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Unravelling the relationship code in ISCOLE Portuguese children physical activity and sedentariness levels and patterns and obesity

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Unravelling the relationship code in ISCOLE Portuguese children physical activity and sedentariness levels and patterns and obesity

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DEDICATION

To mummy, Thacy and Polly…

For your endless love, support and encouragement… For you I am where I am, for you I continue aim higher... I am because you are!

To my dear friend Nanda...

For being “my person”…

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RESUMO

Os propósitos desta tese foram (1) descrever as diferenças inter-individuais nos níveis de atividade física e sedentarismo e de obesidade entre crianças Portuguesas, e (2) investigar suas relações com características individuais, familiares e ambientais.

A amostra foi composta por 777 sujeitos, de ambos os sexos, com idades entre 9-11 anos, participantes do projeto *International Study of Childhood Obesity, Lifestyle and the Environment* (ISCOLE). Foram utilizados procedimentos estandardizados para a recolha de dados oriundos de diferentes níveis: individual, familiar, vizinhança e escola. As análises estatísticas foram realizadas nos *softwares* SPPS, HLM, MIXREGLS e SuperMix.

Os resultados mostraram que, em geral, (I) a variância explicada pelo contexto escolar nas diferenças nos níveis de sedentarismo, atividade física moderada-a-vigorosa e IMC é baixa; (II) há uma variação significativa nos níveis de sedentarismo entre e intra-sujeitos ao longo da semana, com efeito significativo do sexo, estatuto ponderal e maturação biológica nos níveis de "consistência" e "erraticismo"; (III) foi observado um platô na prevalência de sobrepeso/obesidade entre crianças Portuguesas na última década, não obstante a elevada prevalência de crianças com excesso de peso na presente amostra; (IV) crianças com sobrepeso/obesidade e normoponderais diferem, apenas, nas variáveis biológicas, mas não nas variáveis demográficas e comportamentais; enquanto que sexo, níveis de atividade física moderada a vigorosa, hábitos alimentares, maturação biológica e estatuto socioeconómico, parecem ser variáveis preditores do IMC em crianças; (V) a percentagem de crianças que atingem os valores diários recomendados de atividade física moderada-a-vigorosa é baixa-a-moderada; (VI) crianças "ativas e normoponderais", e "ativas e com valores mais elevados de força muscular" apresentam um melhor perfil metabólico comparativamente a seus pares "inativos e com sobrepeso" e "inativos e com valores mais baixos de força muscular", respectivamente; (VII) a atividade física moderada a vigorosa parece não exercer efeito significativo em atenuar os fatores de risco metabólico em crianças com excesso de peso; contudo, níveis elevados de força muscular parecem reduzir o risco de desenvolvimento de fatores de risco metabólico em crianças com baixos níveis de atividade física; (VIII) há uma correlação negativa e significativa entre atividade física moderada-a-vigorosa e sedentarismo, mas o mesmo não foi observado entre sedentarismo e IMC; (IX) o número de irmãos, tempo de sono e estatuto socioeconómico são preditores, simultaneamente, para o sedentarismo e atividade física moderada-a-vigorosa; (X) variáveis do contexto escolar mostraram-se significativas, apenas, para o sedentarismo, e nenhuma das variáveis da escola estudadas mostraram efeito significativo sobre as diferenças nos níveis de atividade física moderada-a-vigorosa ou IMC.

Os resultados sublinharam como estes três traços interagem e como variam entre crianças. Dado os seus determinantes serem distintos, as estratégias de intervenção que tenham por objetivo reduzir o sedentarismo, aumentar os níveis de atividade física e reduzir a prevalência de sobrepeso/obesidade devem ser pensadas de modo contextualizadas para agir sobre cada uma delas, isolada e conjuntamente.

Palavras-chave: atividade física, sedentarismo, obesidade, crianças, Portugal, ISCOLE

ABSTRACT

The purposes of this thesis were (1) to describe inter-individual differences in physical activity and sedentariness levels as well as in obesity in Portuguese children, and (2) to investigate their relationships with individual, familial and environmental characteristics.

The sample comprises 777 subjects, aged 9-11 years, from both sexes, that took part in the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE) project. Standardized procedures were used to collect all data at the individual, family, neighbourhood, and school levels. Data analyses were done in SPPS, HLM, MIXREGLS and SuperMix softwares.

In general, results showed that (I) the variance explained by the school environment on sedentariness, moderate-to-vigorous physical activity and BMI is low; (II) there is a significant between- and within-subject variance in sedentariness across the week, with significant effects of sex, socioeconomic status and biological maturation in children's consistency and erraticsm; (III) it was observed a plateau in the prevalence of overweight/obesity among Portuguese children in last decade, notwithstanding the observed high prevalence of overweight/obesity in the present sample; (IV) overweight/obese and normal-weight children only differ in biological variables, but not in demographic or behavioural variables; however, sex, moderate-to-vigorous physical activity levels, nutritional habits, biological maturation, and socioeconomic status seem to predict children's BMI; (V) the percentage of children complying with the daily recommended guidelines for moderate-to-vigorous physical activity is low-to-moderate; (VI) "normal-weight and active" children and "active with high muscle strength" children have a better metabolic profile than their "overweight and inactive" and "active with low muscle strength" peers, respectively; (VII) moderate-to-vigorous physical activity does not attenuate the metabolic risk factors in children with excess weight, however high levels of muscular strength seems to reduce the metabolic risk in children with low physical activity levels; (VIII) there is a negative and significant correlation between moderate-to-vigorous physical activity and sedentariness, but this was not observed between sedentariness and BMI; (IX) the number of siblings, sleep time and socioeconomic status are predicting both sedentariness and moderate-to-vigorous physical activity; (X) school-level variables were only significant for sedentariness, but no other school level variables were significantly correlated with differences in moderate-to-vigorous physical activity or BMI levels.

These results show how these three traits interact with each other in several ways, and that their variability is clearly observed among children. Since their determinants are different, intervention strategies to reduce sedentariness, increase physical activity levels and decrease the prevalence of overweight/obese in children should be planned and developed to act independently and in conjunction in these three traits.

Key-words: physical activity, sedentariness, obesity, children, Portugal, ISCOLE.

LIST OF SYMBOLS AND ABBREVIATIONS

AHMS	Active and high muscular strength
ALMS	Active and low muscular strength
ANCOVA	Analysis of covariance
ANOVA	Analysis of variance
BMI	Body mass index
BS	Between-subject
CDC	US Centers for Disease Control and Prevention
cm	Centimetre
DBP	Diastolic blood pressure
df	Degrees of freedom
GLU	Glucose
HDI	Human Development Index
HDL-C	High density lipoprotein cholesterol
HLM	Hierarchical Linear and Nonlinear Modelling
$hours \cdot day^{-1}$	Hours per day
IOTF	International Obesity Task Force
ISCOLE	International Study of Childhood Obesity, Lifestyle and the Environment
kg	Kilogram
$Kg.m^{-2}$	Kilogram per metre squared
LR	Likelihood-ratio
M0	Null Model
M1	Model 1
M2	Model 2
m	Metre
m^2	Metre squared
MAP	Mean arterial blood pressure
mg/dl	Milligram to decilitre
$min \cdot day^{-1}$ / $minutes \cdot day^{-1}$	Minutes per day

MIXREGLS	Mixed-effects regression with location scale program
mm	Millimetre
mmHg	Millimetres of mercury
MR	Metabolic risk
MS	Metabolic Syndrome
MS	Muscular Strength
MVPA	Moderate-to-vigorous physical activity
NA	Normal-weight and physically active group
NI	Normal-weight and inactive group
NW	Normal-weight
O/O	Overweight/obese
OA	Overweight and physically active group
OI	Overweight and inactive group
P50	50th percentile
PA	Physical activity
PF	Physical fitness
PHV	Peak height velocity
SB	Sedentary behaviour
SBP	Systolic blood pressure
SD/std dev	Standard deviation
SE	Standard error
SED	Sedentariness
SES	Socioeconomic status
SPSS	Statistical Package for the Social Sciences
ST	Sedentary time
TRI	Triglycerides
WC	Waist circumference
WHO	World Health Organization
WINPEPI	Programs for Epidemiologists for Windows
WS	Within-subject
zMR	Metabolic risk score
zMS	Metabolic syndrome score
α	Alpha (between-subject variance)

β	Beta (regression coefficient)
Δ	Delta (change)
p	p-value
R^2	Coefficient of determination
ρ	Rho (correlation coefficient)
τ	Tau (within-subject variance)
σ^2	Variance
X^2	Chi-squared
%	Percentage
<	Lower than
≤	Lower or equal than
>	Higher than
≥	Higher or equal than
± / + or -	Plus or minus
≈	Approximately

CHAPTER I

General Introduction and Thesis Outline

GENERAL INTRODUCTION

It is generally accepted that the urbanization process observed in the last century, as a consequence of economic, social, cultural and behavioural changes, entailing epidemiological transitions, contributed to modify the population health status of almost nations of the world (Yusuf et al., 2001a, 2001b). This complex process promoted a higher life expectancy and the reduction of death by infectious diseases (World Health Organization, 2003), but it has also induced a more sedentary and inactive lifestyle with a marked reduction in physical activity levels (Hallal et al., 2012) and an increase in sedentariness (i.e. sitting) (Pate et al., 2011). Further, it also prompted the adoption of dietary patterns characterized by the consumption of high-energy dense foods (Rodriguez-Ramirez et al., 2011), leading to unparalleled increases in overweight/obesity incidence in the general population of all age ranges (Lobstein et al., 2004; Ng et al., 2014). In addition, and possibly as a consequence of this "new" urbanized lifestyle, a shift in the major causes of deaths from "traditional risk" (i.e. related to under-nutrition and poor sanitation) has been noted leading to a greater incidence of death by co-morbidities linked to sedentariness, inactive lifestyles and excess weight, so-called non-communicable diseases, which became a leading global cause of death (World Health Organization, 2009, 2011). Moreover, physical inactivity and excess weight are ranked the fourth and fifth leading risks for mortality worldwide, being associated with an increased risk of chronic diseases such as heart disease and cancers (World Health Organization, 2009).

The decreasing levels of physical activity, and the increasing prevalence of sedentariness and overweight/obesity in children and adolescents are of major concern, due to the fact that these traits are closely related to the development of chronic diseases in adulthood (Berenson & Srnivasan, 2005; Deshmukh-Taskar et al., 2006; Ortega et al., 2013). Although physical activity and sedentariness are two different behavioural constructs (Katzmarzyk, 2010; Pate et al., 2011), it has been suggested that their determinants might be similar (King et al., 2011; Pate et al., 2011; Uijtdewilligen et al., 2011), as they both arise from intrapersonal (biological, psychological, demographic), interpersonal (social, cultural), organizational, environmental (built, natural, social), and policy characteristics (Ferreira et al., 2007; Sallis et al., 2000; Van Der Horst et al., 2007). Though childhood obesity is a major public health problem across the world (de Onis et al., 2010; Ng et al., 2014), its individual expression is the result of a complex interaction of behavioural, biological and environmental factors (Kumanyika & Obarzanek, 2003; Damiani & Damiani, 2010). As such, recent investigations have focused their attention on unravelling the roles of physical activity and sedentariness in promoting excess weight in youth (Steinbeck, 2001), and further link these traits to their health.

Adolescence is considered a unique time-window in human development (Steinberg & Morris, 2001), characterized by important changes in life, especially in terms of the interactions between individual lifestyles and environmental conditions. Since behaviours and different aspects of healthy/unhealthy statuses acquired in this period of life tend to track through adulthood (Malina, 2001; Singh et al., 2008), the promotion of healthy lifestyles and education on solid human values in early years are very important venues to also reduce the incidence of non-communicable diseases in adult life. There is evidence that up to 50% of obese adolescents may remain obese in adulthood (Steinbeck, 2001). For example, in the Bogalusa Heart Study, 22.5% of the participants who were overweight in childhood remained overweight in young adulthood, and only 2.3% of the overweight children became normal-weight as adults (Deshmukh-Taskar et al., 2006). Similarly, data from the Fels Longitudinal Study showed a moderate-to-high prediction of adult body mass index (BMI) according to child/adolescent BMI (Guo & Chumlea, 1999; Guo et al., 2000; Guo et al., 2002); and The Physical Activity Longitudinal Study reported that over a 22 year period, about 83% of overweight youth remained overweight as adults, and that almost all healthy weight adults had been healthy weight youths (Herman et al., 2009). With respect to the tracking of physical activity and sedentary behaviour, results are not always conclusive (Herman et al., 2009; Telama, 2009), but a trend exists where adequately active youth become adequately active adults (Azevedo et al., 2007; Malina, 2001). Likewise, metabolic syndrome indicators, linked to low levels of physical activity, high levels of sedentariness and excess weight during childhood and adolescence, tend also to track well from childhood into adulthood (Camhi & Katzmarzyk, 2010; Eisenmann et al., 2004).

These previous pieces of evidence reinforce the significant links between physical activity, sedentariness, overweight/obesity and several co-morbidities, requiring a better understanding of their interwoven net during early life (Andersen et al., 2006; Brambilla et al., 2011; Butte et al., 2005; Steele et al., 2008). However, the complex nature of these traits, whose predictors originate from different sources, requires the use of “substantive” models allowing the examination of their relationship from multiple layers of influence. A useful framework of thought and empirical research is the so-called *Ecological Model of Human Development*, initially developed by Urie Bronfenbrenner (Bronfenbrenner, 1977, 1979) and later adapted for the field of physical activity by Sallis, Owen and Fisher (2008). The focus of this model is on the subject-environment dyad, i.e., the investigation of individual development within the manifold environmental facets. Since active or sedentary behaviours occur in specific “places”, the use of the ecological model is well suited for research, acknowledges the identification of “place” characteristics that facilitate or impair their occurrence. The *Ecological Model of Four Domains of Active Living* (Sallis et al., 2006), includes several levels of interwoven influences, such as intrapersonal factors, perceived environment, behaviour, behaviour settings, and policy environment, and is an interesting approach to ground this purpose (Figure 1). This is also so because the higher order predictors, namely environmental and policy factors, have received less attention than individual ones when correlates of youth physical activity, sedentary behaviour and BMI are investigated and/or interventions are planned (Sallis et al., 2006; Story et al., 2008).

