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Combination of sleep duration, TV time and body mass index is associated with cardiometabolic risk moderated by age in youth

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Abstract

Objectives: The combination of sleep duration, television (TV) time and body mass index (BMI) may be related to the alteration of cardiometabolic risk. However, there are few studies that use these variables grouped, and showing the moderating role of age. This study aimed to verify if the combination of sleep duration, TV time and BMI is associated with cardiometabolic risk and the moderating role of age in this relationship in youth.

Methods: Cross-sectional study conducted with 1411 adolescents (611 male), aged 10–17 years. Sleep duration, TV time and BMI were assessed and grouped into eight categories. Cardiometabolic risk was assessed by a continuous metabolic risk score, including the following variables: low HDL-cholesterol, elevated triglycerides, dysglycemia, high systolic blood pressure, high waist circumference and low cardiorespiratory fitness. Generalized linear models were used to test moderation of age in the relationship between the eight categories of sleep duration/television time/BMI with cardiometabolic risk.

Results: Cardiometabolic risk factor showed association with all overweight or obesity independent of sleep time and TV time. Age moderated the relationship between sleep duration/television time/BMI with cardiometabolic risk. This association was stronger in younger adolescents (11 and 13 years), indicating that individuals with inadequate sleep, prolonged TV time and overweight/obesity present higher cardiometabolic risk values when compared to 15-year-old adolescents.

Conclusion: Overweight/obesity, independently of sleep duration and TV time, is the main risk factor for cardiometabolic disorders in adolescence. When moderated by age, younger adolescents that presented the combination of risk factors had higher cardiometabolic risk.

Keywords: adolescents; health; obesity; prevention; sleep; television.

Introduction

Cardiometabolic diseases present their first signs in childhood [1], through the early development of cardiometabolic risk factors [2]. There is evidence that lifestyle changes, such as high screen time, inappropriate sleep duration and physical inactivity are negatively affecting the cardiometabolic health of adolescents [3]. Evidence also indicates that overweight seems to be related to the development of these diseases in adulthood [4, 5]. However, the alteration in cardiometabolic risk has already been observed in individuals with severe obesity during adolescence [6].

Other behaviors, such as prolonged screen time [7] and inadequate amount of sleep also seem to reflect on the development of diseases [8]. Studies are already pointing out that in individuals who sleep little or too much the risk of all-cause mortality is increased [9, 10]. This is a worrying scenario, once children and adolescents do not meet the recommendations for sleep time and screen exposure [11, 12]. Also overweight and obesity prevalence have increased significantly in recent years in this population [13]. These facts deserve attention, as failure to comply with these recommendations may interfere with the quality of life and overall health [11, 14]. In addition, individuals with inadequate sleep hours, elevated screen time and

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overweight or obesity are more likely to develop cardiometabolic diseases [1, 7, 8].

Thus, it is important to understand the combination between sleep duration, television (TV) time and overweight, since there are few studies that consider these variables grouped. In the literature, the combination of sleep, TV time and physical activity related to cardiometabolic risk is more commonly used [12, 15, 16]. However, it is important to approach sleep, TV time and overweight condition, since cardiometabolic diseases are highly affected by excess adiposity.

It is also important to consider that sociodemographic factors, such as age, may also be related to different cardiometabolic risk status among pre-adolescents and adolescents [17]. Other aspects that could present differently among age groups are aspects related to the environmental, biological and social factors that may intervene in the lifestyle of children and adolescents, [18, 19]. Therefore, it is suggested that age may modify the association between sleep duration, TV time and overweight with cardiometabolic risk. Also, we intend to determine which age category has the strongest association between behaviors lifestyle (sleep and TV time) and nutritional status with the risk of developing cardiometabolic diseases. Our hypotheses were that the combination of inadequate sleep, high TV time and the presence of overweight/obesity are strongly associated with the cardiometabolic risk in this population. In addition, when presenting the combination of inadequate sleep, prolonged TV time and the presence of overweight/obesity there is a difference between younger and older adolescents.

In this sense, the objective of the present study was to verify whether the combination of sleep duration, TV time and BMI is associated with cardiometabolic risk and the moderating role of age in this relationship in youth.

