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ORIGINAL ARTICLE

Cardiometabolic Risk in Adolescents: Associations with Physical Activity, Fitness, and Sleep

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Abstract
Background Physical activity and fitness are independently associated with cardiometabolic dysfunction, and short sleep duration is an emerging marker of obesity. Few have examined interrelations among these factors in a comprehensive risk model.
Purpose Investigate the influence of behavioral and lifestyle risk factors on the metabolic syndrome and inflammation.
Methods A sample of 367 15–17-year-olds (73 % boys) from ethnic minority groups (45.8 % Hispanic, 30.8 % Black), most with elevated blood pressure (72 %), underwent aerobic fitness testing, blood sampling, and completed behavioral questionnaires.
Results Structural model results are consistent with the notion that short sleep duration, poor sleep quality and fatigue, and decreased physical activity are associated with increased risk of metabolic syndrome and inflammation possibly via effects on reduced cardiorespiratory fitness.
Conclusions The combination of negative lifestyle and behavioral factors of physical inactivity, sleep loss, and poor fitness has serious implications for cardiovascular health complications in at-risk youth.

Keywords Cardiorespiratory fitness · Behavioral factors · Metabolic syndrome · Inflammation · Pediatrics

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Introduction

Increased prevalence of obesity, elevated blood pressure (BP), insulin resistance, and inflammation in adolescents has stimulated investigation into how these factors contribute to cardiovascular morbidity and mortality. Although the causes are multifactorial, sedentary behavior and low levels of physical activity contribute to the development of the metabolic syndrome and inflammatory states [1, 2]. Limited work has evaluated the relative contribution of sleep duration as an intermediate risk factor of cardiovascular disease; however, emerging evidence points to parallel trends between increased risk factor prevalence and decline in sleep duration as a potential explanation for how other lifestyle and behavioral factors may converge to influence cardiometabolic disease [3]. A 2006 U.S. National Sleep Foundation Survey found that 62 % of ninth to twelfth graders get an insufficient amount of sleep on school nights (<8 h). More than half of those adolescents report less sleep than they feel they need and are likely to report feeling too tired to engage in exercise and other physical activities [4], thus compounding the physiologic consequences of sleep loss. Given that risk factors often emerge during adolescence as a result of initiating health-compromising behaviors, and both behavioral patterns and their physiologic consequences are thought to extend later into life [5], it is important to examine in youth the relative influence of multiple cardiometabolic risk factors, including sleep.

In adults, persistent low levels of fitness are associated with increases in the metabolic syndrome and specific inflammatory markers such as IL-6, C-reactive protein (CRP), and fibrinogen [2, 6]. Despite evidence that fitness patterns from adolescence persist during adulthood [7], relationships between aerobic fitness and metabolic syndrome and inflammatory outcomes are not well established in youth [1, 2]. It has been proposed that physical activity may, over time, improve

aerobic fitness levels and subsequently decrease cardiometabolic risk. Cugnetto and colleagues [8] found that being physically active was associated with greater aerobic fitness, which in turn was related to lower body mass index (BMI). However, self-reported physical activity is only weakly associated with metabolic syndrome component clusters [9, 10] and individual inflammatory markers [2] when examined in conjunction with more objective fitness measures (e.g., maximal exercise tests). It is unclear if this weak association can be attributed to (1) inconsistencies or error in self-report, (2) the absence of tests for integrated relationships between physical activity and fitness (i.e., meditational pathways), or (3) complexities and variability in conceptualizing outcomes of the metabolic syndrome and inflammation. Additionally, lack of sleep as a factor in these models prevents conclusive interpretation of risk factor processes.

Mounting epidemiologic and observational evidence in youth highlights the deleterious impact of shortened sleep duration on individual metabolic syndrome risk factors, particularly obesity [11, 12]. Two studies in adults provide preliminary support for a "U-shaped" relationship between sleep duration and metabolic syndrome [13, 14]. Though extended sleep is thought to contribute to metabolic syndrome risk, evidence of a stronger effect for short sleep (i.e., <8 h) even after controlling for sociodemographic variables and physical activity suggests that curtailed sleep may have salient and unique cardiometabolic consequences. The effects of sleep loss on cardiovascular health are likely the result of physiologic changes involving increased sympathetic nervous system activity, decreased glucose tolerance, increased inflammation, and changes in appetite regulation and energy expenditure [15]. It is thought that insufficient sleep duration contributes to the development of an inflammatory state [16]. Laboratory sleep restriction results in a temporary increase in IL-6 and CRP in adults [17, 18]; however, there is some question as to whether this effect translates to recurrent sleep loss, given that only a handful of studies directly examined this hypothesis (using CRP), ultimately yielding mixed results [19, 20]. Additionally, total energy expenditure, a measure of physical activity, is conceptually proposed as a behavioral and biological mechanism linking sleep loss and cardiovascular risk [21], but it is unknown how fitness and physical activity are related to sleep duration and if they are associated with the metabolic syndrome and inflammation in a comprehensive model.

The purpose of the current study was to test a structural model with direct association of physical activity, fitness, and sleep with the metabolic syndrome and inflammation, as well as an indirect association of physical activity with outcomes through fitness, covarying for gender and parent education. A secondary aim was to conceptualize the metabolic syndrome and inflammation as latent constructs. Specifically, we planned to determine if inflammatory markers of IL-6, CRP, and fibrinogen load onto a latent inflammation construct. We also sought to replicate the Shen and colleagues [22] adult metabolic syndrome measurement model (comprised of latent variables of obesity, insulin resistance, lipid levels, and BP) in our sample of adolescents, most of them at risk for cardiovascular disease.