Briefly, this model shows (Figure 1), at its centre, broad categories of intrapersonal variables, representing the individual. Individual environmental perception and objective aspects of the environment are distinguished, and both are likely to be important. Behaviour represents the interaction between the person and the environment, and is highlighted because this is the outcome of interest, and the four main domains of active living are presented: active recreation, active transport, occupational activities, and household activities. Behaviour settings are the places where the outcome variable occurs, being relevant to account both access to settings and specific characteristics. The policy environment can influence active living by different pathways, i.e., through the built environment, incentives, and programs. The socio-cultural environment, information environment, and natural environment are shown as cutting across the other levels (Sallis et al., 2006).

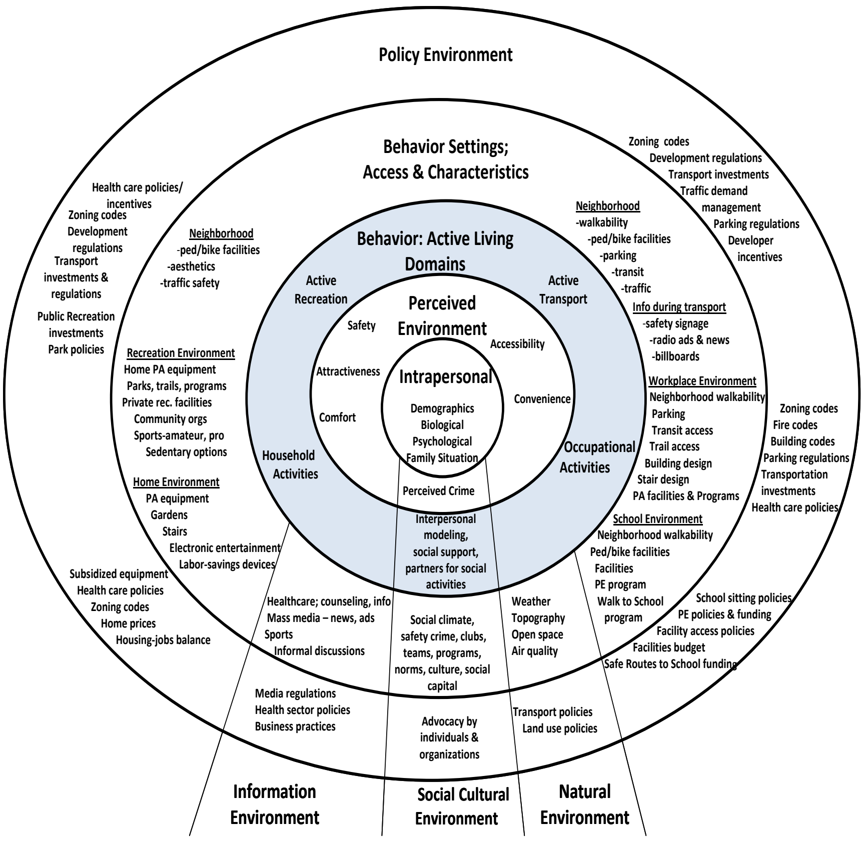

Figure 1. Ecological Model of Four Domains of Active Living, adapted from Sallis et al. (2006).

As such, it is relevant to investigate the role of variables coming from different levels of the ecological model on children's physical activity, sedentariness and obesity, as well as the correlations among them. We will be mostly interested in the roles of biological traits, family demographics and school context. Investigating the impact of lifestyle on childhood overweight/obesity, physical activity and sedentariness in a large study, involving countries from the major world regions (Eurasia & Africa, Europe, Latin America, North America, and Pacific) will provide high-impact results on the development of lifestyle interventions to reduce behavioural risks during childhood that can be culturally adapted for implementation around the world.

The International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE) (Katzmarzyk et al., 2013) is a multi-national cross-sectional study conducted in 12 countries (Australia, Brazil, Canada, China, Colombia, Finland, India, Kenya, Portugal, South Africa, United Kingdom, United States), with the purpose "to determine the relationship between lifestyle behaviours and obesity in a multi-national study of children, and to investigate the influence of higher-order characteristics such as behavioural settings, and the physical, social and policy environments, on the observed associations within and between countries" (p. 4). Using information from ISCOLE-Portugal, this thesis was developed aiming to unravel the relationship between physical activity, sedentariness and obesity, as well as their co-morbidities, in Portuguese children.

The present doctoral thesis has its foundations on four main reasons:

- The first one is grounded in the epidemiology of physical activity framework, which has a strong interest in studying predictors and correlates of physical activity and sedentary behaviour (Caspersen, 1989). Previous studies have suggested that children spend a considerable portion of their awake time in sedentary activities (Biddle et al., 2009; Pate et al., 2011), while their physical activity levels have decreased globally - more than 80% of adolescents aged 13-15 years do not comply with the daily recommended levels (Hallal et al., 2012). A study involving Portuguese children and youth showed that at ages 10-11 yrs, 36% of them were considered sufficiently active; further, a progressive decrease was observed in this prevalence with age, where only 4% of youth aged 16-17 complied with the physical activity daily guidelines. Furthermore, there is also evidence that low childhood physical activity and high levels of sedentariness are potential risk factors for the development of chronic diseases in later life, namely cardiovascular disease (Chau et al., 2013; Dunstan et al., 2010; Katzmarzyk et al., 2009; Steele et al., 2008; Thorp et al., 2010; Vicente-Rodriguez et al., 2008). Furthermore, they are related to a worse metabolic profile in children and adolescents (Steele et al., 2008; Vicente-Rodriguez et al., 2008). As such, it is mandatory to change this scenario, increasing youth physical activity and, in addition, reducing time spent in sedentary behaviour.

However, since both physical activity and sedentary behaviour occur in specific settings, it would be useful to take into account not only biological variables, but also the environment where the child lives and spends his/her daily activities. For example, home demography has a relevant role on children’s sedentariness and active lifestyle, particularly parental and sibling influences (Anderssen & Wold, 1992; Atkin et al., 2013; Beets et al., 2010; Seabra et al., 2009; Tandon et al., 2014; Tandon et al., 2012; Verloigne et al., 2012), family socioeconomic status (Atkin et al., 2013; Brodersen et al., 2007; Newton et al., 2011; Olds et al., 2010; Seabra et al., 2009; Steele et al., 2010), and availability of sports equipment or sedentary entertainment at home (Rosenberg et al., 2010). In addition, children spend most of their awake time at school, and this is an important environment for children to increase their physical activity levels and also decrease the time spent in sedentary activities, and a number of previous investigations reported the role of the school environment and policies in the promotion of an active lifestyle among students (Cradock et al., 2007; Ridgers et al., 2007; Sallis et al., 2001; Verstraete et al., 2006; Wechsler et al., 2000).

Since physical activity and sedentariness are complex and multifactorial behaviours, determined by biological, demographical, psychological, behavioural, sociocultural, and environmental factors (Ferreira et al., 2007; Sallis et al., 2000; Van Der Horst et al., 2007), it is mandatory to examine the influence of these different sets of predictors, arising from different levels, using multilevel models (Diez-Roux, 2000; Duncan et al., 1998; Snikders & Bosker, 1994).

- Secondly, notwithstanding the observed trend of increases in sedentariness and decreasing physical activity levels in youth, their levels and patterns differ according to their intrapersonal traits, interpersonal characteristics, built, physical and social environmental factors (Brodersen et al., 2007; Brodersen et al., 2005; Gorely et al., 2007; Mota et al., 2008; Pate et al., 2011; Pulsford et al., 2013; Sallis et al., 2000; Uijtdewilligen et al., 2011; Van Der Horst et al., 2007). Since there is a periodicity in daily physical activity patterns that approximates seven days or fewer (Rowlands et al., 2015), observations for a whole week of monitored activity and sedentary behaviour seems to be sufficient to study children's daily patterns variability. Exploring covariates related to within- and between-subject variance in sedentariness, i.e., erraticism and heterogeneity, can offer reliable information to be used when developing efficient interventions aiming to decrease sedentary behaviour. Although this is very important information, we are not aware of any study with this purpose in mind, and the lack of this information exists.

- The third reason is related to public health concerns about the current poor health status of youth, namely the high prevalence of overweight/obesity and its co-morbidities, such as elevated metabolic risk factors. Recent data reported that, between 1980-2013, the overweight/obesity prevalence increased in developed countries from 16.2% to 22.6% in boys, and from 16.9% to 23.8% in girls, whereas in developing nations these increases were from 8.1% to 12.9% in boys and from 8.4% to 13.4% in girls (Ng et al., 2014). Although this trend appears to reach a plateau in some countries (Olds et al., 2011), its prevalence remains high, and data from the World Health Organization estimates that there were about 43 million preschool children with overweight/obesity in 2010, and this prevalence is expected to reach approximately 60 million in 2020 (de Onis et al., 2010). In the Portuguese context, similar trends have been observed, with an increase in children's BMI in the last decades (Padez et al., 2004), and prevalence of overweight/obesity, in youth aged 10-18 yrs, ranging from 22.6% to 31.7%, depending on the cut-point used (Sardinha et al., 2011). Since excess weight is linked with the development of metabolic health problems in both children and adults

(Berenson & Srnivasan, 2005; Deshmukh-Taskar et al., 2006; Tailor et al., 2010; Weiss et al., 2013), exploring the importance of its correlates is a relevant endeavour when designing and implementing strategies to reduce overweight/obesity in youth.

The actual incidence of metabolic risk in the paediatric population also calls for attention to reduce its incidence and investigate associated factors. It is known that metabolic risk in youth is closely connected to excess weight. For example, Saland (2007) reported that among obese youth the prevalence of metabolic syndrome varied between 18% and 50%, being 1% or less among normal-weight youth[1]. Tailor et al (2010), in a review study, noted that the prevalence of metabolic risk in children and adolescents ranged from 1.2% and 22.6% in the general population, but when only youth with overweight/obesity are considered, this prevalence increased to 60%. In Portuguese children, Maia (2010) found a metabolic syndrome prevalence of 7.2% in adolescents non-stratified by their weight status, while Braga-Tavares and Fonseca (2010), studying Portuguese obese adolescents, reported a metabolic syndrome prevalence ranging from 8.9% to 34.9%, varying according to different cut-off points. In addition, reports have also showed that physical activity and physical fitness levels play relevant roles on children's metabolic profiles - those with higher physical activity levels and/or higher physical fitness levels tend to have a better metabolic profiles than their counterparts who are less active and/or less fit, independent of their weight status (Andersen et al., 2006; Brambilla et al., 2011; Eisenmann et al., 2005; Eisenmann et al., 2007; Steele et al., 2008). So, exploring the relationship between metabolic risk factors and children behavioural traits is a pertinent venture.

- The fourth and last reason is connected with the exploration of the complex relationship between physical activity, sedentariness and BMI. It has been previously proposed that the obesogenic environment, characterized by decreases in daily physical activity, increases in time spent in sedentariness,

[1] "While the details vary, it is clear that the prevalence of the metabolic syndrome in obese pediatric patients ranges from 18 to 50%, while 1% or fewer of normal weight children have the metabolic syndrome by any definition." (Saland, 2007, p. 184)

and changes in dietary intake, are the responsible links for the high incidence of excess weight in the population, as well as for the increase in deaths by non-communicable diseases (Yusuf et al., 2001a). In addition, some research has reported that low levels of physical activity, or high levels of sedentariness, induce increases in body weight, resulting in overweight/obese children and adolescents (Epstein et al., 2000; Epstein et al., 1995; Jago et al., 2005; Liao et al., 2014; Rabbee & Betensky, 2004; Tremblay et al., 2011; Trinh et al., 2013). However, it is not clear if this relationship is direct or mediated by other behavioural or environmental variables, as suggested by Epstein et al (1995), when conjecturing that sedentary behaviours, such as watching TV, stimulate food consumption, increase energy intake and, as a consequence, increase body weight.

The relationship between physical activity and sedentariness has been explored in many different ways. Although they have been seen as two different constructs and not different sides of the same coin (Biddle et al., 2004; Katzmarzyk, 2010; Pate et al., 2011), they can co-exist in any individual (Leech et al., 2014; Marshall et al., 2002; Owen et al., 2000), although there is no consensus that by increasing one the other will necessarily decrease (Biddle et al., 2004; Marshall et al., 2002; Tammelin et al., 2007).

Considering data from the Portuguese youth population, available results are not consistent. For example, Carvalhal et al (2007), and Mota et al (2006) found an association between sedentary behaviour (namely playing games and computer use, respectively) and weight status in both boys and girls, but Mota et al (2006) were not able to find a relationship between physical activity and weight. On the other hand, Bingham et al (2013), exploring the influence and risk of socio-demographic and behavioural factors on childhood obesity, reported that performing at least 1 hour of moderate physical activity every day is one of the protective factors against childhood overweight/obesity in Portuguese children.

Therefore, a more comprehensive understanding of how BMI, physical activity, and sedentariness are interconnected is both necessary and important.

Taken together with the four reasons described above, we delineated this thesis whose main aims are (1) to describe inter-individual differences in physical activity and sedentariness levels as well as obesity in Portuguese children, and (2) to investigate their relationships with individual, familial and environmental characteristics. Further, specific aims are:

- To estimate between-school variability in children's sedentariness, identifying individual- and school-level correlates, and to determine if these correlates differ among weight status groups.

- To investigate the between- and within-individual variances in sedentariness over an entire week of objective monitoring as well as their correlates.

- To determine if there has been an increase in the prevalence of overweight/obesity in Portuguese children. In addition, to investigate differences in behavioural characteristics among normal-weight and overweight/obese children, and individual and school level correlates linked to children's BMI variation.

- To explore the role of physical activity and BMI on metabolic risk, analysing differences in risk factors across distinct BMI and physical activity groups.

- To study the joint roles of physical activity and muscular strength on metabolic risk factors in children.

- To jointly analyse physical activity and sedentariness, as a multivariate multilevel model, as well as their correlates at the child and school levels.

- To investigate the relationship between BMI and sedentariness from a multilevel perspective.