Methods

Study design and participants

This cross-sectional study was conducted with 1411 adolescents of both sexes (611 males), aged 10–17 years, from public and private schools in the urban and rural areas of a city in southern Brazil (Santa Cruz do Sul, Rio Grande do Sul state). To calculate the sample size in order to determine whether there was enough test power for the analysis applied, considering also the effect size, the G*Power 3.1 program (Heinrich-Heine-Universität - Düsseldorf, Germany) was used. According to Faul et al. [20], the most appropriate statistical test to use was the multiple linear regression, which determined a minimum sample of 934 adolescents. The following reference parameters were used: test power $(1-\beta)=0.95$, effect size $(f^2)=0.02$ and significance level $\alpha=0.05$.

In order to form a cohort, since 2004, the same schools have been invited to participate in our research, entitled Schoolchildren's Health. For this, a survey was conducted in the city to verify the number of schools (n=50) and school children (n=17,688) enrolled. Then, the population density of students from all regions of the city (public networks, including municipal and state and private schools) was calculated. Finally, the total number of individuals to be included in the evaluation was reached. Participating schools were defined by lot. To be included in the sample of the present study, individuals should be aged between 10 and 17 years old, agreed to participate in the research by signing the free and informed consent form by parents or guardians and individuals aged ≥12 years should have signed the consent form. Adolescents who did not complete all questions related to the study aim, who were not fasting for 12 h and had not completed the cardiorespiratory fitness test (CRF) were excluded from the study (Figure 1). The evaluations were carried out at the University facilities in 2016 and 2017.

Ethics

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. In addition, this study has been conducted in accordance with Resolution 466/2012 of the National Council of Health in Brazil. Informed consent was obtained from all individual participants included in the study. The study was approved by the Research Ethics Committee of the University of Santa Cruz do Sul (UNISC) under Opinion No. 2936223.

Measures

To calculate BMI, height and weight were determined using the anthropometric scale (Filizola®), which contains a stadiometer. To do this, the students were instructed to stand barefoot and stand together. The results were classified according to the percentile curves of the World Health Organization [21], for age and sex, in which individuals with percentile ≥85 were classified as overweight and percentile ≥ 97 were classified as obese.

Data regarding sleep duration, TV time, age and color/ethnicity were collected through a self-reported questionnaire. Sleep duration

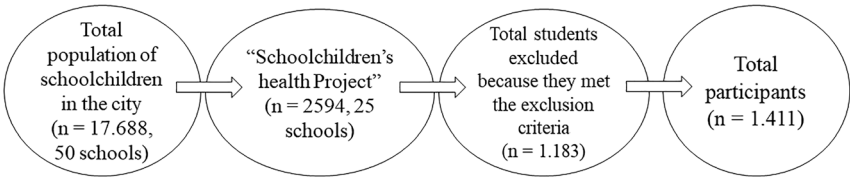


Figure 1: Sample selection flowchart.

was obtained by asking: "What time do you usually sleep during the week?/weekend?" and "what time do you usually wake up during the week?/weekend?". Because the questions refer to the week and the weekend, both were averaged to obtain total sleep time. Sleep duration classification was performed according to National Sleep Foundation's reference values for short time, adequate time and long sleep time (≤8 h, 9–11 h and ≥12 h, for individuals 10–13 years and ≤7 h, 8–10 h and ≥11 h for teenagers 14–17 years old, respectively) [22], being recategorized into adequate sleep and inadequate sleep (short and long sleep). The following question was asked to evaluate TV time: "Indicate the number of hours or minutes you watch TV per day". The adolescents answered in hours, which were turned into minutes for statistical purposes. The classification followed the parameters proposed by the American Academy of Pediatrics [23], considering the short time in front of the screens (<2 h daily) and the long time in front of the screens (≥2 h daily). The time in front of the videogame and computer were also evaluated, however, the longest time reported by the adolescents was in front of the TV, therefore, only this time was included in the present study.

The combination of behaviors and nutritional status included sleep duration, TV time and BMI, categorizing them as "adequate sleep/low TV time/normal weight"; "adequate sleep/ prolonged TV time/normal weight"; "adequate sleep/short TV time/overweight"; "adequate sleep/ prolonged TV time/overweight"; "inadequate sleep/ short TV time/normal weight"; "inadequate sleep/ prolonged TV time/ normal weight"; "inadequate sleep/short TV/overweight" and "inadequate sleep/ prolonged TV/overweight". It was considered as "overweight", individuals who were overweight or obesity.

Cardiometabolic risk assessment

Blood pressure was evaluated using a sphygmomanometer, a stethoscope on the right arm and a cuff that was adequate to the adolescent's arm circumference. Subjects were instructed to sit and rest for 5 min. The classification followed the parameters of the VII Guidelines of the Brazilian Society of Cardiology [24], through the 90th and 95th percentiles for the borderline range and hypertension, respectively, considering sex, age and height of the adolescent.