Methods

Participants

The participants for the current study were 367 (268 boys; 99 girls) adolescents, ages 15–17 years (M=16.1, SD=0.7). The majority of the sample was from self-identified racial or ethnic minority groups (45.8 % Hispanic, 30.8 % Black). More than half of the participants reported a positive parental history of hypertension, and parents completed an average of some college education (M=13.3, SD=2.5). Individuals had relatively short hours of sleep duration for adolescents (M=7.7, SD=1.2). Results from laboratory studies indicate that sleep need, defined as the amount of sleep achieved during a 10 h nocturnal period, reaches approximately 9 h in adolescents [23].

Participants were drawn from three data cohorts and were part of larger studies examining cardiovascular risk in adolescents. Prospective participants for the study were identified during an annual high school BP screening from 2000–2005. Individuals in the first participant cohort were recruited based on criteria of persistently elevated BP (systolic [S]BP and/or diastolic [D]BP≥95th percentile, adjusted for their age, gender, and height) measured during two separate school screenings, and SBP and/or DBP≥90th percentile during a home BP assessment (n=61). Normotensive adolescents were identified through friend recruitment during home visit (n=104). Participants in cohorts 2 and 3 were recruited based on criteria of persistently elevated BP during two screenings at school and home assessment (SBP and/or DBP≥90th percentile). Additional study recruitment details have been outlined previously [8]. Though participants in cohorts 2 and 3 were part of a larger intervention study, only pretreatment or baseline assessment variables were used in these analyses.

Inclusion criteria consisted of age <18 years, US residency for at least 4 years, BP<160/100 mmHg, not taking any medication with potential cardiovascular effects (including oral contraceptives), and being otherwise healthy (e.g., no history of asthma, heart murmurs, diabetes, severe allergies, renal disease, secondary hypertension, seizure disorder, developmental disabilities, cancer, bronchial conditions, ventricular arrhythmias, heart disease, amputation or related birth defect, major psychological disorder, spinal cord injury, or arthritis). Participants with an abnormal electrocardiogram or echocardiogram

at baseline assessment were excluded from the larger study. Participants were not assessed for presence or absence of sleep disorders. A total of 809 students were screened, and 464 were eligible. Of the eligible students, 383 consented to participate, and 367 completed the baseline assessment. A total of 16 withdrew from the study prior to baseline. The present study, therefore, included 367 students with both elevated and normal BP. Included subjects did not differ from those not included on all major demographic variables (p values>0.05).

Procedures

Written informed consent and assent for the study procedures were obtained from participants and their parents at a home visit during which eligibility for the study was determined. Casual BP and sociodemographic variables (e.g., age, gender, parent education, and parental history of hypertension) were assessed. Participants with elevated BP during the home visit were asked to participate in the study. The nature of the assessment study was described to participants, along with a review of eligibility requirements, benefits of the study, and time commitment. Recruitment of a friend to serve as the normotensive control was also determined for participants in the first cohort. The first physical assessment appointment was made, and participants were instructed to fast after midnight (not to eat or drink anything) before the appointment. Trained personnel working in the laboratory conducted all physical assessments.

During the first medical laboratory assessment, BP, body size, lipid, glucose, insulin, and inflammation and procoagulation measures were assessed. Participants also completed a 7-day activity recall, as well as psychosocial questionnaires (e.g., Children's Depression Inventory). The second physical assessment visit included a laboratory fitness assessment. For details regarding the specific variables sampled for each participant cohort, see Table 1.

Physical Assessments

Casual BP and Body Size Measures

Blood pressure was assessed using a mercury sphygmomanometer (Baumanometer) and a cuff on the right arm. Participants were instructed to sit quietly for 5 min with their right arm rested at heart level, feet flat on the floor. Three BP readings were taken at 5, 7, and 9 min, and the cuff was removed. The average of 7 and 9 min was used to determine metabolic syndrome BP criteria. Height (inches) and weight (pounds) were assessed with a balance beam scale and height rod, and were used to calculate BMI (kilograms per square meter). Waist circumference in inches was calculated from the average of two waist measurements taken at the level of the navel.

Fasting Blood Measures

Blood collection tubes were labeled with appropriate participant information, date, and time of collection, prior to phlebotomy. Chemistry assays were performed by autoanalyzer (Roche Cobas-Mira or Cobas-Mira Plus) using commercially available kits and according to manufacturer instructions for instrument set-up, run procedures, and maintenance policies [24]. Samples were processed on the same schedule as other samples already received (stored in appropriate-sized cryogenic vials at −80 degrees until analysis) and reported usually within 48 h after collection.