THESIS OUTLINE

The thesis comprises a collection of manuscripts that are published, accepted or submitted for publication in peer-reviewed journals. Chapter I presents the general introduction to the thesis, its aims and the overall outline; chapter II is dedicated to the study methods; chapter III comprises the published manuscripts as well as those accepted and submitted. A general overview and conclusions, as well as the limitations of this thesis, are presented on chapter IV. References are at the end of each chapter according to journal guidelines; chapters I, II and IV references were presented according to Faculty of Sports, of University of Porto, guidelines.

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Table 1. Thesis outline

Chapter I	Presents the general introduction and thesis aims.
Chapter II	Presents the methods section and describes the sample, instruments and protocols used in data collection.
Chapter III	**Paper I** **Correlates of sedentary time in children: a multilevel modelling approach** Purposes: to estimate the between-school variability in sedentary time of Portuguese children; to identify individual- and school-level correlates of sedentary time, and also to test cross-level interactions between BMI and school climate variables; and to determine if individual- and school-level correlates of sedentary time are similar among normal-weight and overweight/obese children. *Published in BMC Public Health (2014), doi:10.1186/1471-2458-14-890* Authors: Thayse Natacha Gomes; Fernanda Karina dos Santos; Daniel Santos; Sara Pereira; Raquel Chaves; Peter T. Katzmarzyk; José A. R. Maia.
	Paper II **Why are children different in their daily sedentariness? An approach based on the mixed-effects location scale model.** Purpose: to investigate the between- and within-individual variances in sedentariness over seven days of objective monitoring. *Under review in PloS One* Authors: Thayse Natacha Gomes; Donald Hedeker; Fernanda Karina dos Santos; Sara Pereira; Peter T. Katzmarzyk; José A. R. Maia.

Chapter III

Paper III

Overweight and Obesity in Portuguese Children: Prevalence and Correlates

Purposes: to conduct a meta-analysis on overweight/obesity prevalence in 9 to 11 year old Portuguese children; to detect significant differences in behavioural characteristics among normal-weight and overweight/obese children; and to investigate the importance of individual- and school-level correlates on variation in children's BMI.

Published in International Journal of Environmental Research and Public Health (2014), doi: 10.3390/ijerph111111398

Authors: Thayse Natacha Gomes; Peter T. Katzmarzyk; Fernanda Karina dos Santos; Michele Souza; Sara Pereira; José A. R. Maia.

Paper IV

"Fat-but-active": Does physical activity play a significant role in metabolic syndrome risk among children of different BMI categories?

Purpose: to explore the idea of "fat-but-active" by analysing differences in metabolic syndrome risk factors across distinct BMI and physical activity groups.

Published in Journal of Diabetes and Metabolism (2014), doi: 10.4172/2155-6156.1000421

Authors: Thayse Natacha Gomes; Fernanda Karina dos Santos; Daniel Santos; Raquel Chaves; Michele Souza; Peter T. Katzmarzyk; José A. R. Maia

Paper V

"Active and strong": physical activity, strength and metabolic risk in children

Purpose: to explore the joint roles of physical activity and muscular strength on metabolic risk factors in children.

Submitted

Authors: Thayse Natacha Gomes; Peter T. Katzmarzyk; Fernanda Karina dos Santos; José A. R. Maia.

Paper VI

Relationship between sedentariness and moderate-to-vigorous physical activity in youth. A multivariate multilevel study

Purpose: to jointly analyse moderate-to-vigorous physical activity and sedentariness as well as their correlates in children within their school contexts.

Submitted

Authors: Thayse Natacha Gomes; Donald Hedeker; Fernanda Karina dos Santos; Michele Souza; Daniel Santos; Sara Pereira; Peter T. Katzmarzyk; José A. R. Maia.

Chapter III	**<u>Paper VII</u>** **Are BMI and sedentariness correlated? A multilevel study in children** Purposes: to study the relationship between BMI and sedentariness in Portuguese children; and to investigate the importance of child and school correlates in BMI and sedentariness variation. *Under reivew in Nutrients* Authors: Thayse Natacha Gomes; Peter T. Katzmarzyk; Fenanda Karina dos Santos; Raquel Chaves; Daniel Santos; Sara Pereira; Catherine M. Champagne; Donald Hedeker; José A. R, Maia.
Chapter IV	General overview and conclusions

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CHAPTER II

Study Sample and Methods

STUDY SAMPLE AND METHODS

The International Study of Childhood Obesity, Lifestyle and the Environment – ISCOLE

The main purpose of ISCOLE, a cross-sectional study, is to "determine the relationship between lifestyle behaviours and obesity in a multi-national study of children, and to investigate the influence of higher-order characteristics such as behavioural settings, and the physical, social and policy environments, on the observed associations within and between countries" (Katzmarzyk et al., 2013, p. 2). The study hypothesizes that differences in the relationship between lifestyle behaviour and obesity will be different across countries and distinct environmental settings. It is expected that results coming from ISCOLE will provide relevant information that should be used in the development of interventions to address and prevent childhood obesity, suitable to be adapted for implementation around the world.

The ISCOLE sample comes from 12 countries (Australia, Brazil, Canada, China, Colombia, Finland, India, Kenya, Portugal, South Africa, United Kingdom, and United States of America) from five geographic regions of the world (Europe, Africa, the Americas, South-Eastern Asia, Western Pacific). In each country, a random sample of at least 500 children (final projected total sample for at least 6000 children), gender balanced, aged 9-11 years (mean age of 10 years), from urban or suburban areas, was recruited. Children should be enrolled in schools which were stratified by socioeconomic status whenever possible. By design, the ISCOLE sampling frame was not representative of each country, but maximized socioeconomic status variation at each site. The Pennington Biomedical Research Center, in Baton Rouge, USA, is the ISCOLE Coordinating Center, responsible for the overall administration the study, and in each site there is a principal investigator, who is responsible for the all aspects of study implementation at the local level.

The data collection was conducted during the school year, covering, whenever possible, different seasons, and started in September 2011. Each site should have completed their data collection across one school year (12 months).

Objective and subjective information from distinct levels were obtained, as follows:

- <u>At the child level:</u> age, anthropometry and body composition [height, weight, sitting height, waist circumference, mid-upper-arm circumference, body mass index (BMI), impedance, body fat]; somatic maturation (percentage of predicted adult stature and the maturity offset); accelerometry (physical activity, sedentary behaviour, steps count, sleep time); self-reported physical activity; outdoor time; television viewing and computer use; physical education class; active transport; motivation for and attitudes towards physical activity; food consumption (food frequency; eating in front of TV; frequency of eating breakfast; lunches at school and outside of the home; emotional eating); self-reported sleep duration and quality; self-rated health and well-being.

- <u>At the family level:</u> ethnicity of participants; family health and socioeconomic factors; family structure; education level, and self-reported height and weight of biological parents; home social environment; home food environment; home physical activity environment.

- <u>At the neighbourhood level:</u> neighbourhood social capital; neighbourhood food environment; neighbourhood physical activity environment; neighbourhood built environment.

- <u>At the school level:</u> number of students; number of days students attend school during the academic year; school facilities; healthy eating and physical activity policies; extracurricular activities; frequency of physical education and breaks (recess); amount of class time mandated for physical education; promotion/support of active transportation; availability of healthy and unhealthy foods; directly-observed information pertaining to the school built and food environment (sports and play amenities, aesthetics), and competitive food environment (food environment of the area surrounding the school).

All staff members were trained and certified by trained experts as competent to make the required measurements in regional training sessions organized by the ISCOLE Coordinating Center. The same standardized measurement protocols were used in all sites, and given that the main purpose of ISCOLE is to take into account the role of lifestyle behaviours and environmental characteristics on obesity, multilevel/random-effects models (children within schools, and schools within countries) are used in all major analyses.

ISCOLE – Portugal

In Portugal, the ISCOLE study was conducted by the Kinanthropometry Laboratory from the Faculty of Sport, University of Porto, coordinated by Prof. Dr. José António Ribeiro Maia (the ISCOLE Portugal principal investigator). The staff comprised 9 members (6 with master degree, 3 undergraduates in Physical Education and/or Sport Science). All staff members were trained and certified, and were involved full-time in the data collection, control and management of the study.

A total of 23 schools, from the North of Portugal, were enrolled in the ISCOLE project. In each school, all 5th grade students were invited to take part in the study, and those aged 9-11 years were considered eligible. From those, parental or legal guardian consent was obtained, and approximately 30 to 40 children (50% of each sex), per school, were randomly selected. The response rate was 95.7%.

Schools selection and inclusion in the project were done in a series of steps. Firstly, from a list provided by the North Regional Education Directory Board, eligible schools were selected. Since there is little variability in socioeconomic status at the school level in the Portuguese North Region, only public schools were selected; further, schools should be located in different regions and socioeconomic neighbourhoods. Secondly, selected schools were contacted and the project was presented to the Physical Education Department coordinator, and if he/she agreed with the project implementation at the school, a presentation of ISCOLE for the Physical Education Department took place (purposes of the project, strategies for data collection, feedback about the collected information, and benefits for the implementation of the project to the school community). After approval by the Physical Education Department, the project was, in this order, presented to school Principal and Pedagogical Council, and Parental Council, and should be approved from all of them to be implemented. If a non-compliant school was found (no approval by at least one of these “groups”), it was replaced by the next school selected from the list. Thirdly, signed parental or legal guardian consents were sent to all 5th grade students and, as mentioned above, from those aged 9-11 years and with signed consent form, approximately 30-40 children were selected. Fourthly, a calendar with data procedure routines was developed and sent to each school. The data collection was obtained during a whole week per school, mostly during physical education classes. Fifthly, after the data collection, each school received a report containing the major results, and for each child enrolled in the project a report was sent to his/her parents/legal guardian containing information about anthropometry, body composition, physical activity and sedentariness levels, lifestyle, nutritional habits, and also metabolic risk indicators (blood analyses and blood pressure as explained below).

Since it was of interest in Portugal to explore other research venues not comprised in the ISCOLE original project, and given that ISCOLE principal investigators were encouraged to develop ancillary studies that could enhance the scientific output of ISCOLE, information regarding children’s physical fitness levels and metabolic risk indicators [blood pressure, High Density Lipoprotein Cholesterol (HDL-C), glucose, triglycerides] were also obtained. All measurements proposed by ISCOLE original project were taken following the ISCOLE protocols (Katzmarzyk et al., 2013). The ISCOLE-Portugal and the ancillary study proposals were approved by the University of Porto ethics committee.

Sample

The sample for this thesis comprises 777 children (419 girls), aged 9-11 years, enrolled in 5th grade from 23 elementary schools from the North region of Portugal. All these 777 children were evaluated according to ISCOLE procedures.

A sub-sample of 421 children (233 girls) accepted to participate in an ancillary study to understand the relationship between physical activity, physical fitness, weight status and metabolic risk. All data were collected from September 2011 to January 2013.

Procedures

The procedures described below correspond to all measurements made in the ISCOLE-Portugal project. Not all available information were used in this thesis. In addition, all procedures followed the protocol defined by the ISCOLE Coordinating Center, except for those variables belonging to the ancillary study.

Anthropometry and body composition

Height, sitting height, weight, waist circumference, and mid-upper-arm circumference were taken according to standardized ISCOLE procedures and instrumentation (Katzmarzyk et al., 2013).

Height, sitting height and mid-upper-arm circumference were measured according to procedures described by Lohman et al (1988). For height and sitting height, children were without shoes, with heads positioned to the Frankfurt Plane, using a Seca 213 portable stadiometer rounding up to the nearest 0.1 cm (Hamburg, Germany). For height, children were fully erect, feet together, and the measurement was taken at the end of a deep inhalation, while for sitting height, children were seated on a table with legs hanging freely and arms resting on the thighs. Mid-upper-arm circumference was measured on the right arm, in the midway between the acromion and olecranon processes, with arm hanging loosely at the side of the body, using a non-elastic tape. Waist circumference measurement followed the procedures described by the World Health Organization (WHO) (2011), and were taken at the midway point between the lower rib margin and the iliac crest, at the end of a gentle expiration, using a non-elastic tape.

Body weight, impedance and body fat were measured using a portable Tanita SC-240 Body Composition Analyzer scale (Arlington Heights, IL). Children were without shoes and socks, and wearing light clothes.

All measurements were taken twice, and the average was used for analysis. A third measurement was obtained if the difference between the previous two measurements was outside the permissible range for each measurement and its replica: 0.5 cm for height, sitting height, waist circumference, and mid-upper-arm circumference; 0.5 kg for weight; and 2.0% for body fat. In this case, the closest two measurements were averaged and used for analysis.

The BMI [weight(kg)/height(m)^2], waist-to-height ratio, and sitting height-to-height ratio were computed. In addition, children were classified in their weight status using cut-points suggested by WHO (de Onis et al., 2007), International Obesity Task Force (Cole et al., 2000; Cole et al., 2007), and the US Centers for Disease Control and Prevention (Kuczmarski et al., 2002).

Biological maturation

Biological maturation was assessed by two methods: percentage of predicted adult height (Khamis & Roche, 1994), and the maturity offset (Mirwald et al., 2002).

Using information from children's chronological age, height, weight and mid-parent height (average of father's and mother's height), the final adult height of children was estimated; the closer to adult height children are, the more advanced in their somatic maturation. Regarding to maturity offset, the timing to peak height velocity (PHV) occurrence was estimated using information on sex, age, and physical growth characteristics (sitting height, leg length, height, and weight); a positive maturity offset expresses the number of years a child is beyond PHV, while a negative maturity offset means the number of years a child is before the PHV; a value of zero indicates that a child is experiencing the PHV.

Objective measured physical activity, sedentariness and sleep time

Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL) were used to objectively estimate children's physical activity, sedentariness and sleep time. Children were instructed to wear the accelerometer for at least 7 consecutive days (plus an initial familiarization day and a part of the final day), including two weekend days, 24 hours/day. Accelerometer data were divided into daytime activities and nocturnal sleep time using an automated algorithm (Barreira et al., 2014; Tudor-Locke et al., 2014). Any sequence of at least 20 consecutive minutes of zero activity counts during the awake period was considered as "non-wear time" (Barreira et al., 2014; Tudor-Locke et al., 2014). To be considered eligible, i.e. valid information, children had to have at least 4 days (with at least one weekend day) with a minimum of 10 hours of wear time per day. At the final day of data collection, accelerometers were returned to the ISCOLE staff, and the research team verified the data for completeness using the most recent version of the ActiLife software (version 5.6 or higher; ActiGraph, Pensacola, FL) available at the time. If a child was not considered eligible, ISCOLE staff could ask the child to wear the accelerometer again, for the same period or higher (up to a maximum of 14 days).

