in this study. Benefits may include knowledge about your health status. You will receive information on your body composition, including percent body fat and bone mineral density, an assessment of your sleep quality and risk for sleep apnea, and an assessment of your blood vessel health. Indirect benefits of participating in this study will be helping the researchers better understand the relationship between habitual snoring, sleep apnea, and cardiovascular disease risk.

Inquiries

If you have any questions or concerns or you would like to receive a copy of the final aggregate results of this study, please contact Dr. Trent Hargens at hargenta@jmu.edu or (540) 568-5844. In the case of any immediate concerns or adverse reactions during the study, contact Dr. Hargens on his cell phone (540) 810-1310.

Questions about Your Rights as a Research Subject
Dr. David Cockley
Chair, Institutional Review Board
James Madison University
(540) 568-2834
cocklede@jmu.edu

Confidentiality

All data and results will be kept confidential. You will be assigned an identification code. At no time will your name be identified with your individual data. The researcher retains the right to use and publish non-identifiable data. All data will be kept secured in a locked cabinet. Final aggregate results will be made available to participants upon request.

Freedom of Consent

Your participation is entirely voluntary. You are free to choose not to participate. Should you choose to participate, you can withdraw at any time without consequences of any kind.

I have read this consent form and I understand what is being requested of me as a participant in this study. I freely consent to participate. I have been given satisfactory answers to my questions. The investigator provided me with a copy of this form. I certify that I am at least 18 years of age.

_____ _____
Name of Subject (Printed) Name of Researcher (Printed)

_____ _____
Name of Subject (Signed) Name of Researcher (Signed)

_____ _____
Date Date

Appendix E

International Physical Activity Questionnaire – Short Form (IPAQ-SF)

INTERNATIONAL PHYSICAL ACTIVITY QUESTIONNAIRE

We are interested in finding out about the kinds of physical activities that people do as part of their everyday lives. The questions will ask you about the time you spent being physically active in the **last 7 days**. Please answer each question even if you do not consider yourself to be an active person. Please think about the activities you do at work, as part of your house and yard work, to get from place to place, and in your spare time for recreation, exercise or sport.

Think about all the **vigorous** activities that you did in the **last 7 days**. **Vigorous** physical activities refer to activities that take hard physical effort and make you breathe much harder than normal. Think *only* about those physical activities that you did for at least 10 minutes at a time.

1. During the **last 7 days**, on how many days did you do **vigorous** physical activities like heavy lifting, digging, aerobics, or fast bicycling?

   _____ **days per week**

   ☐ No vigorous physical activities → ***Skip to question 3***

2. How much time did you usually spend doing **vigorous** physical activities on one of those days?

   _____ **hours per day**

   _____ **minutes per day**

   ☐ Don't know/Not sure

Think about all the **moderate** activities that you did in the **last 7 days**. **Moderate** activities refer to activities that take moderate physical effort and make you breathe somewhat harder than normal. Think only about those physical activities that you did for at least 10 minutes at a time.

3. During the **last 7 days**, on how many days did you do **moderate** physical activities like carrying light loads, bicycling at a regular pace, or doubles tennis? Do not include walking.

   _____ **days per week**

   ☐ No moderate physical activities → ***Skip to question 5***

4. How much time did you usually spend doing **moderate** physical activities on one of those days?

_____ **hours per day**

_____ **minutes per day**

☐ Don't know/Not sure

Think about the time you spent **walking** in the **last 7 days**. This includes at work and at home, walking to travel from place to place, and any other walking that you have done solely for recreation, sport, exercise, or leisure.

5. During the **last 7 days**, on how many days did you **walk** for at least 10 minutes at a time?

_____ **days per week**

☐ No walking → ***Skip to question 7***

6. How much time did you usually spend **walking** on one of those days?

_____ **hours per day**

_____ **minutes per day**

☐ Don't know/Not sure

The last question is about the time you spent **sitting** on weekdays during the **last 7 days**. Include time spent at work, at home, while doing course work and during leisure time. This may include time spent sitting at a desk, visiting friends, reading, or sitting or lying down to watch television.

7. During the **last 7 days**, how much time did you spend **sitting** on a **week day**?

_____ **hours per day**

_____ **minutes per day**

☐ Don't know/Not sure

This is the end of the questionnaire, thank you for participating.

Appendix F

STOP Questionnaire

Subject ID_____ Date Completed ____/____/____

Height (Inches) _____ Weight (lbs) _____ Age _____

Neck Circumference (cm) _____

The **STOP** Test consists of four questions:

1. **Snoring**
   Do you *snore* loudly (louder than talking or loud enough to be heard through closed door)? **Yes No**

2. **Tired**
   Do you often feel *tired*, fatigued or sleepy during the day? **Yes No**

3. **Observed**
   Has anyone *observed* you stop breathing during your sleep? **Yes No**

4. **Blood Pressure**
   Do you have or are you being treated for high blood *pressure*? **Yes No**

High risk of OSA: answering yes to *two or more* questions
Low risk of OSA: answering yes to *less than two* questions

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