While participants were supine, a catheter was inserted in their arm, and fasting blood samples were collected for serum cholesterol, triglycerides, lipoprotein determinations, glucose, and insulin. Triglyceride level was measured in EDTA plasma after hydrolysis by lipoprotein lipase by assay of released glycerol and fatty acids. High-density lipoprotein-cholesterol (HDL-C) was measured in dextran sulfate (50,000 MW), and magnesium precipitate low-density lipoproteins (LDL) and very-low-density lipoproteins in plasma or serum [25]. HDL-C remained in the supernatant after centrifugation and was assayed using cholesterol enzymatic reagent. Inter-assay CVs are 5.3 % and 2.5 % for triglycerides and HDL-C, respectively. Glucose was measured in plasma by the hexokinase technique that converts glucose to glucose-6-phosphate (G-6-P). G-6-P is oxidized by G-6-P dehydrogenase to yield NAD, which was measured spectrophotometrically at 340 nm. Intra-assay and interassay CVs are <2 % and 2.7 %, respectively. Insulin was analyzed using a solid-phase ^{125}I radioimmunoassay (Coat-A-Count, Diagnostic Products Corp), which utilizes a tube-immobilized anti-human insulin antibody with 3 h incubation after addition of cold antigen and ^{125}I-insulin label in buffer, followed by decanting of the supernatant, and gamma counting. Intra-assay and interassay CVs are 3.1–9.3 % and 4.9–10.0 %, depending on antigen concentration. The catheter was removed, and a bandage was placed on the participant's arm.

Inflammation and Procoagulation

An indwelling catheter was inserted in the participants' arm while lying down, and fasting blood samples were collected and analyzed for fibrinogen, high-sensitivity CRP, and IL-6. Fibrinogen and high-sensitivity CRP were measured on the Behring BN-100 auto-analyzer nephelometer using the manufacturer's reagents, quality controls, and methods. Tests used a serum standard that is diluted to provide a reference range, in order to avoid matrix effects in this matrix-sensitive methodology. Fibrinogen was measured in citrated plasma, and CRP was measured in serum. Intra-assay CVs are 2.7 % and <4.4 % for fibrinogen and CRP,

Table 1 Baseline characteristics: mean (SD)

	Participant groups			
Variable	Cohort 1 (n=166)	Cohort 2 (n=38)	Cohort 3 (n=163)	Total (n=367)
Age (years)	16.1 (0.8)	16.3 (0.6)	16.1 (0.6)	16.1 (0.7)
BMI (kg/m^2)	28.2 (7.5)	31.0 (7.0)	29.6 (7.7)	29.1 (7.6)
SBP (mmHg)	118.7 (11.7)	130.8 (10.8)	124.5 (11.7)	122.5 (12.2)
DBP (mmHg)	71.6 (9.8)	77.6 (9.4)	73.9 (11.4)	73.2 (10.6)
CDI	–	–	6.2 (4.5)	–
CRP (nmol/L)	–	–	17.1 (28.6)	–
Fibrinogen (μmol/L)	–	–	8.8 (3.2)	–
Glucose (mmol/L)	4.9 (0.4)	4.5 (0.5)	4.4 (0.4)	4.6 (0.5)
HDL-C (mmol/L)	1.1 (0.2)	1.1 (0.2)	1.2 (0.2)	1.1 (0.2)
IL-6 (IU/mL)	–	–	0.2 (0.2)	–
Insulin (pmol/L)	84.0 (108.3)	94.5 (62.5)	95.8 (81.3)	90.3 (93.8)
Parent education (years)	13.3 (2.4)	13.5 (2.5)	13.2 (2.7)	13.3 (2.5)
Peak VO_2 (mL/kg/min)	36.5 (11.6)	35.4 (9.4)	36.1 (9.3)	36.2 (10.3)
Physical activity[a]	33.5 (7.0)	30.9 (6.3)	29.5 (6.1)	31.4 (6.8)
Sleep duration (min)	454.9 (60.8)	445.1 (72.4)	468.0 (76.2)	459.7 (69.5)
Triglycerides (mmol/L)	1.0 (0.6)	1.1 (0.8)	1.0 (0.8)	1.0 (0.6)
Waist circumference (cm)	90.9 (18.0)	94.5 (19.1)	94.5 (19.1)	93.0 (18.5)

Cohort 1 consists of individuals with persistently elevated BP (n=59) and normotensive controls, and cohorts 2 and 3 consist of individuals with elevated BP. Cohorts 1 and 2 were sampled for metabolic syndrome and all predictor variables. Cohort 3 was sampled for metabolic syndrome and all predictor variables, as well as inflammatory markers

BMI body mass index, *SBP* systolic blood pressure, *DBP* diastolic blood pressure, *CDI* Children's Depression Inventory, *CRP* C-reactive protein, *HDL-C* high density lipoprotein cholesterol, *IL-6* interleukin-6

[a] Derived from calculation of kilocalories per kilogram per day adjusted for the removal of energy expenditure associated with sleep

respectively; interassay CVs are 2.6 % and <5.7 %, for fibrinogen and CRP, respectively. IL-6 was measured by enzyme immunoassay manufactured by R&D Systems (Quantikine, Minneapolis, MN), while intra-assay CV is <7.6 % and interassay is <9.8 %.

Seven-Day Physical Activity Recall

A research assistant administered the activity recall. The participant reported approximate bed times and wake-up times for the past 7 days, gave a detailed description of daily activities (e.g., walking, cleaning house, or playing outside), and reported activity duration and intensity (light, moderate, hard, or very hard). Activity recalls were scored to estimate sleep time in minutes per night (sleep duration) and estimate kilocalories per kilogram per day (kcal/kg/day; physical activity energy expenditure), based on procedures by Sallis and colleagues [26]. Because sleep time is included in the calculation of kilocalories per kilogram per day, a modified estimate obtained by removing sleep time from the kilocalories-per-kilogram-per-day score was used in all analyses. The activity recall is valid and reliable (r=0.81 for 11th graders) in adolescents [27]. Scores obtained for the physical activity recall had a Cronbach's alpha of 0.72.