Children received the accelerometers when anthropometric measurements were taken, and all instructions were given to them and a letter to their parent or legal guardian was sent, explaining the purpose of the device, and how children should use it. At the 10th day of monitoring, children returned the accelerometer for the ISCOLE staff (which went to the school with this purpose, as previous combined with children).

Cut-points defined by Evenson et al (2008) and Treuth et al (2004), using 15 second epochs and 1 minute epochs, respectively, were used to define different activity phenotypes and sedentariness. The nocturnal sleep time was determined using a novel and fully-automated algorithm specially developed for use in ISCOLE and epidemiological studies employing a 24-hour waist-worn accelerometer protocol in children (Barreira et al., 2014; Tudor-Locke et al., 2014).

Diet and lifestyle information

A compilation of questions and measures obtained from several different sources was done and “The Diet and Lifestyle Questionnaire” was developed (Katzmarzyk et al., 2013). This questionnaire contains items related to dietary intake, physical activity, sedentary behaviour, and sleep, and was answered by all children, at school, on the same day that anthropometric measures were taken, under the supervision of at least one ISCOLE staff member, and all questionnaires were checked for completeness at the time of data collection.

Concerning nutritional habits, the questionnaire provides information regarding to children’s food frequency, asking children about several different types of food consumed in a usual week, and also the consumption of different types of snacks while watching TV, which were exemplified by individual food items (not portion size). In addition, the questionnaire also provides information regarding breakfast consumption, lunches consumed at schools, and meals consumed prepared away from home.

Subjective information about physical activity and sedentary behaviours was obtained by asking children about the amounts of time they spent in physical activity before/after school and during weekends, sedentary behaviours (namely time spent watching TV, using the computer, or playing video games, and the availability of TV in their bedroom), the number of physical education classes attended per week, and the method used to go to/from school (active or motorized).

Sleep patterns, duration and quality were also obtained, as well as psychological constructs related to physical activity and dietary behaviour, such as motivation and self-efficacy for physical activity, and emotional eating, and health-related quality of life (health and mental well-being).

Parental questionnaires

Two questionnaires were answered by parents or legal guardians: "Demographics and Family Health Questionnaire" and "Neighbourhood and Home Environment" (Katzmarzyk et al., 2013). On the day of the anthropometry measurements, and after children answered the "Diet and Lifestyle Questionnaire" and received the accelerometer, they also received an envelope containing questionnaires that should be answered by their parents or legal guardians, as well as a letter explaining to parents or legal guardians about the relevance and procedures to answer the questionnaires; further, it also explained the use of the accelerometer by their child (as mentioned before). Children were instructed to give the envelope to their parent or legal guardian and return it, with all questionnaires answered, at the day where the accelerometer should be returned. After the receipt of the questionnaires, ISCOLE staff checked for completeness and possible mistakes during answering procedures, and if the existence of blank or doubled answers was observed, phone calls (whenever possible) were done to the parent or legal guardian to solve the problem. If it was not possible to contact the parent or the legal guardian, the respective questions were assigned as "missing information".

The "Demographics and Family Health Questionnaire" considered information on basic demographics, ethnicity, child's birth date and weight, child history of breast feeding, home socioeconomic factors, biological parental height and weight.

The "Neighbourhood and Home Environment Questionnaire" includes items related to neighbourhood social capital (collective efficacy and the degree to which persons in the neighbourhood know each other and engage socially), the home social environment (parental support for and modelling child physical activity), home and neighbourhood food environments (availability of healthy or unhealthy food and drink in the home, the availability and types of food stores in the neighbourhood), the home and neighbourhood physical activity environment and neighbourhood built environment (availability of electronics for the child's personal use, access to and use of play equipment in home, access to and use of places where child can be physically active, and the suitability of the neighbourhood environment for walking and physical activity).

School environment

Two instruments were used to assess the school environment: a questionnaire (School Environment Questionnaire), answered by the physical education teacher or school principal; and a direct audit of the school environment, performed by the ISCOLE-Portugal principal investigator or the site coordinator (Katzmarzyk et al., 2013).

The "School Environment Questionnaire" provides information regarding the number of students per school, number of days that students attend school during the academic year, amount of mandatory class time for physical education, school facilities, healthy eating and physical activity policies, extracurricular activities, frequency of physical education classes and breaks (recess), promotion of active transportation, and availability of healthy and unhealthy foods in the cafeteria and vending machines. The questionnaire was delivered to the physical education teacher designed to answer it on the first day of data collection in each school, and all instruction was given. At the last day (end of the week) of data collection, the questionnaire should be delivered to one of ISCOLE staff members, who had to check the questionnaire for completeness; if any question was blank or with more than one answer, the ISCOLE staff would ask to physical education teacher to solve the problem. If it was not possible to solve the problem, for any reason, respective questions were assigned as "missing information".

The school audit, in each school, was designed to obtain directly-observed information on the school built and food environments, and whether fast food restaurants were visible from any of the school entrances (to characterize the food environment of the area surrounding the school). In addition, ISCOLE staff also obtained information about weekly class schedules from each of the classroom with students enrolled in ISCOLE, which were used to determine the amount of weekly time children spent at school and time spent in recess, breaks, and physical education classes.

Physical fitness

Measures of physical fitness components (muscular strength, agility, cardiorespiratory fitness and flexibility) were obtained by use of eight tests from two distinct batteries: AAHPERD (1980) and Fitnessgram (Cooper Institute for Aerobics Reseach, 1987; Welk & Meredith, 2008). All tests were performed according to the following protocols:

- *Handgrip strength* was measured (kg) using a digital hand dynamometer (Takei TKK 5401, Tokyo, Japan), with children in the standing position. Children squeezed the dynamometer with maximal force, maintaining it away from the body with the arm extended, using the preferred hand.
- *Standing long jump* was measured as the distance from the take-off line to the heel or other part of the body that touched the floor nearest to the take-off line. Children stood with feet apart behind the take-off line and were instructed to jump as far as possible.
- *Curl-up* required children to perform the maximum number of elevations and descents of the trunk properly, up to a limit of 75 repetitions. The number of curl-ups performed was recorded.
- *Push-up* required children performed the maximum number of push up at a rate of one every three seconds. The number of push ups performed was recorded.
- *Trunk-lift* was measured as the distance from the floor to child chin. Child should be lying on the floor, in a face down position, with toes pointed back behind the body and hands placed under the thighs; when ready, child lifts the upper body off the floor, maintaining the head in a straight alignment with the spine, for enough time for a measurement to be taken.

- *Shuttle-run* required children to run as fast as possible between two lines 10m apart. Two blocks of wood were placed behind one line distance 10m from the starting line, where is the child. Children have to run from behind the starting line to the other line, pick up one block, run back to the starting line, putting the block behind the line and repeat the process with the second block. Time was recorded in seconds.
- *50 yard dash* required children to cover the 45.7m distance running as fast as they can though the finish line, and time was recorded in seconds.
- *1-mile run/walk test* required children to cover the 1-mile distance in the shortest time as possible, running or walking, and time was recorded in seconds.

Metabolic risk indicators

Metabolic risk indicators measured were as follows: waist circumference (previously described), systolic (SBP) and diastolic (DBP) blood pressure, fasting glucose, triglycerides and HDL-C. Resting SBP and DBP were measured using a digital Omron sphygmomanometer (5 Series^{TM} Upper Arm Blood Pressure Monitor – BP742, England) after the children had been at rest for at least 10 minutes (National High Blood Pressure Education Program Working Group on High Blood Pressure in Children and Adolescents, 2004). Three measurements were taken with a 3-minute interval between successive measurements, and the mean value was used. The mean arterial pressure (MAP) was computed as: [(SBP-DBP)/3 + DBP].

Finger-stick blood samples were collected after 10-12 hours of fasting, and glucose, triglycerides and HDL-C were analysed with a Cholestech LDX point of care analyser (Cholestech Corporation, Hayward, CA, USA) (LDX C, 2003).

A standardized metabolic risk score was computed using MAP, waist circumference, glucose, triglycerides, and HDL-C, as previously advocated

(Eisenmann, 2008). All the metabolic risk indicators were previously adjusted for sex and biological maturity using a stepwise regression analysis, and the sum of the continuously distributed metabolic residuals (the HDL-C z-score was multiplied by -1) was computed to obtain the metabolic risk score. The lower the metabolic risk score is, the better the metabolic profile is (Eisenmann, 2008).

Data management and control

Except for information from physical fitness and metabolic risk, all the data entered and management were done in a secure web-based system, developed by the ISCOLE Coordinating Center and the Research Information Systems group at Wake Forest School of Medicine (Winston-Salem, NC) (Katzmarzyk et al., 2013), allowing both study site staff and the Coordinating Center to monitor progress and produce missing data reports in real time.

The data quality control was monitored by each ISCOLE site and the Coordinating Center (Katzmarzyk et al., 2013). During the entire data collection period, quality control was monitored through remote source document verification, monitoring data entry errors, and study site visits by a Coordinating Center staff member. If necessary, additional training series were provided to ISCOLE site staff to maintain quality control. Data errors and quality control issues observed during source data verification were discussed with site staff and principal investigators, and deficiencies identified were immediately corrected.

Data quality control from the information from the ancillary study was performed by training sessions for research team by experienced investigators; re-testing of a random sample (for physical fitness tests); direct observation of the author of this thesis of all the data collection; further, data entry checks and exploratory analysis for outliers were also done.

Statistical analysis

Exploratory and descriptive data analyses were performed in SPSS 20.0 or 21.0, and WinPepi version 11.26 (Abramson, 2011). T-test, and Analysis of Variance (ANOVA) and Covariance (ANCOVA) to test mean differences in the metabolic risk indicators and metabolic risk score across physical activity/weight status and physical activity/muscular strength groups, with a Bonferroni adjustment for multiple comparisons, were done in SPSS.

Since the main focus of the thesis was to explore the multilevel data structure, univariate and multivariate mixed-effects regression models, and a mixed-effects location scale model were performed in HLM 7.02 (Raudenbush et al., 2011), MIXREGLS (Hedeker & Nordgren, 2013) and SuperMix v.1 (Hedeker et al., 2008) software.

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CHAPTER III

Research Papers

Paper I

Correlates of sedentary time in children: a multilevel modelling approach

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ABSTRACT

Background: Sedentary behaviour (SB) has been implicated as a potential risk factor for chronic disease. Since children spend most of their awake time in schools, this study aimed to identify individual- and school-level correlates of sedentary time using a multilevel approach, and to determine if these correlates have a similar effect in normal-weight (NW) and overweight/obese (O/O) children.

Methods: Sample comprised 686 Portuguese children aged 9-11 years from 23 schools that took part in the ISCOLE project. Actigraph GT3X+ accelerometers were used 24 hours/day for 7 days to assess sedentary time (daily minutes <100 counts/min); BMI was computed and WHO cut-points were used to classify subjects as NW or O/O. Sex, BMI, number of siblings, family income, computer use on school days, and sleep time on school days were used as individual-level correlates. At the school level, school size (number of students), percentage of students involved in sports or physical activity (PA) clubs, school promotion of active transportation, and students' access to equipment outside school hours were used. All multilevel modelling analysis was done in SPSS, WINPEPI, and HLM.

Results: School-level correlates explain ≈ 6.0% of the total variance in sedentary time. Results (β ± SE) showed that boys (-30.85 ± 5.23), children with more siblings (-8.56 ± 2.71) and those who sleep more (-17.78 ± 3.06) were less sedentary, while children with higher family income were more sedentary (4.32 ± 1.68). At the school level, no variable was significantly correlated with sedentary time. Among weight groups, variables related to sedentary time in NW were sex, sleep time and family income, while in O/O sex, number of siblings and sleep time were significant correlates. No school-level predictors were significantly associated in either of the weight groups.

Conclusion: Notwithstanding the relevance of the school environment in the reduction of children's sedentary time, individual and family characteristics played a more relevant role than the school context in this study.

Keywords: sedentary behaviour; children; school; multilevel modelling.

BACKGROUND

Sedentariness is emerging as a potential risk factor for chronic disease [1-6]. For example, among adults, positive associations between sedentary behaviour (SB) such as sitting time and television viewing, and cardiovascular disease and adverse metabolic profiles have been reported [1-4]. In children, the link is also consistent between SB and increased prevalence of overweight/obesity [5], and an increase in metabolic risk factors [6]. Furthermore, systematic reviews have shown that screen time and overall sedentary time (objectively measured) track moderately during childhood and adolescence [7,8], which means that reducing their sedentary time may be a way to induce health benefits into adulthood [9].

Understanding the correlates of sedentary time may aid in developing preventive strategies [10]. Sedentary time may be best represented by a construct that is different from physical activity (PA) [11,12]; however, their determinants might be similar [11,13]. Recently, it has been proposed that ecological approaches may provide a sound basis for a better understanding of sedentary time [14]. These approaches examine interactions between the subject and multiple levels of influence across intrapersonal (biological, psychological), interpersonal (social, cultural), organizational, physical environment (built, natural), and policy (laws, rules, regulations, codes) domains [10]. As such, factors that influence sedentary time in children could be different in home, neighbourhood and school settings, emphasising the necessity to understand the setting-specific multilevel factors that influence this complex behaviour.

Since children spend considerable time at school, this multifaceted environment could be an important venue for reducing their sedentary time. The school social and physical environments provide potential opportunities for children to avoid extended periods of sedentary time such as active transportation to and from school, large campus size or playground areas, sports equipment and sporting facilities, recess periods, lunch breaks, and physical education classes [15-19]. However, children spend most of their school time in sedentary activities [20]. The examination of school correlates of sedentary time among children, attempting to scrutinise the influence of factors coming from multiple levels, is not abundant in the literature [21,22].