For biochemical analysis, serum samples and commercial kits (DiaSys Diagnostic Systems, Germany), performed on Miura 200 automated equipment (I.S.E., Rome, Italy), were used. Blood collection was performed after 12 h-fasting. To classify glycemia, the American Diabetes Association cut-off points were used [25] and the classification of triglycerides (TG) and high density lipoproteins cholesterol (HDL-c) followed the protocols of National Heart, Lung, and Blood Institute [26]. The biochemical results were converted from ml/dL to mmol/L for inclusion in the cardiometabolic risk score, using the following formulas: glycemia = glycemia*0.0555, TG = TG/88,57 and HDL-c = HDL-c/38,67.

Waist circumference (WC) was measured at the narrowest part of the trunk between ribs and iliac crest using a 1 mm-resolution inelastic tape (Cardiomed®). The classification was obtained according to the protocols of Fernández et al. [27], which considers abdominal obesity when WC is above the 75th percentile, according to age and sex. The CRF was evaluated through the 6-min run/walk test on the sports track and classified according to the protocols of *Projeto Esporte Brasil* - PROESP-BR [28]. The result was obtained in meters and to convert to VO_{2peak} we applied the formula: VO_{2peak} = 41.946 + 0.022 (6 min)– 0.875 (BMI) + 2.107 (sex) [29].

Cardiometabolic risk was assessed by calculating the continuous metabolic risk score (cMetS), including the following continuous variables as risk factor: HDL-c, TG, glycemia, systolic blood pressure (SBP), WC and CRF [30]. For this, a sum of the z-score of each variable is calculated. The z-score was calculated using the formula: z-score=([value of continuous variable - cutoff points]/standard deviation). The cutoff points and standard deviation used was indicated in the study of Stavnsbo et al. [31]. Sex and age of adolescents were considered in the z-score calculation of each of the variables included as a risk factor. Because of the inverse relationship between CRF and HDL-c with cardiometabolic risk, the score of these variables was multiplied by –1 [30]. There is evidence indicating that for the assessment of the cardiometabolic risk of children and adolescents, the cardiometabolic risk score is the best to be used, given that each risk factor is considered according to age and sex [30–32].

Statistical analysis

For the characterization of the adolescents, descriptive statistics was applied through absolute and relative frequency, mean and standard deviation. The following generalized linear models were tested: Model 1: association between sleep duration/TV time/BMI and age and cardiometabolic risk; Model 2: interaction (sleep duration/TV time/ BMI x age) and cardiometabolic risk. The PROCESS macro was used for the Statistical Package for the Social Sciences (SPSS) program, using linear regression models to verify the role of age categories in the relationship between sleep duration/TV time/BMI and cardiometabolic risk. The age categories were provided by the Johnson–Newman technique in which age was classified according to tertiles (11, 13 and 15 years). Values of $p<0.05$ were considered significant. All analyzes were adjusted by sex and skin color/ethnicity and tested using the statistical program SPSS vs. 23.0 (IBM, Armonk, USA).

Results

The sample of the present study was composed of 1411 adolescents, of both sexes, presenting mean age 12.74 ± 1.96. It was found that 28.1% of adolescents had inadequate sleep, 48.9%, prolonged TV time and 38.3% showed overweight and obesity. In addition, 5.7% presented inadequate sleep/prolonged TV time/overweight (Table 1). Present data also indicate that age group 12–13 years demonstrated higher frequency of inadequate sleep (34.3%, p=0.001) and combination of inadequate sleep/prolonged TV time/overweight (8.1%, p=0.001). The age group 10–11 years presented higher frequency of overweight (46.3%, p=0.001) compared to the age group 14 to 17 to years (Data not presented in tables).