Aerobic Fitness

A maximal treadmill test was completed using a modified Balke (walk–jog) exercise protocol to determine peak VO_2 (volume of oxygen; milliliters per kilogram per minute). Speed was set at 4.5 mph (2.01 m/s) and remained constant, while initial grade was set at 2.5 % and increased by 2.5 % at the beginning of each 2-min stage. At the end of each 2-min stage, participants were asked to rate their level of physical exertion based on the Borg rate of perceived exertion scale. During the test, expired gases were continuously collected using a low-resistance mass flow sensor and analyzed with a Sensor Medics Vmax229 metabolic cart. Exercise termination criteria were defined as three or more of the following: (1) a Borg PRE >17; (2) a respiratory exchange ratio >1.1; (3) a maximal HR (within 1 standard deviation of predicted values); (4) an increase in workload accompanied by an increase in absolute VO_2 less than 0.11.

Children's Depression Inventory

The Children's Depression Inventory (CDI) is a 27-item, self-report, symptom-focused scale for youth age 7 to 17 years and is considered a downward extension of the

Beck Depression Inventory for adults. The CDI is both valid and reliable (r=0.84) in children and adolescents [28]. Items 16 and 17 were summed to create a composite score based on self-reported sleep quality and fatigue, which was included in analyses. Composite scores obtained using the CDI had a Cronbach's alpha of 0.78.

Statistical Analyses

Descriptive statistics using Statistical Analysis Systems version 9.2 were performed for demographic (age and parent education), physiological (BP, body size, lipids, fasting insulin and glucose, inflammation), physical activity (kilocalories per kilogram per day minus sleep time), physical fitness (peak VO_2), and sleep (average of seven nights of sleep duration) variables and are reported in Table 1. Normality assumptions were assessed by identifying variables with outliers, and kurtosis>10 and skewness≥2. Sleep duration had 13 outliers, whose sleep data were excluded because they did not seem plausible (e.g., >1,000 min). Insulin, CRP, IL-6, and fibrinogen values were naturally log-transformed to improve normality.

All model tests were conducted using Mplus version 5.1 [29]. Confirmatory factor analysis (CFA) was used to examine a model with four first-order metabolic syndrome component factors (and their respective indicators) comprised of obesity (waist circumference and BMI), insulin resistance (fasting glucose and insulin), lipid level (HDL-C and triglycerides), and BP (SBP and DBP) based on the adult model from Shen and colleagues [22]. Next, the model was tested for an overarching common metabolic syndrome factor that incorporates the four lower-order component factors. A three-factor measurement model was then tested for (1) sleep: Seven indicators of sleep duration (minutes of sleep for each night during the week) and one indicator of sleep quality and fatigue (composite score of CDI items 16 and 17) load onto latent sleep duration factor; (2) inflammation: Inflammatory markers of IL-6, CRP, and fibrinogen load onto the latent inflammation variable; and (3) metabolic syndrome: Four components load onto a higher-order latent metabolic syndrome factor. Finally, structural regression was used to test for direct effects of physical activity, physical fitness, and sleep on the latent factors of metabolic syndrome and inflammation, as well as an indirect effect of physical activity and sleep on metabolic syndrome and inflammation outcomes through physical fitness.

Full information maximum likelihood (FIML) estimation was used to account for data that are assumed to be missing at random [30]. Although inflammatory markers were assessed only for cohort 3 (45 % missing data across all subjects), which may indicate the presence of nonrandom missing data, inflammatory variables were correlated with metabolic syndrome outcome measures, and thus all missing data (including inflammatory markers in cohorts 1 and 2) were handled with FIML. Given the sensitivity of the chi-square (χ^2) fit index to large sample sizes, the comparative fit index (CFI), root mean square error of approximation (RMSEA), and standardized root mean square residual (SRMR) were used to evaluate the various models [31].

Results

Measurement Model

Results of the metabolic syndrome CFA suggested adequate fit to the data (CFI=0.98; RMSEA=0.05 [90 % CI=0.029–0.047]; SRMR=0.03), supporting the hypothesized metabolic syndrome measurement model. According to the results, all factor loadings were significant (ps<0.01). The three-factor measurement model also yielded adequate fit to the data (CFI=0.97; RMSEA=0.03 [90 % CI=0.018–0.041]; SRMR=0.05). All freely estimated factor loadings were statistically significant (ps<0.05). Standardized regression coefficients for the observed indicators of the three latent variables are displayed in Fig. 1. The zero-order correlation matrix revealed several small correlations that comprised the foundation for factor analyses. Additionally, several standardized factor loadings were below 0.5 (e.g., indicators of sleep quality and fatigue, fibrinogen, and glucose). Although these variables did not load as strongly as nights of sleep duration, other inflammatory markers, and insulin, respectively, they were retained as indicators due to their conceptual and statistical significance (ps<0.01). More specifically, there is theoretical support for relationships among sleep quality, fatigue, and restricted sleep [32], as well as for affiliated biological pathways among fibrinogen and other hemostatic risk markers in the prediction of cardiovascular disease [33]. The metabolic syndrome factor was strongly correlated with the inflammation factor (r=0.8; p<0.01), and there was a trend for correlation with the sleep factor (r=−0.12; p=0.07). The sleep and inflammation factors were not correlated (r=0.01; p=0.94). The model explained 54 % of the variance in CRP, 34 % of the variance in IL-6, 6 % of the variance in fibrinogen, 11 to 50 % of the variance in nights of sleep, and 5 % of the variance in sleep quality and fatigue.