Given that students are influenced by shared and unique characteristics within and between schools, the correlates of sedentary time are ideally investigated using multilevel modelling [23]. Multilevel modelling analysis allows for the simultaneous examination of the effects of school- and individual-level predictors; accounts for the non-independence of observations within schools; does not treat subjects and school environment as unrelated, but they are seen as coming from a larger population; and examines both inter-individual and inter-school variation (as well as the contributions of school- and individual-level variables to these variations), allowing the investigation of individual and school contexts simultaneously [24-26].

The purposes of this study were to (1) estimate the between-school variability in sedentary time of Portuguese children, (2) identify individual- and school-level correlates of sedentary time, and also test cross-level interactions between BMI and school climate variables, and (3) determine if individual- and school-level correlates of sedentary time are similar among normal-weight (NW) and overweight/obese (O/O) children.

METHODS

Sample

A two-level random cluster sample of 777 5th grade Portuguese children (419 girls, 358 boys) from 23 schools, aged 9-11 years old, was assessed. After exclusion criteria (non-valid accelerometer data), the final sample comprises 686 children (381 girls, 305 boys). The students were part of the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE), a research project conducted at sites in 12 countries from all major world regions. In short, ISCOLE aims to determine the relationship between lifestyle behaviours and obesity in a multi-national study of children, and to investigate the influence of higher-order characteristics such as behavioural settings, and the physical, social and policy environments, on the observed relationships within and between countries [27].

After a first initial contact with a physical education teacher from each school, the project was presented to the physical education department. Following their approval, the project was then presented to the school principal as well as to the parental council; it was only after obtaining these consents that the project was implemented in each school. All 5th grade children were invited to be part of the ISCOLE; however, only children aged between 9 and 11 years old were classified as “eligible” to be part at the project. From those “eligible” children, a sample of ≈ 30-40 children per school was randomly selected (50% for each gender). Non-response was negligible (response rate was 95.7%), and missing information was at random, since differences between subjects with missing information and those included in the present study were not statistically significant (data not shown).

Data were collected from September 2011 to January 2013. All assessments were done during a full week per school. The study protocol was approved by the University of Porto ethics committee, as well as by the schools' directorate councils. Written informed consent was obtained from parents or legal guardians of all children. All data collection and management activities were performed and monitored under rigorous quality control procedures, implemented by the ISCOLE Coordinating Center, as described in detail by Katzmarzyk et al. [27].

Anthropometry

Height and weight measures were obtained according to standardized ISCOLE procedures [27]. Each child was measured twice and, when necessary, a third measurement was taken if the difference between the previous two was outside the permissible range for each measure and its replica (0.5 cm for height and 0.5 kg for weight). The mean value of each measured variable was used for analysis.

Body mass index (BMI) was calculated using the standard formula [weight(kg)/height(m)^2], and subjects were classified as normal-weight, overweight, or obese according to the cut-off points from the World Health

Organization (WHO) [28]. In the present paper, and to pursue our second aim, two BMI groups were formed: NW group, and O/O group. Since the number of children classified as underweight was very low (8 cases), they were included in the NW group.

Family data

Family information was obtained by a questionnaire completed by parents or legal guardians [see ISCOLE Demographic and Family Health Questionnaire in Katzmarzyk et al [27]]. The questionnaire collected information on basic demographics, ethnicity, family health and socioeconomic factors. For the present study, we only use information on family income [as an indicator of socioeconomic status (SES)] and number of siblings.

Subjects were classified into one of eight categories of annual family income, ranging from < €6000 to ≥ €42000, where category 1 represents lowest family income, and category 8 represents the highest. In the analysis strategy used in this paper, the family income socioeconomic variable was centered at category 4. Parents were also asked about family size, i.e., number of siblings.

Sleep and sedentary time

Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL) were used to monitor sleep and sedentary time. Children wore the accelerometer at their waist on an elasticized belt, placed on the right mid-axillary line 24 hours/day, for 7 days, including 2 weekend days. To be eligible for this analysis, children had at least 4 days with a minimum of 10 hours of wear time per day; a total of 686 children fulfilled this condition.

Accelerometer data were first divided into awake time and nocturnal sleep time using an automated algorithm [29,30]. After exclusion of the nocturnal sleep episode time, waking non-wear time was defined as any sequence of at least 20 consecutive minutes of zero activity counts [30].

Sedentariness is a multi-faceted characteristic that includes behaviour at work/school, at home, during transport, and in leisure-time including screen-time, motorized transportation, and sitting (to read, talk, do homework, or listen to music) [31]. In the present study, sedentary time was defined as minutes/day spent at less than 100 counts/min (using 1 minute epochs) as advocated by Treuth et al [32]. Further, information was also collected about children's SB, by asking them about time spent in computer use during school days [ISCOLE Diet and Lifestyle Questionnaire, described in Katzmarzyk et al [27]].

School environment

Information concerning the school environment (context and climate) was obtained via a questionnaire [ISCOLE School Environment Questionnaire presented in Katzmarzyk et al. [27]] which was completed by the physical education teacher or the school principal. The questionnaire includes items related to school facilities, healthy eating and PA policies, extracurricular activities, frequency of physical education and breaks (recess), promotion of active transportation, availability of healthy and unhealthy foods in the cafeteria and vending machines, number of days that students attend school during the academic year, and the amount of class time mandated for physical education. For the present study we considered primarily the (i) school context information regarding school size (number of students), and (ii) school climate which includes percentage of students participating in school sports or PA clubs, school promotion of active transportation (allowing children to bring their bicycles), and students' access to sports equipment outside of school time. These variables were chosen firstly because there is evidence that they are correlated with PA and sedentariness in school children; and secondly, because of the multilevel data structure.

Data analysis

Descriptive statistics, t and chi-square tests were computed in IBM SPSS version 20.0, and WinPepi version 11.26 [33]. Modelling the relationship between children's sedentariness, their individual characteristics (level-1), and school environmental factors (level-2) was done in HLM 7.02 software within the framework of the multilevel approach using maximum likelihood estimation procedures [34].

A series of hierarchical nested models were fitted, and the Deviance statistic was used as a measurement of global fit. It is expected that as models increase in complexity by adding predictor variables, a significant decrease in Deviance is expected to occur, and the significance of the decrease is tested with a chi-square test [35]. In addition, the relevancy of predictors to explain sedentary behaviour was assessed with a pseudo-R^2 statistic which is interpreted as a proportional reduction in variance for the parameter estimate that results from the use of one model as compared to a previous one [34]. Modelling was done in a “stepwise” fashion as generally advocated [see, for example, Hox [35], and Snijders and Bosker [36]]. Firstly, a null model (M0) was fitted to the data to compute the intraclass correlation coefficient to estimate the variance accounted for by school effects in sedentariness. Secondly, Model 1 (M1) was fitted to the data using only children predictors of sedentariness (gender, BMI, number of siblings, family income, computer use, and sleep time). BMI and sleep time were centred at the grand mean. Thirdly, Model 2 (M2) was fitted by adding all school predictors and cross-level interactions. This analysis was firstly done using the total sample (i.e., all subjects), and then repeated using the two sub-samples based on WHO cut-offs for BMI (normal-weight and overweight/obese).

RESULTS

Tables 1 and 2 show descriptive statistics (Mean ± SD and percentages) for level 1 and level 2 variables. Boys and girls had similar ($p > 0.05$) heights, weights, BMI, number of siblings, and mean sleep time. Also, no differences were found in overweight prevalence among genders ($\chi^2 = 0.772$, $p = 0.380$), but obesity had a higher frequency in boys ($\chi^2 = 9.895$, $p = 0.002$). Girls had higher sedentary time than boys ($t = 6.085$, $p < 0.001$).

Table 1. Descriptive statistics for variables at the child level (level-1)

Child-level variables (mean ± SD)				
	Boys (N = 305)	Girls (N = 381)	*t*	*p*-value
Height (cm)	143.46 ± 6.42	143.49 ± 7.06	0.060	0.952
Weight (kg)	40.52 ± 9.23	40.28 ± 9.23	-0.332	0.740
BMI (kg/m^2)	19.54 ± 3.45	19.41 ± 3.36	-0.511	0.610
Number of siblings	0.97 ± 0.80	0.95 ± 0.83	-0.230	0.818
Sedentary time	449.73 ± 73.07	482.21 ± 66.45	6.085	<0.001
Sleep time (hours/day)	8.14 ± 1.02	8.21 ± 0.96	0.964	0.335
		BMI classification (%)	χ^2	*p*-value
Normal-weight	49.5%	58.0%	4.920	0.026
Overweight	15.1%	17.6%	0.772	0.380
Obese	35.4%	24.4%	9.895	0.002
	Annual Family Income			
Category 1	14.1%	22.7%		
Category 2	33.2%	29.3%		
Category 3	21.2%	16.7%		
Category 4	11.6%	9.3%		
Category 5	7.1%	7.0%		
Category 6	4.6%	6.0%		
Category 7	2.9%	3.7%		
Category 8	5.4%	5.3%		
	Computer use on school days			
Did not use	36.4%	46.2%		
<1 hour	23.3%	29.4%		
1 hour	21.0%	16.8%		
2 hours	12.8%	5.2%		
3 hours	3.3%	1.8%		
4 hours	1.6%	0.3%		
5 or more hours	1.6%	0.3%		

Table 2. Descriptive statistics for variables at the school level (level-2)

School-level variables	
Number of students (mean ± SD)	782 ± 309
Children participation in sports or PA clubs	
Not available	4.3%
Less than 10%	4.3%
10-24%	34.8%
25-49%	13%
≥50%	43.5%
Promoting active transportation (bike)	
No	21.7%
Yes	78.3%
Student’s access to equipment outside school hours	
No	47.8%
Yes	52.2%

More than 90% of the schools have children engaged in sports participation or PA clubs, more than 75% of them promote active transportation among their students, and about 50% of them allow the students to have access to sports equipment outside of school hours. The mean number of students per school is 782 ± 309, ranging from 239 to 1589.

Results of the null model, as well as for the other two models from the full sample, are presented in Table 3. Estimated variance at the school level suggests significant inter-individual differences across schools in sedentary time ($\chi^2 = 67.32$, $p < 0.001$). The estimated school-level effects from the intraclass correlation coefficient was 0.0609, meaning that ≈ 6.0% of the total variance in sedentary time among all children is explained by school effects, and 94% is explained by children's distinct characteristics at their individual level. Also, the reliability estimate of 0.65 is an indicator of how well each school sample mean estimates the overall schools mean sedentary time parameter.

Results from M1 related to individual-level predictors show that the sedentary time mean for a girl with a mean age of 10.5 years is 484 minutes·day^{-1}. Boys, children with more siblings and those who sleep more are less sedentary, i.e. spend less time in sedentary activities ($p < 0.05$), but those with higher family income tend to be more sedentary ($p = 0.013$). No statistically significant associations were found for BMI and time spent using a computer on school days in mean sedentary time ($p > 0.05$). The reduction in the variance component at the children's level allowed the estimation of the proportion (34.4%) of children's characteristics explaining the inter-individual variance in sedentary time.

Table 3. Results summary of hierarchical linear modelling for all sample: estimates, standard-errors, and p-values

Parameters	Null Model			Model 1			Model 2		
	Estimates	*Standard Error*	*p-value*	*Estimates*	*Standard Error*	*p-value*	*Estimates*	*Standard Error*	*p-value*
Intercept	467.02	4.53	<0.001	484.46	5.67	<0.001	491.70	13.05	<0.001
Sex				-30.44	4.97	<0.001	-30.85	5.23	<0.001
BMI				-0.22	0.69	0.752	1.06	1.85	0.566
BMI X Participation in sports or PA clubs							-0.14	0.65	0.829
BMI X Promoting active transport							-1.75	1.74	0.316
BMI X Access to equipment outside school hours							0.74	1.67	0.656
Number of siblings				-8.50	2.67	0.002	-8.56	2.71	0.002
Family income				4.24	1.70	0.013	4.32	1.68	0.010
Computer using on school days				2.68	3.07	0.383	2.62	3.10	0.399
Sleep time				-17.90	2.96	<0.001	-17.78	3.06	<0.001
School size							-0.001	13.05	0.910
Participation in sports or PA clubs							-1.81	3.78	0.637
Promoting active transport							-2.33	8.53	0.787
Variance components: random effects									
School mean		309.50			202.89			190.05	
Children level effect		4765.57			3854.52			3852.97	
Model summary									
Deviance statistic		7781.240599			5551.69			5550.73	
Number of estimated parameters		3			9			15	

The final model, M2, investigated school effects as well as cross-level interactions. In this model, we assumed that the intercept parameter (sedentary time) varies at level 2. The mean sedentary time of a girl from a school where students are not involved in sports or PA clubs, and do not promote active transportation to school is 492 minutes·day^{-1}. No significant associations were found for school size, percentage of students engaged in sports or PA clubs, or school promotion of active transportation. Similarly, cross-level interactions between BMI and school climate variables tested did not show any significant interaction.

Table 4 shows the results for the two weight groups (NW and O/O). Since BMI was used to classify subjects in weight groups, this variable was excluded in these analyses, as well the cross-level interactions between BMI and school climate variables. Among NW children, significant associations were found for sex, sleep time and family income, where boys and children who sleep more are less sedentary ($p < 0.05$); those with higher family income have higher sedentary time ($p = 0.008$). For O/O, being a boy, children with more siblings and those who sleep more have a significantly lower mean sedentary time ($p < 0.001$). Similar to the overall sample, no significant associations were found between sedentary time and school variables in NW and O/O groups.

Table 4. Summary of results of final model for two BMI groups (normal-weight and overweight/obese groups): estimates (standard-errors), and p-values

Parameters	**Normal-weight (N = 340)**			**Overweight/Obese (N = 272)**		
Regression coefficients: fixed effects	***Estimates***	***Standard Error***	***p-value***	***Estimates***	***Standard Error***	***p-value***
Intercept	483.23	13.35	<0.001	514.03	19.96	<0.001
Sex	-32.93	8.45	<0.001	-29.12	7.33	<0.001
Number of siblings	-4.25	4.48	0.344	-10.25	5.18	0.049
Family income	6.29	2.34	0.008	2.59	2.58	0.317
Computer using on school days	1.63	3.52	0.643	5.44	4.44	0.222
Sleep time	-25.85	4.45	<0.001	-9.52	3.04	0.002
School size	-0.02	0.01	0.105	0.01	0.01	0.502
Participation in sports or PA clubs	-0.18	4.56	0.969	-6.20	5.75	0.294
Promoting active transport	7.67	8.85	0.397	-20.29	13.43	0.147
Variance components: random effects						
School mean		8.75			389.16	
Children level effect		4043.96			3492.64	
Model summary						
Deviance statistic		2919.962555			2223.24	
Number of estimated parameters		11			11	

DISCUSSION

This study aimed to identify the magnitude of child- and school-level correlates of sedentary time and to determine if their importance was similar in NW and O/O children using a multilevel modelling approach.