Table 2 shows associations between sleep duration/TV time/BMI and cardiometabolic risk. In model 1, cardiometabolic risk values are higher when individuals have adequate sleep/short TV time/overweight (β=3.838; CI95% = 3.344; 4.332), adequate sleep/prolonged TV time/ overweight (β=4.096; CI95% = 3.612; 4.579), inadequate

Table 1: Characteristics of adolescents.

	n (%) (n=1411)
Sex	
Male	611 (43.3)
Female	800 (56.7)
Age classification	
10–11 years	428 (30.3)
12–13 years	484 (34.3)
14–17 years	499 (35.4)
Skin color/ethnicity	
White	1121 (79.4)
Black	97 (6.9)
Brown/Mulatto	174 (12.3)
Indigenous/yellow	19 (1.3)
Sleep duration	
Adequate sleep	1015 (71.9)
Inadequate sleep	396 (28.1)
TV time	
Low TV time	721 (51.1)
Prolonged TV time	690 (48.9)
BMI classification	
Underweight/normal weight	871 (61.7)
Overweight/obesity	540 (38.3)
Sleep duration/TV time/BMI	
Adequate sleep/low TV time/normal weight	347 (24.6)
Adequate sleep/ prolonged TV time/normal weight	302 (21.4)
Adequate sleep/short TV time/overweight	177 (12.5)
Adequate sleep/ prolonged TV time/overweight	189 (13.4)
Inadequate sleep/short TV time/normal weight	103 (7.3)
Inadequate sleep/ prolonged TV time/normal weight	119 (8.4)
Inadequate sleep/short TV/overweight	94 (6.7)
Inadequate sleep/ prolonged TV/overweight	80 (5.7)
	Mean (SD)
Cardiometabolic risk, z-score	−0.23 (3.31)
WC, cm	67.99 (8.93)
$VO_{2\ peak}$, mL/kg/min	44.41 (6.73)
SBP, mmHg	107.57 (12.97)
TG, mmol/L	0.81 (0.36)
HDL-c, mmol/L	1.48 (0.27)
Glycemia, mmol/L	4.91 (0.38)

n, number of subjects; %, percentage; TV, television; BMI, body mass index; SD, standard deviation; WC, waist circumference; VO_{2peak}, peak oxygen uptake; SBP, systolic blood pressure; TG, triglycerides; HDL-c, high density lipoproteins cholesterol.

sleep/short TV time/overweight (β=3.998; CI95% = 3.379; 4.617) and inadequate sleep/prolonged TV time/overweight (β=4.304; CI95% = 3.645; 4.963). Age also presented a positive association with cardiometabolic risk (β=0.097; CI95% = 0.023; 0.170). Model 2, on the other hand, indicates a negative interaction between inadequate sleep/prolonged TV time/obese x age and cardiometabolic risk (β=−0.558; CI95% = −0.925; −0.191), indicating that with increasing age the association between inadequate sleep/prolonged TV time/obese and cardiometabolic risk is weakened. For the other categories of the combination of sleep duration/TV time/BMI, age does not intervene in the association with cardiometabolic risk.

We have also tested the association between BMI with the cardiometabolic risk score, excluding the WC in the score. Indeed, the association remains significant between the combination of sleep duration, TV time and BMI with the cardiometabolic risk, indicating that the associations are not driven by the presence of WC in the score (see Supplemental Material, Supplemental Table 1).

The interaction indicates that the relationship between sleep duration/TV time/BMI and cardiometabolic risk was stronger in younger adolescents (11 and 13 years) than in older adolescents (15 years) (Figure 2). In addition, individuals with inadequate sleep, prolonged TV time and overweight present higher cardiometabolic risk values when compared to 15-year-old adolescents (Figure 2).

Discussion

Main findings of the present study indicate that individuals who are overweight, regardless of sleep duration and TV time, have higher cardiometabolic risk values. In addition, our data suggest that younger adolescents (11 and 13 years) that presented combination of risk factors (inadequate sleep/prolonged TV time/overweight) showed higher values of cardiometabolic risk factors when compared to 15 years or older. These findings deserve attention at the public health level, due to the cardiometabolic risk being more strongly associated with overweight individuals, regardless of whether or not they comply with sleep and TV recommendations. Therefore, lifestyle factors, especially the ones related to the increase in obesity cases, need to be monitored since childhood, in order to avoid major health problems in adolescence and adulthood. This association can be mainly explained by obesity condition, that already present changes in cardiometabolic risk values [1, 6] and by adopting an inappropriate lifestyle in this age group [3, 33] reflecting on excess body weight and consequently impairments on cardiometabolic health. In addition, there is a need to consider that the precarious security in public places destined to physical activity practice may lead to a longer time at home and consequently to increased sedentary behaviors [34]. Based on this, it is suggested that the first symptoms of cardiometabolic diseases are present in childhood through the increase of body weight and the deleterious effects that childhood obesity causes on cardiometabolic health can already be observed in adolescence [35].

Table 2: Association between sleep duration/TV time/BMI and age with cardiometabolic risk.