Structural Model

The final structural model is displayed in Fig. 2 and contains standardized and unstandardized coefficients. With respect to links from physical activity to endogenous variables, physical activity was associated with aerobic fitness (unstandardized coefficient=0.4, z=4.45, p<0.01), controlling for sleep. Additionally, consistent with our hypotheses, aerobic fitness

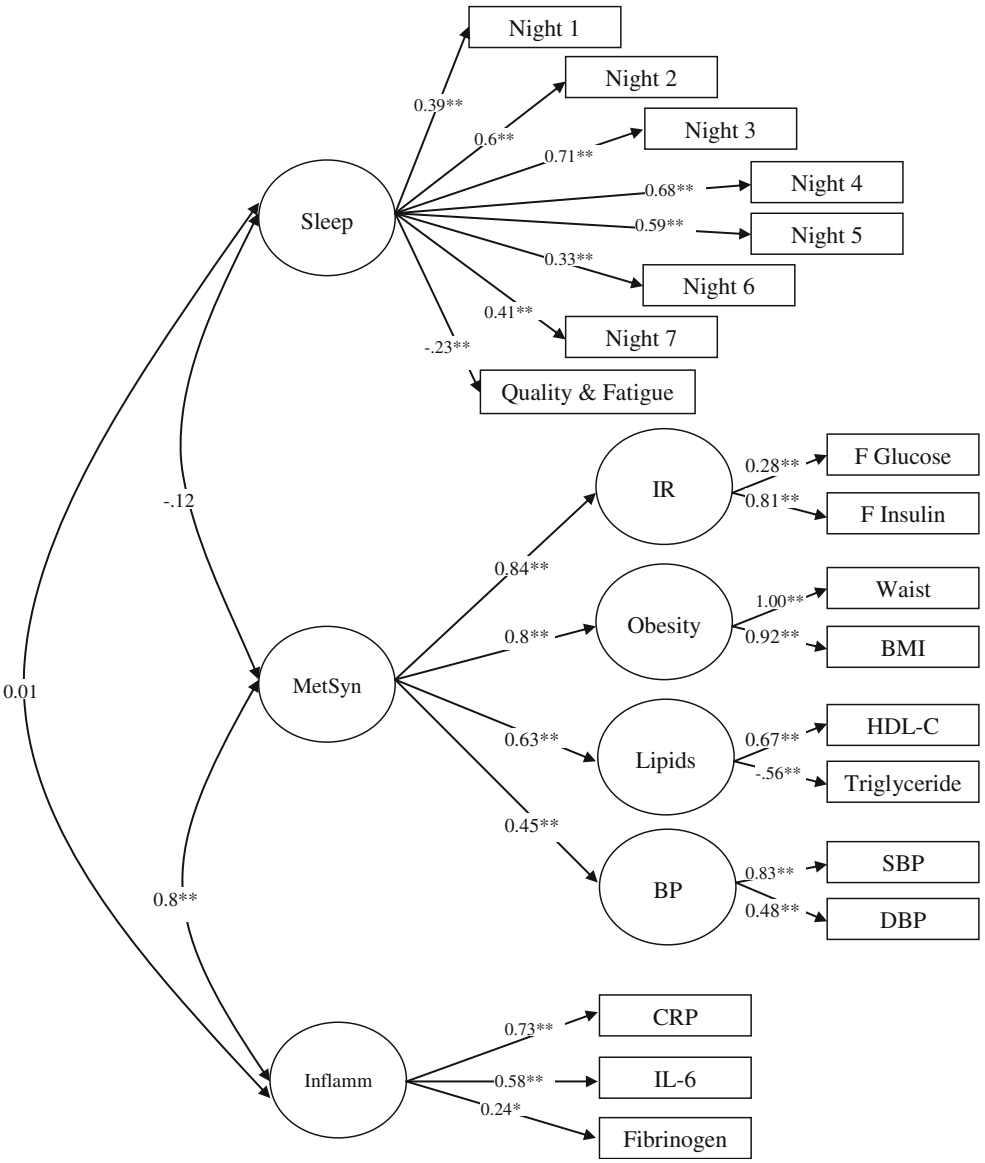

Fig. 1 Measurement model with sleep, inflammation, and metabolic syndrome latent variables. All values represent standardized coefficients. Error variances not pictured. *BMI*=body mass index; *BP*=blood pressure; *SBP*=systolic blood pressure; *DBP*=diastolic blood pressure; *CRP*=C-reactive protein; *F glucose*=fasting glucose; *F insulin*=fasting insulin; *HDL-C*=high-density lipoprotein cholesterol; *IL-6*=interleukin-6; *Inflamm*=inflammation; *IR*=insulin resistance; *MetSyn*=metabolic syndrome; *Waist*=waist circumference. *$p<0.05$, **$p<0.01$

(peak VO_2) was associated with the metabolic syndrome and (coefficient=−0.4, $z=-11.59$, $p<0.01$) inflammation factors (coefficient=−0.07, $z=-5.95$, $p<0.01$), controlling for the sleep latent variable. Results also indicated significant potential indirect associations (product of two direct path coefficients) of physical activity on the metabolic syndrome (coefficient=−0.16, $z=-4.14$, $p<0.01$) and inflammation (coefficient=−0.03, $z=-3.53$, $p<0.01$) via aerobic fitness. Interestingly, the combination of direct and indirect associations between physical activity and outcome variables as a function of aerobic fitness did not yield significant total effects (metabolic syndrome total effect, coefficient=−0.06, $z=-1.0$, $p=0.32$; inflammation total effect, coefficient=0.01, $z=0.3$, $p=0.76$).