At the child level, most of the variables included in the model were significantly linked to sedentary time. Sex differences in sedentary time are well documented [37], showing that girls spend more time in sedentary activities [11], which was confirmed in the present study. Van Stralen et al [38] studied the time devoted to sedentary activities at school in children aged 10-12 years from five European countries, and reported that girls spent a significant larger amount of school-time in sedentary activities (67%) than boys (63%, $p < 0.0001$), which can be related to differences in sex options for engagement in activities during recess time, with boys engaging more in competitive games while girls prefer socialising with friends [39]. Similarly, Verloigne et al [40] also found that girls spend more time in sedentary activities (511 minutes·day^{-1}) than boys (478 minutes·day^{-1}) taking into account the whole day, not only school time. Since in the present study children were monitored 24 hours·day^{-1}, the sedentary time variable represents the entire day, not just sedentary time while at school. As such, in association with the explanation for the sex differences in sedentary time during school hours, it is also possible that these differences may be potentiated by dissimilarities in boys' and girls' leisure time activities. Since boys tend to devote more time in PA and/or in sports participation [41] during their leisure time, this behaviour may be relevant to decrease their sedentary time.

The influence of siblings on children's sedentariness is not clear. It has also been reported, in a longitudinal study, that children with more siblings exhibit smaller increases in objectively measured sedentary time [42]. On the other hand, Verligne et al [43] investigated the effect of an intervention program on 10-12 year old Belgian children's total sedentary time, and reported that those with one or more siblings were less likely to reduce sedentary time after the intervention program. Further, Tandon et al [44] reported that children watched TV/DVD's with siblings more days per week, on average, than they did

PA's, reinforcing a potentially positive influence of the sibling for sedentary behaviour. On the other hand, it was suggested that the presence of more children at home (i.e., more siblings) is highly related with more moderate-to-vigorous PA overall and at home, and more sedentary time at home but less screen time [45]. We found a negative association between number of siblings and sedentary time in children, implying that the more siblings children have, the less sedentary they are. Since at this age there is a high peer influence in children behaviour [41], it is possible that those with less sedentary siblings tend to also become less sedentary.

Sleep time was negatively associated with sedentary time, indicating that children that slept more spent less time in sedentary activities. Several studies have shown that SB may interfere with sleep [46-48], but the results are not conclusive. For example, Belgium students who spent more time in sedentary activities, such as watching TV, playing video games, and using the internet went to bed later, spending less time in bed on weekdays [47]. However, in Taiwanese adolescents [49] no association was found between the time they spent watching TV or using a computer and getting sufficient sleep.

A positive association was found between family income and sedentary time, although the results from other studies have not always been clear about the magnitude and direction of this association [11]. For example, Olds et al [50] studied the socio-demographic correlates of SB in children aged 9-16 years, and found that children from higher SES reported greater engagement in non-screen sedentary time (such as sitting or lying down), but those from lower SES spent more time in screen-based sedentary time (watching TV, playing videogames, using computer), and no significant difference across income bands was found for total sedentary time (sum of non-screen sedentary time and screen sedentary time). Similarly, Foley et al [51] reported that 10-18 years old adolescents from areas of lower deprivation (i.e., higher SES) tended to accumulate more total sedentary time, which was determined by the concomitant use of an accelerometer and a recall diary. Furthermore, Klitsie et al [52], also using an objective and subjective method to access sedentariness, reported that 9-10 year old children with higher SES spent more time in non-screen SB; however, those from low SES and those from high SES both had higher sedentary time than those of medium SES. Using an objective method to measure sedentary time, namely accelerometers, Steele et al [53] did not find any difference in sedentary time according to SES, while Atkin et al [42] reported an increase in sedentary time, after a one-year period, among children from higher SES. Our findings add to this body of evidence, and suggest that Portuguese children with higher family income have greater sedentary time than those with low family income.

There is some prior evidence that children with a higher BMI are more sedentary, spending more time watching TV [54,55]. However, in the present study no significant association was found between sedentary time and BMI. Further, the interaction between BMI and school climate variable did not reveal a mediation effect of school characteristics on the role of BMI on sedentary time. However, TV watching was not specifically measured in the present study, and the relationship with BMI may differ across different sedentary behaviours.

Schools offer extracurricular activities and policies that could potentially reduce sedentary time among students [15-19]. In this study, only 6.0% of the total variance in sedentary time was explained by school-level variables. It is known that schools with a larger campus size or playground areas provide more opportunities for their students to engage in PA during recess time, potentially decreasing their sedentary time [15,17,19]. In addition, athletic facilities such as school sports or PA clubs appear to be good opportunities to decrease sedentary time and increase PA in youth [56]. Moreover, active commuting to school is associated with higher PA levels among youth [57,58], and children who drive to/from school are less likely to achieve recommended levels of daily PA [59]. However, despite the suggestion that school context has the potential to reduce children’s sedentary time, in the present study we did not find such an association. Our study was potentially under-powered to identify school level effects, given the sample size of only 23 schools (versus a sample size of 686 children for individual-level correlates). Further, there was limited variance in some of the school-level variables measured in this study (i.e. more than 90% of the schools have children engaged in sports participation or PA clubs). Thus, a study with a larger sample size of school, and with greater variability among schools in the environmental variables, may be better suited to detect school-level correlates.

When the analyses was stratified by body weight status, sex and sleep time were related to sedentary time in both NW and O/O groups; family income was only related to sedentary time in the NW group, while number of siblings was related to sedentary time in O/O; further, no school-level predictor was significantly associated with sedentary time in either group. Differences in individual-level sedentariness correlates among weight groups suggests that attention should be paid to weight status when implementing strategies to decrease sedentary time in children, such that the chosen activities should be easily and playfully performed by both NW and O/O children; additionally, body weight should not be a barrier to those children with higher weight.

This study has several limitations and strengths. Firstly, as we did not study distinct SB's (screen time, reading, listening to music, transportation to/from school, etc.), rather we focused on objectively determined overall sedentary time. Thus, it was not always possible to compare our results with previous studies that did not assess sedentary time objectively using accelerometry [11,60]. Secondly, the present sample comes from only one Portuguese region and its results do not necessarily generalize to all children. However, a comparison of the present sample characteristics with information available from the Portuguese population of the same age and gender was done. For example, in data not shown here, no differences were found in the prevalence of overweight/obesity [61], in the percentage of children attaining sufficient levels of PA [62], and SES distribution [63]. Thirdly, despite the evidence that moderate-to-vigorous PA attenuates the association between SB and health risk [64], we did not include this information as a covariate. Notwithstanding these limitations, the study has several important strengths: (1) the use of an objective method to estimate sedentary time; (2) the use of the accelerometer for 7 days; (3) inclusion of objective information regarding sleep time; (4) using standard methods and highly reliable data, and (5) the use of multilevel modelling to capture the complexity of nested information available at the child and school levels.

CONCLUSIONS

In summary, this study investigated the role of individual- and school-level variables with children's sedentary time within the multilevel modelling framework. School context explained 6.0% of the total variance in children's sedentary time. At the individual level, sex, number of siblings, family income and sleep time explained 34.4% of the 94% of the variance fraction of the individual level. No significant association was found between sedentary time and BMI, as well as between sedentary time and school-level correlates. Notwithstanding the relevancy of school diversified environments to reduce sedentary time in children, enhancing their opportunities for being less sedentary in their awake time, requires further analysis with a more diversified list of markers than those explored in the present study. Furthermore, differences in sedentary time correlates among NW and O/O children suggest that different strategies may be needed to reduce sedentary time in these two groups. Moreover, given the association between sedentary time and health risks, future studies should be conducted using direct measures of total sedentary time, distinguishing different types of SB and examining different patterns in which sedentary time is accumulated. Furthermore, the use of an inclinometer, in association with the accelerometer, could be useful to provide information regarding postural changes. In addition, since sedentariness and PA are two distinct phenotypes, and being physically active does not imply being less sedentary, future studies should also investigate the relationship between these two variables on health risk factors, independently and in association.

ABBREVIATIONS

SB, Sedentary behaviour; PA, Physical activity; NW, Normal-weight; O/O, Overweight/obese; ISCOLE, International study of childhood obesity, lifestyle and the environment; BMI, Body mass index; WHO, World health organization; SES, Socioeconomic status.

COMPETING INTERESTS

The authors declare that they have no competing interests.

AUTHOR CONTRIBUTIONS

TNG collected the data, undertook the data analysis and interpretation, and led the writing of the article. FKS and DS collected the data and contributed to drafting the paper. SP and RC collected the data. PTK conceptualized and designed the study and contributed to drafting the paper. JM organized and supervised data collection and management, and contributed to drafting the paper. All authors read and approved the final manuscript.

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Paper II

Why are children different in their daily sedentariness? An approach based on the mixed-effects location scale model

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ABSTRACT

This study aimed to investigate the between- and within-individual variability in sedentary time over seven days, using a mixed-effects location scale model. The sample comprised 686 Portuguese children (381 girls) aged 9-11 years, from 23 schools. Sedentary time was estimated by the Actigraph GT3X+ accelerometer, which was used 24 hours/day for 7 consecutive days; height, sitting height, and weight were measured, BMI was computed (WHO cut-points were used to classify subjects as normal-weight or overweight/obese), and maturity offset was estimated. Information regarding the home environment was obtained by questionnaire. Results revealed that: (i) children were more sedentary on Friday, but less so on Saturday and Sunday (compared to Monday), with significant variation between- and within-subjects (between-subject variance=0.800, within-subject variance=1.793, intra-subject correlation=0.308); (ii) there is a sex effect on sedentariness, with boys being less sedentary than girls (p<0.001), and the between-subject variance was 1.48 times larger for boys than girls; (iii) in terms of the within-subject variance, or erraticism, Tuesday, Wednesday and Friday have similar erraticism levels as Monday (Thursday has less, while Saturday and Sunday have more); in addition, girls (variance ratio=0.632, p<0.001), overweight/obese children (variance ratio=0.861, p=0.019), and those later mature (variance ratio=0.849, p=0.013) have less erraticism than their counterparts; (iv) the within-subject variance varied significantly across subjects (scale std dev=0.342±0.037, p<0.001); and (v) in the fixed part of the model, only biological maturation was positively related to sedentariness. This study demonstrated that there is significant between- and within-subject variability in sedentariness across a whole week. This implies that a focus on intra-individual variability, instead of only on mean values, would provide relevant information towards a more complete map of children's sedentary behaviour, which can be helpful when developing more efficient strategies to reduce sedentariness.

Keywords: sedentariness; children; Portugal; ISCOLE; mixed-effects location scale model.

INTRODUCTION

The last years witnessed an augmented interest in monitoring and understanding sedentary behaviours [1], their correlates [2], and their relationships with health hazards and reduced quality of life [3,4]. There is now compelling evidence that children and adolescents spend a large proportion of their day in sedentary behaviours [5]. However, sedentary behaviour differs among youth according to their intrapersonal traits and interpersonal characteristics, as well as built and physical environmental factors [5,6,7]. For example, several studies have identified distinct clusters of youth based on their levels and patterns of sedentary behaviour alongside their physical activity levels [1,8,9]. Further, sex [10], age [11], and maturity status [12] have also been identified as correlates of sedentariness.

Notwithstanding the intensified interest in having a more comprehensive understanding of patterns and correlates of sedentary behaviour [5,10,13,14], the available research has focused largely on mean differences [15], used sets of covariates in multiple regression models to predict sedentariness [2], or studied contextual inter-individual differences using multilevel models [16,17,18]. To our knowledge, exploring factors related to intra-individual variability in sedentary behaviour across days, which can help to better understand observed between-subject differences and effectiveness of interventions beyond mean changes, has never been addressed.

Studying variability in intra-individual differences in daily behaviours is relevant to provide an understanding of patterns of sedentary behaviour over time [19,20]. Further, seven-day objective monitoring is an acceptable window to study different physical activity and sedentary behaviour expressions [21]. Thus, the purpose of the current study is to investigate the between- and within-individual variances in sedentariness over seven days of objective monitoring, in order to answer the following questions: (i) Is there a trend in children's sedentary behaviour over an entire week? (ii) Is this trend similar in boys and girls? (iii) Is there appreciable variability in sedentary time across days? (iv) Does variability in sedentary behaviour differ for each subject or is its magnitude similar for all children? (v) Which variables are associated with variability in sedentary behaviour? To answer these questions, we used a mixed-effects location scale model [22,23] which allows both the mean and variance structures to be modelled in terms of covariates.

METHODS

Sample

The sample of this study is part of the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE), a research project conducted in 12 countries from all major world regions. In short, ISCOLE aims to determine the relationship between lifestyle behaviours and obesity in a multi-national study of children, and to investigate the influence of higher-order characteristics such as behavioural settings, and the physical, social and policy environments, on the observed relationship within and between countries [24]. Since the purpose of ISCOLE was to study children with a mean age of 10 years, ranging from 9 to 11 years, our sample recruitment was only done in 5th grade students. A total of 777 5th grade Portuguese children (419 girls), aged 9-11 years, were assessed, and after the inclusion criteria (accelerometer valid data for at least 4 days, as described below), the final sample comprised 686 children (381 girls). These students belong to 23 schools from the metropolitan area of Porto, North of Portugal, which were selected from a list provided by the North Regional Education Directory Board, taking account their location (schools should be located in different socioeconomic neighbourhoods).

After a first initial contact with a physical education teacher from each school, the project was presented to the physical education department. Following their approval, the project was then presented to the school principal as well as to the parental council; it was only after obtaining these approvals that the project was implemented in each school. All 5th grade children were invited to be part of ISCOLE; however, only children aged between 9 and 11 years were classified as “eligible” to be part at the project. From those “eligible” children, a sample of ≈30-40 children per school was randomly selected (50% for each sex). Non-response was negligible (response rate was 95.7%).