	Cardiometabolic risk		
	B	**(IC, 95%)**	**P**
Model 1 (AIC: 6843.902; BIC: 6922.683)			
Sleep Duration/TV time/BMI			
Adequate sleep/short TV time/ normal weight	1		
Adequate sleep/prolonged TV time/normal weight	0.153	(−0.266; 0.571)	0.475
Adequate sleep/short TV time/ overweight	**3.838**	(3.344; 4.332)	**0.001**
Adequate sleep/prolonged TV time/overweight	**4.096**	(3.612; 4.579)	**0.001**
Inadequate sleep/short TV time/ normal weight	0.344	(−0.254; 0.942)	0.260
Inadequate sleep/prolonged TV time/normal weight	0.148	(−0.417; 0.713)	0.608
Inadequate sleep/short TV time/ overweight	**3.998**	(3.379; 4.617)	**0.001**
Inadequate sleep/prolonged TV time/overweight	**4.304**	(3.645; 4.963)	**0.001**
Age	**0.097**	(0.023; 0.170)	**0.010**
Model 2 (AIC: 6843.584; BIC: 6959.130)			
Sleep Duration/TV time/BMI X age			
Adequate sleep/short TV time/ normal weight X age	1		
Adequate sleep/prolonged TV time/normal weight X age	0.047	(−0.162; 0.257)	0.659
Adequate sleep/short TV time/ overweight X age	−0.049	(−0.294; 0.197)	0.698
Adequate sleep/prolonged TV time/overweight X age	−0.041	(−0.304; 0.222)	0.762
Inadequate sleep/short TV time/ normal weight X age	0.078	(−0.220; 0.375)	0.610
Inadequate sleep/prolonged TV time/normal weight X age	−0.175	(−0.469; 0.119)	0.243
Inadequate sleep/short TV time/ overweight X age	−0.252	(−0.570; 0.067)	0.121
Inadequate sleep/prolonged TV time/overweight X age	**−0.558**	(−0.925; −0.191)	**0.003**

β, linear regression (the models were adjusted for sex and skin color/ethnicity); CI, confidence interval; AIC, Akaike Information Criterion; BIC, Bayesian Information Criterion; TV, television; BMI, body mass index.

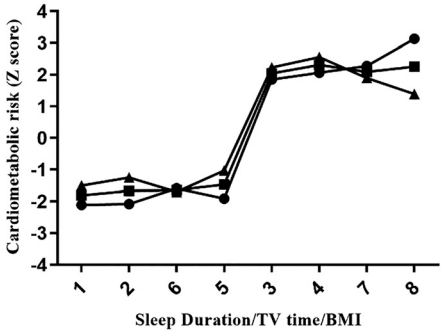
Model 1: association between sleep duration/TV time/BMI and age with cardiometabolic risk; Model 2: the statistic significative moderation role of age on the relationship between sleep duration/TV time/BMI with cardiometabolic risk factors. In Figure 2, it is possible to verify the higher risk of the combination factors on younger individuals. It was considered as "overweight", individuals who were overweight or obesity.

Bold numbers represents the significative values.

However, the relationship between the combination of hours of sleep, TV time and BMI with cardiometabolic risk is stronger in younger adolescents with inadequate sleep, prolonged TV time and overweight. Indeed, in adolescents aged 11–13 years, not meeting the recommendations of these risk factors is more harmful to the development of cardiometabolic risk than in 15-year-olds, than only being overweight as observed in adolescents independently of the age classification. In the sense, it is observed that sleep duration and TV time when grouped with BMI cause effects in cardiometabolic risk only in younger adolescents when compared to older adolescents. Indeed, our data showed that the age group 10–11 years presented higher frequency of overweight and the age group 12–13 years demonstrated higher frequency of inadequate sleep and combination of risk factors. These findings could be explained by different aspects, for example, the urban process reflects in the security of publics places destined for physical activity practice, then parents may not allow younger adolescents to frequent these places [36]. In addition, older adolescents are joining in the labor market and involved with others activities that are related with smaller time spent in sedentary activities. It is known that having inadequate sleep duration and TV time [11, 37] and overweight [35] can negatively affect the quality of life among this age group.

Moreover, the literature studies show that overweight [1, 5], short sleep [8, 38, 39] and the high time spent in front of TV [7, 40] when observed in early childhood may increase the chances of cardiovascular disease. In this context, our data indicated that overweight/obesity, independently of sleep duration and TV time, is the main risk factor for cardiometabolic disorders in adolescence. When accounting for the role of age, younger adolescents that presented the combination of risk factors had higher cardiometabolic risk. However, no studies were found in our review that addressed the combination of sleep duration, TV time and BMI in this population. Based on this, it is suggested that this is one of the first studies to verify the moderating role of age in the relationship between the combination of sleep duration, TV time and BMI with cardiometabolic risk in young.