As part of an ancillary analysis, we found an indirect association of sleep on the metabolic syndrome (coefficient=−1.0, $z=-2.6$, $p=0.01$) and inflammation (coefficient=−0.17, $z=-2.39$, $p=0.02$) involving aerobic fitness. The sleep latent variable was positively associated with aerobic fitness (coefficient=2.52, $z=2.67$, $p=0.01$), controlling for physical activity. Similar to above, there were no significant overall or total associations between sleep and the metabolic syndrome (coefficient=−1.18, $z=-1.89$, $p=0.06$) and inflammation (coefficient=−0.01, $z=-0.09$, $p=0.93$). Contrary to our hypotheses, the associations between physical activity and the metabolic syndrome and inflammation, as well as between the latent construct of sleep and the metabolic syndrome and inflammation, did not reach statistical significance. These results are consistent with the notion that the intermediary aerobic fitness factor substantially accounted for covariations between physical activity and the metabolic syndrome and inflammation, as well as between sleep and cardiometabolic outcomes.

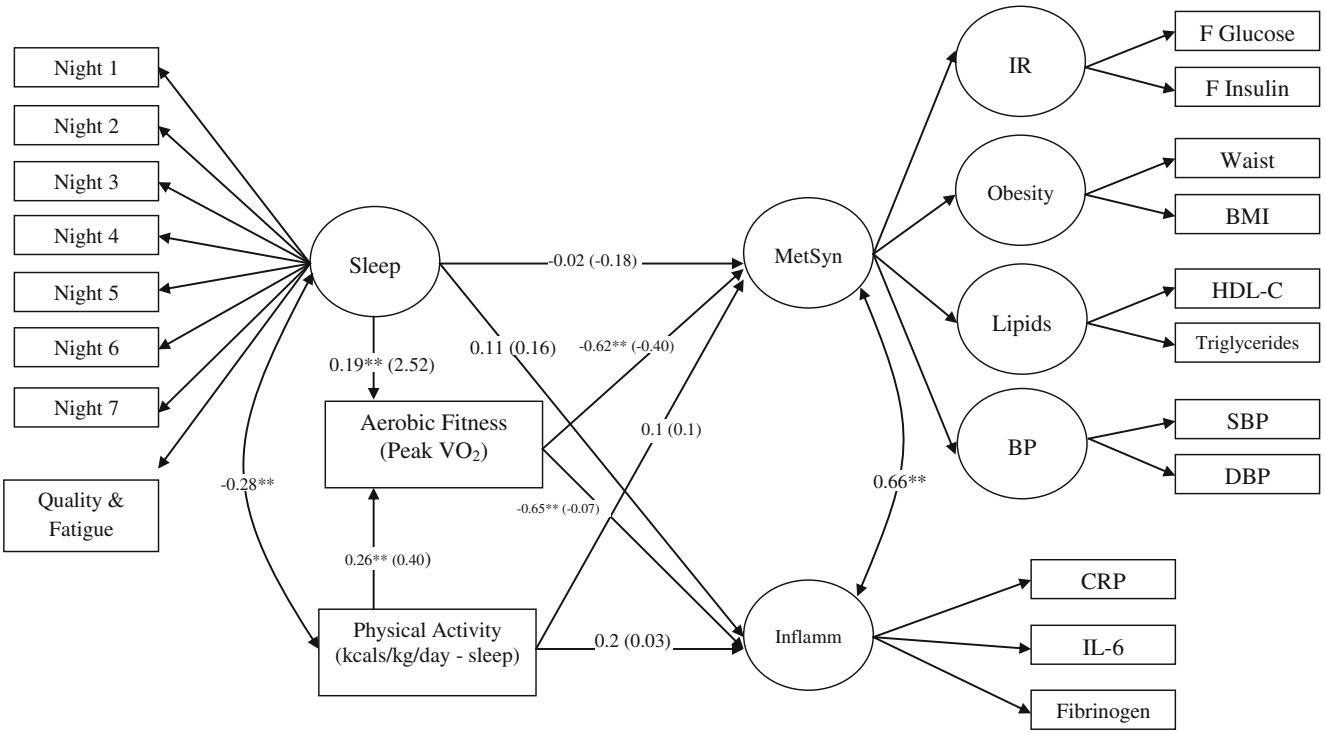

Fig. 2 Structural equation model for the prediction of metabolic syndrome and inflammation from sleep and physical activity, mediated by aerobic fitness. Paths are represented with standardized coefficients (unstandardized in *parentheses*). Latent variable factor values (not pictured) are significant at $p<0.05$. Error variances not pictured. *BMI*=body mass index; *BP*=blood pressure; *SBP*=systolic blood pressure; *DBP*=diastolic blood pressure; *CRP*=C-reactive protein; *F glucose*=fasting glucose; *F insulin*=fasting insulin; *HDL-C*=high-density lipoprotein cholesterol; *IL-6*=interleukin-6; *Inflamm*=inflammation; *IR*=insulin resistance; *MetSyn*=metabolic syndrome; *Waist*=waist circumference. *$p<0.05$, **$p<0.01$

Overall, physical activity and sleep were significantly correlated ($r=-0.28$; $p<0.01$), and there was a significant partial correlation for metabolic syndrome and inflammation ($p<0.01$). The final model explained 8 % of the variance in aerobic fitness, 38 % of the variance in the metabolic syndrome, and 39 % of the variance in inflammation.