Data were collected from September 2011 to January 2013. All assessments were done during a full week per school by trained personnel from the Kinanthropometry Laboratory of the Faculty of Sport (University of Porto) following certification from the ISCOLE Coordinating Center; the questionnaires were answered by each child, at their school, after anthropometric measures were taken, and under the supervision of at least one ISCOLE staff member. The study protocol was approved by the University of Porto ethics committee, as well as by the schools' directorate councils. Written informed consent was obtained from parents or legal guardians of all children. All data collection and management activities were performed and monitored under rigorous quality control procedures, implemented by the ISCOLE Coordinating Center, as previously described in detail [24].

Anthropometry

Height, sitting height, and weight measures were taken according to standardized ISCOLE procedures [24]. For height and sitting height, children were measured without shoes, with head positioned in the Frankfurt Plane, using a portable stadiometer (Seca 213, Hamburg, Germany); height was measured with children fully erect, feet together, and at the end of a deep inhalation, while sitting height was measured with children seated on a table with legs hanging freely and arms resting on the things. Leg length was computed by subtracting sitting height from standing height. Weight was determined using a portable Tanita SC-240 body composition analyzer (Arlington Heights, IL), with children wearing light clothes and without shoes or socks. Each child was measured twice and, when necessary, a third measurement was taken if the difference between the previous two was outside the permissible range for each measure and its replica (0.5 cm for height and sitting height; 0.5 kg for weight). The mean value of each measured variable was used for analysis.

Body mass index (BMI) was calculated using the standard formula [weight(kg)/height(m)^2], and subjects were classified into two groups (normal-weight and overweight/obese) according to the cut-off points from the World Health Organization (WHO), based on BMI z-scores (normal-weight: <+1SD; overweight/obese; ≥+1SD) [25].

Family data

Family information was obtained by a questionnaire completed by parents or legal guardians [see ISCOLE Demographic and Family Health Questionnaire [24]]. The questionnaire collected information on basic demographics, ethnicity, family health and socioeconomic factors, and was answered by parents/legal guardians during the same week their children were assessed at school. For the present study, we used information about media availability in the child's bedroom. Media availability in the child's bedroom was determined by asking parents if children had a computer or video game in their bedroom. The existence of TV in the child's bedroom was informed by the children. Using information regarding media availability in the child's bedroom (TV, computer or game), a "media bedroom" variable was computed to determine if there is, at least, one media available at children's bedroom; so subjects were classified as "having media in bedroom" or "not having media in the bedroom".

Sedentary time and sedentary behaviour

Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL) were used to monitor sedentary time. Children wore the accelerometer at their waist on an elasticized belt, placed on the right mid-axillary line 24 hours/day, for at least 7 days, including 2 weekend days. To be eligible for this analysis, children had at least 4 days (from which at least one of them should be a weekend day) with a minimum of 10 hours of wear time per day; 686 children fulfilled this condition. Accelerometer information was divided into daytime activities and nocturnal sleep time using an automated algorithm, and any sequence of at least 20 consecutives minutes of zero activity counts during awake period was considered as non-wear time [26,27]

Sedentariness is a multi-faceted characteristic that includes behaviour at work/school, at home, during transport, and in leisure-time including screen-time, motorized transportation, and sitting (to read, talk, do homework, or listen to music) [28]. In the present study, sedentary time objectively measured by the accelerometer is our primary dependent variable, and is defined as equal to or less than 25 counts/15 seconds as advocated by Evenson et al. [29]. Further, information was also collected about children's sedentary behaviour, by asking them about time spent watching TV during school days [ISCOLE Diet and Lifestyle Questionnaire [24]], and they were classified as ≤2hours/day or >2hours/day.

Biological maturation

Using information on sex, age, and physical growth characteristics (sitting height, leg length, stature and body mass), an estimate of biological maturity, namely somatic maturation, was obtained using the Mirwald et al. [30] maturity offset method. This method estimates, in decimal years, the status of the child relative to their age at peak height velocity (PHV) occurrence. A positive maturity offset expresses the number of years a child is beyond PHV; a negative maturity offset indicates the number of years before PHV.

Data analysis

All exploratory data analysis and descriptive statistics, as well t-tests, were done in SPSS 20, and Excel was used to plot differences in sedentariness trajectories and patterns of two boys and girls with similar mean sedentariness values. The specifics of the mixed-effects location scale model has been described elsewhere in great detail [22,23]. Briefly, the model for the sedentary measurement y, of child i (i=1, 2, 3, …, N subjects) on day j (j=1, 2, 3, …, n_i days) is,

$$y_{ij} = x'_{ij}\beta + \upsilon_i + \varepsilon_{ij}, \qquad (1)$$

where x_{ij} is the vector of regressors and β is the corresponding vector of regression coefficients. The regressors can either be at the subject level, vary across occasions, or be interactions of the subject-level and occasion-level variables. The random subject effect υ_i indicates the influence of subject i on his/her sedentariness measures; these random effects are assumed to be normally distributed with zero mean and variance σ^2_v. The errors ε_{ij} are also assumed to be normally distributed with zero mean and variance σ^2_ε, and independent of the random effects. Here σ^2_v represents the between-subjects (BS) variance and σ^2_ε is the within-subjects (WS) variance. The mixed location scale model allows both of these variances to be modelled in terms of regressors using log link functions (to ensure positive variances). The coefficients from these variance models can be exponentiated to yield variance ratio estimates for the regressors (i.e., relative change in the variance per unit change in the regressor). Additionally, a random subject effect is included in the WS variance specification, which permits this variance to vary at the subject level, above and beyond the influence of regressors [22,23].

RESULTS

Table 1 shows descriptive statistics (Mean±SD and percentages) for the sample. There is a relatively high frequency (about 46%) of overweight/obesity. On average, children are about 2 years from their PHV. Almost 90% of the children reported 2 or less hours watching TV on week days, and 80% of them have at least one electronic media (TV/PC/game) in their bedroom.

Time spent in sedentary behaviour for each of the 686 subjects over an entire week is presented in Table 2 (for both sexes), and Fig 1 (for boys) and Fig 2 (for girls) show time spend in sedentariness from 20 random boys and girls. On average, boys spend less time in sedentary behaviour than girls in all week days (Monday to Friday), but no statistically significant difference was observed on Saturday and Sunday. The data reveal variability in sedentary time, with differences in their trajectories from Monday to Sunday. To give a sense of this variability at the subject level, we plot the gender-specific means in Fig 3, along with one subject from each group that is highly variable, and one subject that is rather consistent. Additionally, the two subjects in each gender group have the same average across time, though they differ quite a lot in terms of their variability. Individual difference in the WS variation is precisely what the mixed-effects location scale model allows for and attempts to explain.

Table 1. Descriptive characteristics of children

VARIABLE		**Mean±sd or Percentage (%)**
BMI (kg·m^{-2})		19.5±3.4
Maturity Offset (years to PHV)		-1.90±0.89
BMI (classification)		
	Normal-weight (0)	54.2%
	Overweight/Obese (1)	45.8%
Time watching TV on school days		
	≤2 hours/day (0)	89.7%
	>2 hours/day (1)	10.3%
Electronic media in bedroom		
	No (0)	19.8%
	Yes (1)	80.2%

Table 2. Mean±standard deviation for daily sedentary time (hours·day^{-1}) for boys and girls

Days	**Girls**	**Boys**
Monday*	9.52±1.46	8.92±1.66
Tuesday*	9.62±1.44	8.95±1.60
Wednesday*	9.55±1.40	8.88±1.65
Thursday*	9.43±1.44	8.87±1.64
Friday*	9.55±1.57	9.23±1.61
Saturday	9.08±1.60	8.86±1.83
Sunday	8.96±1.59	9.04±1.88

*statistically significant difference between boys and girls ($p<0.05$)

Fig 1. Time spent in sedentary behaviour over a week, for boys

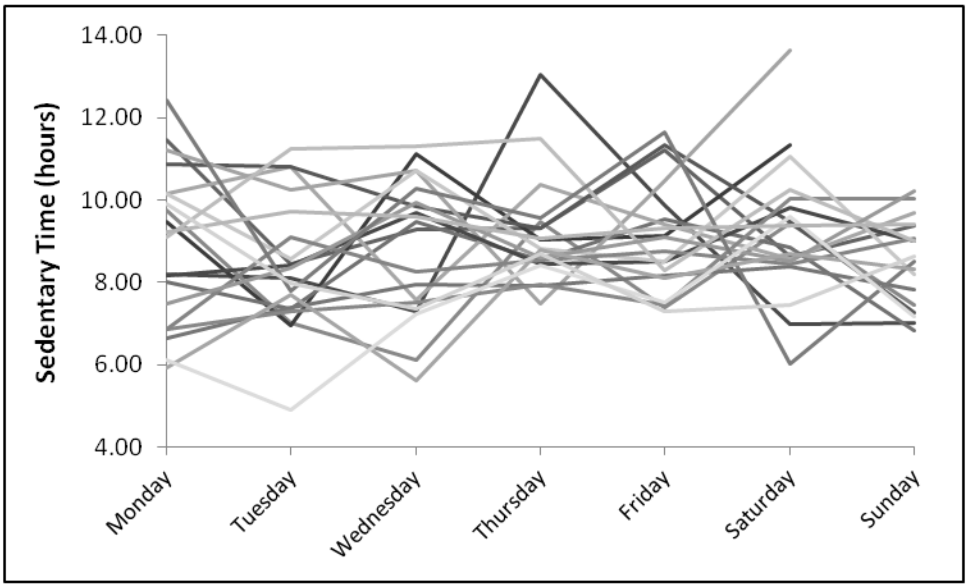


Fig 2. Time spent in sedentary behaviour over a week, for girls

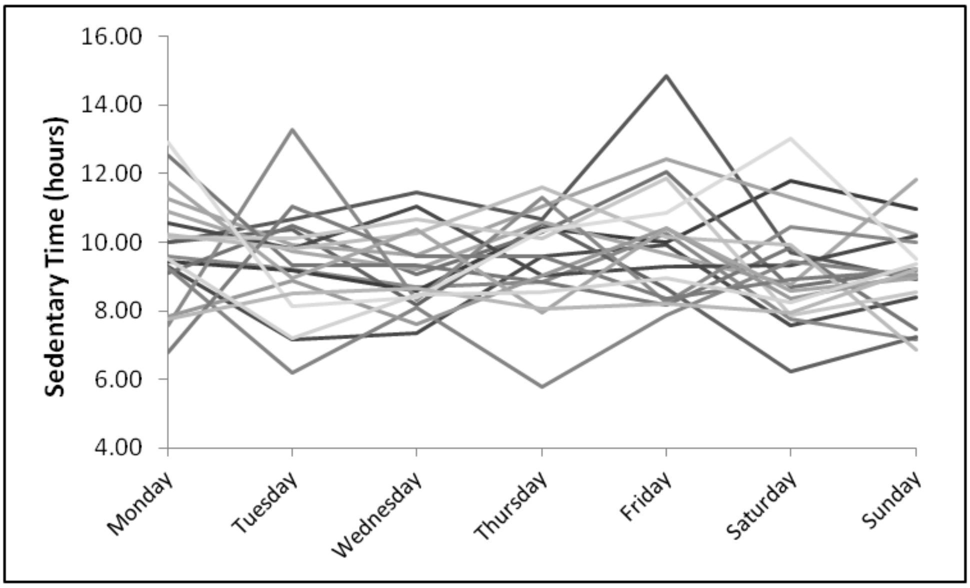
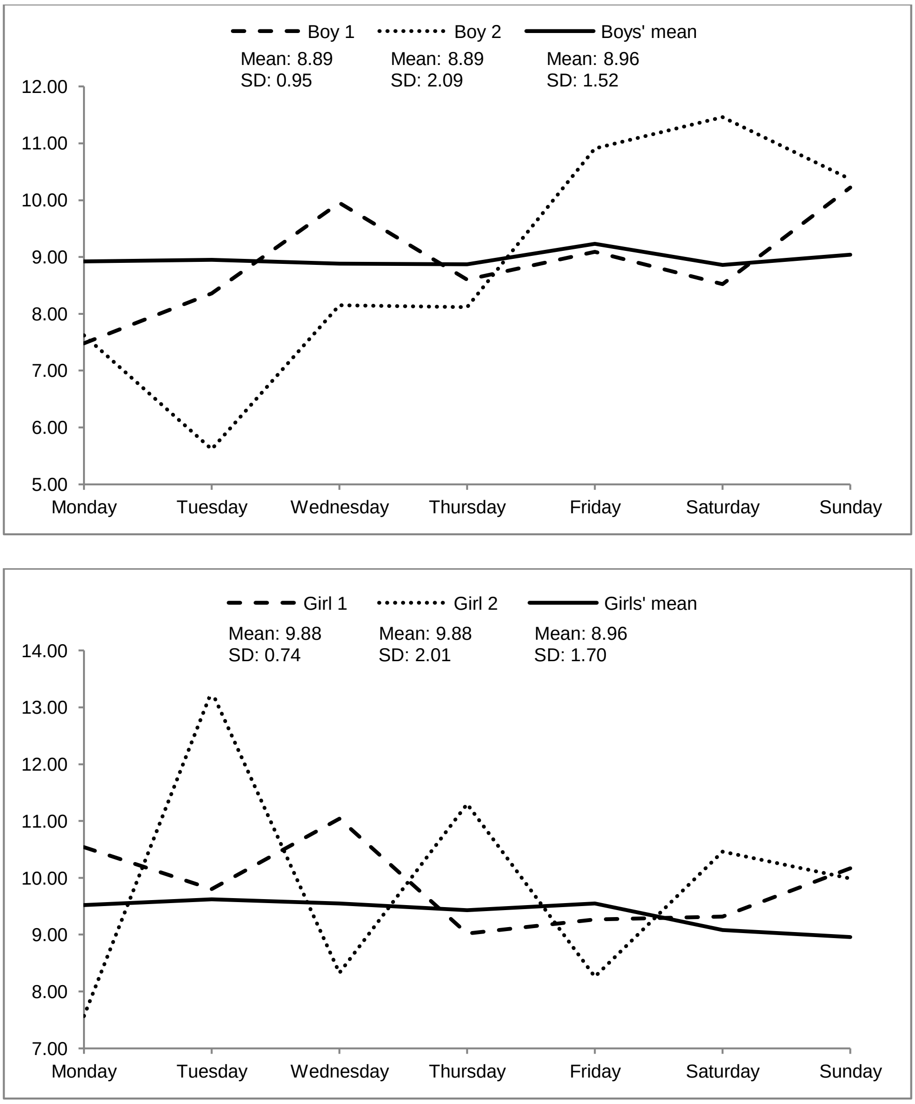


Fig 3. WS differences in sedentariness along a whole week, for 2 boys (up) and two girls (down), with same mean sedentariness time across the week



Results concerning the five research questions are included in Table 3. Note that although we have a three-level model, daily observations nested within children nested within schools, the software MIXREGLS does not accommodate 3 levels. In any case, we add school-indicators (22 dummy variables given that we have 23 schools) into the model. The results did not change in terms of the overall interpretation, and so, for simplicity, we are presenting the results from the models without these indicators.