Thus, it is suggested that actions that encourage the adoption of a healthy lifestyle should be carried out urgently, in order to reduce the diseases development in adulthood. For this, multicomponent interventions are indicated with the monitoring of professionals of Physical



1: adequate sleep/low TV time/normal weight
2: adequate sleep/ prolonged TV time/normal weight
3: adequate sleep/short TV time/obese
4: adequate sleep/ prolonged TV time/obese
5: inadequate sleep/short TV time/normal weight
6: inadequate sleep/ prolonged TV time/normal weight
7: inadequate sleep/short TV/obese
8: inadequate sleep/ prolonged TV/obese

11 years
2: B=0.029; CI=-0.561; 0.620; p=0.922
3: B=3.966; CI=3.348; 4.584; p=0.001
4: B=4.177; CI=3.559; 4.795; p=0.001
5: B=0.205; CI=-0.641; 1.051; p=0.635
6: B=0.525; CI=-0.317; 1.367; p=0.221
7: B=4.388; CI=3.554; 5.222; p=0.001
8: B=5.239; CI=4.333; 6.146; p=0.001

13 years
2: B=0.041; CI=-0.280; 0.561; p=0.512
3: B=3.851; CI=3.332; 4.370; p=0.001
4: B=4.119; CI=3.604; 4.634; p=0.001
5: B=0.339; CI=-0.262; 0.940; p=0.269
6: B=0.163; CI=-0.406; 0.732; p=0.573
7: B=3.896; CI=3.267; 4.524; p=0.001
8: B=4.066; CI=3.392; 3.740; p=0.001

15 years
2: B=0.252; CI=-0.351; 0.855; p=0.412
3: B=3.736; CI=2.931; 4.541; p=0.001
4: B=4.061; CI=3.218; 4.905; p=0.001
5: B=0.472; CI=-0.382; 1.328; p=0.278
6: B=-0.199; CI=-1.001; 0.603; p=0.627
7: B=3.403; CI=2.444; 4.363; p=0.001
8: B=2.893; CI=1.806; 3.980; p=0.001

Interactions:
2: B=0.056; CI=-0.156; 0.267; p=0.605
3: B=-0.058; CI=-0.306; 0.190; p=0.649
4: B=-0.029; CI=-0.294; 0.236; p=0.830
5: B=-0.067; CI=-0.234; 0.368; p=0.663
6: B=-0.181; CI=-0.478; 0.116; p=0.232
7: B=-0.246; CI=-0.568; 0.075; p=0.133
8: B=-0.587; CI=-0.957; -0.218; p=0.002

Figure 2: Age moderation role in the relationship between sleep duration/TV time/BMI and cardiometabolic risk.

Education, Nutrition and Psychology, as well as public policies that encourage the physical activity practice, adoption of healthy eating habits [41]. In addition, holding lectures with the participation of parents demonstrating the importance of adopting a healthy lifestyle at early ages is important to reduce cases of cardiometabolic diseases [42].

The possible associations between the combination of sleep duration, TV time and BMI with cardiometabolic health, the moderating role of age and the representative sample of a city in southern Brazil are the strengths of the study. However, some limitations also stand out, such as data on sleep duration and TV time being obtained through a self-reported questionnaire by participants, which may differ from the reality of adolescents. Likewise, this study did not consider screen time, due TV is the screen most used by participants compared to video games and computers. In addition, no mobile phone time evaluation was performed. Also, we did not evaluated physical activity levels, which could be a relevant potential confounder for the present study.

In conclusion, the main factors associated with adolescents' cardiometabolic risk are overweight, regardless of sleep duration or sedentary behavior. In addition, in adolescents aged 11 and 13, non-compliance with sleep, TV time and body weight are more detrimental to the development of cardiometabolic risk. Thus, it is suggested that lifestyle aspects should be monitored from childhood to prevent the onset of cardiometabolic diseases in adolescence and adulthood.

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Authors contributions: APS, CPR, JDPR participated in data organization and designed the study. APS, ARG, AFD, CB, JDPR and CPR performed the statistical analysis. All the authors contributed to the elaboration of the manuscript with critical comments about it.

Competing interests: The authors declare no conflict of interest.

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