Inclusion of Covariates

We controlled for the potential influence of gender and parent education on the metabolic syndrome by specifying those direct paths, as well as covariances between control variables and all permitted observed variables. Overall, direct and indirect effects did not significantly change, with the exception of the correlation between the metabolic syndrome and inflammation latent variables, which was attenuated and no longer significant ($p>0.05$).

Discussion

This study sought to determine how behavioral and lifestyle risk factors of physical activity, fitness, and sleep relate to the metabolic syndrome and inflammation. Though physical activity and fitness are relatively well-established risk factors, the literature is unclear concerning whether sleep duration is associated with physical activity and fitness and if these parameters act together to influence the metabolic syndrome and inflammation in adolescents. We addressed inconsistencies and limitations in the literature by examining a comprehensive risk factor model and by conceptualizing outcomes using CFA. Key findings in the current study support our hypotheses of a potentially direct association of aerobic fitness, as well as indirect association of physical activity via fitness, with the metabolic syndrome and inflammation. Results also showed that sleep was associated with the metabolic syndrome and inflammation via cardiorespiratory fitness. However, an important caveat is that the cross-sectional nature of the data does not allow for statements about mediation because the design does not permit established directionality; rather, directionality can only be assumed based on previous literature.

Relation of Physical Activity and Fitness with Cardiometabolic Variables

The association between poor aerobic fitness and increased risk of the metabolic syndrome and inflammation in our

model provides further evidence that objective measures of cardiorespiratory fitness are inversely associated with metabolic syndrome risk factor clusters and inflammatory markers in youth [1, 2]. In terms of conceptualizing outcomes, our hierarchical four-factor model was similar in structure, as well as in specific factor loading values, to the adult models depicted by Shen and colleagues [22]. It appears that insulin resistance, obesity, and unfavorable lipid values are in fact essential features of the metabolic syndrome in adolescents, which is consistent with current adult definitional guidelines (e.g., the American Heart Association/National Heart, Lung, and Blood Institute [AHA; 34]). Similarly, though fibrinogen was not significantly correlated with fitness (a finding mirrored in other investigations [35]), fibrinogen, CRP, and IL-6 cluster together and are predicted by fitness level.

We also provide limited support for a potential mediation effect of fitness, since physical activity was associated with increased aerobic fitness, which, in turn was related to decreases in the metabolic syndrome and inflammation. Though the model generating analysis is exploratory rather than confirmatory given the use of cross-sectional data, we might speculate that physical activity influences cardiometabolic risk in part through effects on aerobic fitness. This argues against prior evidence of independent and separate pathways for physical activity and fitness in the prediction of health outcomes [10] and supports integrated predictive relationships among these risk factors. In children and adolescents, physical activity is associated with variations in fitness, and regular participation in moderate and vigorous physical activity predicts improvements in fitness level [36]. Prior work in adults has supported the marked effect of change in fitness on cardiometabolic risk as a response to regular or vigorous physical activity. For example, stronger associations between physical activity and a metabolic syndrome risk score have been documented in adults with the lowest fitness levels [37].

Although the exact mechanisms are unknown, decreased aerobic fitness has been shown to directly influence individual cardiometabolic variables of BP (through effects on increased systemic vascular resistance, cardiac output, and plasma norepinephrine levels), insulin (reduced muscle insulin sensitivity), and lipids (reduced lipoprotein lipase activity in skeletal muscle leading to accumulation of plasma triglycerides; 38–40). Similar mechanisms involved in the relationship between fitness and inflammation include the presence of proinflammatory cytokines in the buildup of adipose tissue, as well as activation of inflammatory cytokine release from endothelial cells as a result of low exercise levels [41]. Additionally, obesity acting in combination with genetic vulnerability may underlie the cascade of events that ultimately influence the development of cardiovascular disease. For example, an "unfit" phenotype, or a genetic predisposition to low oxidative capacity, reflects an increased vulnerability to cardiometabolic risk. These individuals are described as having a lower capacity for physiologic modification through exercise or physical activity intervention [37] and are more likely to maintain an unfavorable caloric balance and be overweight [42].

Contribution of Sleep to Cardiometabolic Outcomes

Our findings suggest that sleep may in part influence cardiometabolic outcomes through associations with fitness. Specifically, reduced sleep duration (and composite poor sleep quality and fatigue) is associated with decreased fitness, which is related to increased risk of the metabolic syndrome and inflammation. Few investigations have examined the connection between exercise and sleep in adolescents and young adults, and bidirectional effects have been reported. Adolescent athletes engaging in high levels of exercise have favorable sleep patterns, including higher sleep quality and better psychological functioning [43]. In physicians, sleep loss during clinic shift-work rotations leads to adverse effects on aerobic fitness [44]. Differences in slow wave sleep, along with other homeostatic sleep regulating mechanisms, are thought to contribute to associations between sleep loss and poor fitness [45]. For example, exercise supports nocturnal temperature downregulation and reduced light sleep and sleep onset latencies [46], which may reciprocally decrease daytime fatigue and allow for individuals to engage in continued exercise and activity following restorative sleep.

No study has directly examined the hypothesis that fitness mediates the relationship between sleep and cardiometabolic outcomes. One potential mechanism could involve changes in appetite regulation and energy balance. Specifically, short sleep duration increases the ghrelin-to-leptin ratio, which promotes hunger, appetite, and cravings for sweets, starches, and salty snacks [47]. In a recent study, adolescent girls reporting short sleep had elevated ghrelin levels and consumed more carbohydrates compared with normal sleepers [48]. Reduced motivation for physical exercise is also thought to interact with increased caloric intake to produce energy balance disruptions that result in weight gain [21], a significant contributor to insulin resistance and inflammatory states.

The decreased sleep duration reported in our sample may also be a marker for pathophysiologic processes characterized by increased activation of the HPA axis and release of proinflammatory cytokines, which in the presence of lack of exercise, may become more toxic. Vgontzas and colleagues [49, 50] proposed that daytime IL-6 hypersecretion and HPA axis activation, in combination with visceral obesity, lead to fatigue and poor sleep. Sleepiness and fatigue may compound the effect of reduced levels of exercise on insulin resistance and adiposity, as others have found that insufficient physical

activity is associated with increased fatigue [51] and that lack of regular exercise in men with sleep apnea strongly predicts excessive daytime sleepiness [52]. Thus, pathways from sleep to cardiometabolic dysfunction through decreased aerobic fitness may reflect a combination of physiologic changes including change in appetite, disruption in the stress system and cytokine response, and behavioral changes that include decreased energy and exercise interest.

Regarding the notion that sleep is related to physical activity or total energy expenditure, results from the model revealed a significant inverse correlation between the sleep latent variable and physical activity. While it may be that longer sleep duration is related to decreased physical activity and that presence of sleep apnea or obesity underlies this association in some adolescents, another likely explanation involves the artifact of calculation. Although we removed sleep duration from the direct calculation of total energy expenditure (as both are derived from the 7-day physical activity recall of 24-h activity patterns), it makes intuitive sense that reduction in sleep time would result in a concomitant increase in other physical activity levels (e.g., low intensity activities) in our sample [53].

Our hypothesis of a direct association of the sleep latent variable with metabolic syndrome and inflammation latent outcomes was not supported. While depriving individuals of sleep in a laboratory environment results in temporary inflammatory marker changes among adults [17, 18], it may be that chronic shortened sleep duration is not as conclusively linked to static measures of inflammatory status in adolescents. Given that long sleep duration is potentially related to metabolic syndrome components [13, 14], observation of continuous latent indicators of sleep duration rather than examination of groups of long versus short sleepers may have masked effects of both short and long sleep duration on the metabolic syndrome. Additionally, sleep apnea may drive associations between long sleep duration and the metabolic syndrome [54], but it is unknown how the presence of sleep-disordered breathing may contribute to variability in sleep duration for our sample.

Limitations and Future Directions

Our cross-sectional design prevents interpretation of mediation model results, specifically whether physical activity and sleep cause variations in cardiorespiratory fitness, which then lead to changes in metabolic syndrome inflammation. Although we tested alternative models of association (and these did not provide adequate fit compared to the current model), experimental and longitudinal investigations are needed to draw conclusions about the etiologic influence of physical activity, fitness, and sleep on cardiometabolic risk, as well as definitively determine if reciprocal links among these variables exist. As few studies applied a similar metabolic syndrome modeling approach in adolescents, future efforts should focus on replicating the current findings using a longitudinal design so as to determine the progressive influence of metabolic risk factors on the development of cardiovascular disease. Other health behaviors (i.e., diet, smoking, and psychosocial variables) involved in the interplay between sleep, physical activity, fitness, and cardiometabolic risk also warrant further investigation. Additionally, given that the current sample was at increased risk for the metabolic syndrome by virtue of elevated BP status, findings may not generalize to healthier adolescents not at increased risk of cardiovascular illness.

The associations among physical activity, sleep, and cardiometabolic variables may have been attenuated due to measurement error. Self-reported physical activity is subject to inaccuracies and recall bias in youth [55]. Given that there is limited evidence examining correlations between physical activity assessed with accelerometry and cardiometabolic risk [1], research is needed to determine the extent to which fitness is influenced as a function of physical activity and sleep and if this corresponds to cardiovascular health changes. Pathway estimates in the model, though statistically significant, may not translate into clinically meaningful effects. However, reduced fitness had a large effect on increased disease risk (standardized coefficients or effect size estimates> 0.5), while shorter sleep and decreased physical activity had small to moderate effects on poor fitness (coefficients=0.19 and 0.26, respectively). Finally, the current study was limited in assessment of sleep and could have benefited from use of overnight polysomnography and validated sleep questionnaires (e.g., Pittsburg Sleep Quality Index, Cleveland Adolescent Sleepiness Questionnaire, Insomnia Severity Index) in order to extrapolate if unrecognized morbidity or presence of a sleep disorder contributed to observed indirect relationships between sleep and cardiometabolic outcomes.

Conclusion

In sum, this investigation demonstrated that lifestyle factors of decreased physical activity and short sleep duration, poor sleep quality and increased fatigue were associated with increased risk of the metabolic syndrome and inflammation possibly through reduced cardiorespiratory fitness in adolescents at risk for cardiovascular disease. The associated increased prevalence of obesity and sleep disturbances, along with declines in energy expenditure, suggest that

substantial public health benefits may be achieved through modification of these risk factors in youth.

Conflict of Interest Statement The authors have no conflict of interest to disclose.

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