The models are presented in increasing complexity, starting with a simple multilevel model to assess levels of BS and WS heterogeneity. Thus, it was started with a conventional random intercept model that only includes covariate effects on the mean (Model 1). Then, in Model 2, gender was added as a covariate for both the mean and BS variance model. Models 3 and 4 then introduced the novel aspects, which is the WS variance modelling in terms of covariates and (random) subject effects. The difference between Models 3 and 4 is the inclusion of the variables listed in Table 1 in both the mean and WS variance modelling. So, Model 1 addressed the question of a possible trend in sedentary behaviour across days of the week (Monday is the reference day). On average, the number of sedentary hours on Monday was 9.2±0.06, and no significant differences were found for Tuesday, Wednesday and Thursday (relative to Monday). Children were more sedentary on Friday, but less so on Saturday and Sunday (parameter estimates have a negative sign). There is significant variation BS across days as these estimates are from log link functions, the BS variance estimate is exp(-0.232)=0.79, and the WS estimate is exp(0.584)=1.79, resulting in a intra-subject correlation of 0.306. Concerning question two (Model 2), boys and girls (reference category) differ in the average level of sedentariness, with boys being less sedentary than girls, and the BS variance was seen to be 1.48 times larger for boys than girls (exp(0.390) = 1.48). Taken together, boys are, on average, less sedentary, but more heterogeneous than girls.

Model 3 addressed questions three and four, and fits significantly better than Model 2 (likelihood-ratio chi-square =16855.689-16712.490=143.2, which on 9 degrees of freedom is highly significant, $p<0.0001$). The fixed and BS variance parts are similar to Model 2. The novelties are located first in the WS variance modelling, and second in the random scale standard deviation. Tuesday, Wednesday and Friday have similar variability to Monday, i.e., same erraticism; Thursday has less, while Saturday and Sunday have more; further, significant sex differences were observed in this erraticism. The random scale standard deviation indicates whether the WS variance varies across subjects (over and above the effects of regressors on the WS variance). In other words, do subjects differ in how consistent/erratic they are in sedentary behaviour? It is highly significant, and so the degree of sedentary behaviour consistency/erraticism does vary significantly across subjects (over and above the day of week and sex effects).

Model 4 addressed question five about which variables, from the previously described set (Table 1), might be associated with this variability. Results in the fixed part of the model showed that sex, media, and time watching TV do not affect mean sedentariness time; BMI category is marginally associated; whereas children advanced in their biological maturation are more sedentary. There are significant inter-individual (BS) differences among subjects, as well as a significant sex effect. Erraticism is now significant only on Thursday, Saturday and Sunday; further, from the set of covariates, sex, BMI category, and maturation were significant, meaning that girls (variance ratio=0.632), overweight/obese children (variance ratio=0.861), and those less mature (variance ratio=0.849) have less erraticism than their counterparts. In terms of the effect of a person's mean on their WS variance, note that those subjects above average exhibited more erraticism than their peers with lower averages; in addition, the random scale standard deviation remains highly significant confirming that the WS variance does vary across subjects.

DISCUSSION

This study aimed to investigate the influence of several predictors of the BS and WS differences and variability in their sedentariness across seven days, following a set of five different questions. The first question was if there was a trend in children's sedentary behaviour over an entire week. Results showed that children tended to be more sedentary on Friday and less so on the weekend, when compared to Monday. Previous studies [31,32,33], aiming to identify children differences between weekdays and weekend days in their sedentary time, showed that they are more sedentary during weekdays. For example, Carson et al. [31] investigated the levels and bouts of measured sedentary time accumulated during different days of the week by 12-15 years old Australian girls, and reported that their sedentary time was higher on weekdays as compared to weekend days. Similarly, Steele et al. [32] also examined volume and patterns of sedentary activities during different segments of the week in UK children aged 9-10 years, and Harrington et al. [33] found higher levels of sedentary behaviour during weekdays when compared to weekend days among Irish female adolescents. This consistent pattern in sedentariness during the week days of children from different countries is governed, to some extent, by their school schedules and activities, which contribute to more sedentary behaviour among students. Harrington et al. [33] highlighted that during the period children spend at school they usually accumulate more sedentary time, implying that the school setting appears to impose sedentariness in children, especially promoting unbroken continuous periods of sitting. On the other hand, during the weekend, children have more opportunities to be physically active, spending less time in sedentary activities, such as sitting, reading or using the computer.

The second question addressed the issue of sex differences in sedentary behaviour, and boys were not only less sedentary but also showed more heterogeneity in their sedentariness across the seven days. Sex differences on average levels of both physical activity and sedentariness have been previously reported showing that girls usually tend to be more sedentary [10,34], which can be related to their options for sedentary activities during their leisure time (such as reading, listening to music, socializing with friends), while boys tend to engage in more physically activities (such as sport participation, competitive games) [35].

Since sex differences were found in the children's sedentary behaviour heterogeneity (Model 2, α=0.390±0.147, p=0.008), this was further explored across all days as well as its difference between boys and girls (Model 3) using the novelty and flexibility of the mixed-effects location scale model [22,23] which allows the WS variance to be modelled in terms of regressors and also allows subjects to vary in their consistency/erraticism. Results showed significant WS variance across days, meaning that children do not have the same sedentary pattern across all days. To our knowledge this is the first time that the idea of BS and WS variance in sedentariness has been jointly explored. It is expected that children vary in their physical activity and sedentariness between and within days [36], and some studies have investigated the within-day variability in sedentariness in children and adolescents considering the school-time and outside school-time. For example, Harrington et al. [33] did not find differences in girls' sedentary time between these two periods of the day, but reported that these adolescents tend to accumulate more sedentary bouts during school time. On the other hand, Steele et al. [32] reported period and sex differences in sedentary time during the day, with boys spending more time in sedentary activities out of school while no differences in sedentariness between "in-school" and "out-of-school" periods were observed in girls. However, these "trends" may not be similar in all children given that the analysis by Harrington et al. [33] and Steele et al. [32] were based on averages and/or percentages of the total time. We think that differences in children's sedentariness may be more properly addressed by the modelling of the WS variance, which is not always taken into account. Previous research specifically addressing the issue of intra-individual differences, although in aging, reported by Hertzog and Nesselroade [37], clearly stated that using averages to describe changes is not always the best way to detect key features of developmental changes and/or short-term differences. Further, they showed that change can vary within a person over weeks, even when the time of the day and day of the week of testing is kept constant. Additionally, Epstein [38] pointed out that not everyone is equally predictable, highlighting that a significant WS variance exists and that it should be taken into account, independent of the outcome variable. Fig 3 highlights the WS differences in sedentariness along a whole week in two boys and two girls. Children with the same (or similar) mean sedentariness time show different sedentariness trajectories and patterns during the week, revealing that WS variance exists and that should be taken into account when studying correlates of sedentary behaviour.

Table 3. Parameter estimates (±standard errors) of the four models

Model parts	Model 1	Model 2	Model 3	Model 4
	β±SE	β±SE	β±SE	β±SE
Fixed part				
Intercept	9.243±0.062**	9.433±0.068**	9.451±0.066**	9.740±0.168**
Tuesday	0.070±0.074^{ns}	0.069±0.074^{ns}	0.077±0.067^{ns}	0.073±0.067^{ns}
Wednesday	0.004±0.074^{ns}	0.005±0.074^{ns}	0.008±0.068^{ns}	0.006±0.068^{ns}
Thursday	-0.064±0.073^{ns}	-0.064±0.073^{ns}	-0.058±0.065^{ns}	-0.064±0.065^{ns}
Friday	0.167±0.074**	0.167±0.074***	0.184±0.071**	0.187±0.071**
Saturday	-0.261±0.073**	-0.260±0.073**	-0.265±0.076**	-0.267±0.076**
Sunday	-0.267±0.074**	-0.267±0.074**	-0.294±0.077**	-0.297±0.076**
Sex		-0.430±0.079**	-0.475±0.079**	-0.104±0.165^{ns}
BMI				-0.151±0.090*
Maturity Off				0.242±0.090**
Time TV				0.183±0.126^{ns}
Media				0.072±0.095^{ns}
Between Subject (BS) variance				
Intercept	-0.232±0.073**	-0.482±0.104**	-0.401±0.098**	-0.436±0.099**
Sex		0.390±0.147**	0.299±0.145**	0.330±0.146**
Within Subject (WS) variance				
Intercept	0.584±0.022**	0.584±0.022**	0.330±0.071**	0.581±0.132**
Tuesday			-0.106±0.094^{ns}	-0.109±0.094^{ns}
Wednesday			-0.023±0.092^{ns}	-0.024±0.092^{ns}
Thursday			-0.199±0.093**	-0.204±0.093**
Friday			0.124±0.092^{ns}	0.130±0.092^{ns}
Saturday			0.369±0.091**	0.370±0.091**
Sunday			0.360±0.092**	0.356±0.092**
Sex			0.199±0.055**	0.459±0.115**
BMI				-0.150±0.064**
Maturity Off				0.164±0.066**
Time TV				-0.072±0.089^{ns}
Media				0.026±0.068^{ns}
Random location (mean) effect on WS variance				
Loc eff			0.086±0.031**	0.080±0.031**
Random scale standard deviation				
Std dev			0.348±0.037**	0.342±0.037**
Deviance (-2 Log L)	16892.843	16855.689	16712.490	16694.754

ns= non-significant; *<0.10; **<0.05; -2 Log L= -2 Log-likelihood

The usual approach in population studies, taking into account only the mean values of the entire sample, is based on the assumption that results and the distribution of the variables at the population level somehow reflect within-person processes, which allow the generalization from the population to the individual [39]. However, as we showed in Fig 2, results at the person level are different from those at the sample level, and as reported by Hamaker [39], when there are individual differences that cannot be ignored, it is necessary to “investigate to see what predicts them or what they predict” [39, p. 52]. This was the main drive of our last question (results in Model 4) addressing the simultaneous effects of sex, BMI categories, biological maturation, time watching TV, and electronic media in the bedroom in children mean differences (expressed in the fixed part of the model) and intra-individual variability (expressed in the WS variance) in sedentariness across the seven days. From the set of covariates tested, only biological maturation was significantly related to mean levels of sedentariness, indicating that more mature children tend to be more sedentary than their less mature peers. This result is in accordance with previous studies where this association was reported [12,40]. For example, Brodersen et al. [12] described that more advanced puberty was associated with greater sedentariness in youth. Similarly, Machado Rodrigues et al. [40] found that maturity status is a significant predictor of sedentariness, but only in boys. Since girls mature earlier than boys, it is possible that differences in maturation timing and tempo may also explain sex differences in sedentariness, as described in associations with physical activity and exercise [41,42,43], but this issue is still unclear [40].

From those variables related to the WS variance in sedentariness, sex (τ=0.459±0.115, $p<0.001$), BMI categories (τ=-0.150±0.064, $p=0.019$) and biological maturation (τ=0.164±0.066, $p=0.013$) showed significant effects on the variance in sedentariness, meaning that girls, overweight/obese children and those late in their maturation have a lower erraticism in their sedentariness. Further, this result reinforces the need for a careful study of WS variance. Since, in general, the covariates at WS level differ from those at BS level as well as from those at mean level of sedentariness, generalization of the results about covariates from the inter-individual level can extend to intra-individual variability, or considering erraticism a nuisance, may not be appropriate. In addition, as highlighted by Molenaar (cited by Hamaker [39]), this generalization from the population to the individual is only appropriate when the population moments (means, variances, and covariances) are identical to the corresponding within-person moments, which are not the case in almost situations.

Some limitations in the present study should be discussed. Firstly, since children spend a substantial part of their awake time at school, the school environment can have a relevant role at WS variance in sedentariness;

however, school context characteristics were not included in the model, and its effect on children's sedentariness or physical activity is not always clear, given the large variability found in the school effects' intraclass correlation going from ≈0.06 to ≈0.36 [18,44]. Secondly, it is possible that children vary in their sedentariness also within a day, due to their different surroundings (e.g., school, home, sports club), and studies of sedentariness variance during the day can offer relevant information about children's patterns of sedentary behaviour. The use of ecological momentary assessment approaches may be highly useful in unravelling this issue [45,46]. Thirdly, the sample comes from only one Portuguese region, meaning that results cannot be generalized to other Portuguese children. However, in data not shown, similar results were observed in some sample characteristics between our sample and others from previous studies, namely in the prevalence of overweight/obesity [47] and socioeconomic status distribution [48]. In spite of these limitations, several strengths should be pointed out: (1) to our knowledge, this is the first study that explored WS variance in sedentariness, highlighting the relevance of understanding the BS variance, the WS variance as well as their possible predictors; (2) the use of an objective method to estimate sedentariness during a whole week; (3) the use of standard methods and reliable data; (4) and the use of the mixed-effects location scale model to study the complexity of BS and WS variance in sedentariness.

CONCLUSIONS

This study showed that children are significantly different in their sedentariness during the days of the week, and tend to be less sedentary during the weekend (suggesting that the school context may play a relevant role), and that sex difference exists regarding to sedentariness. Within-child consistency/erraticism showed high variability across days, meaning that children do not have the same sedentariness patterns along the week; further, sex, BMI, and biological maturation have significant effects on the sedentariness variance. Since results from between- and within-child are not the same, namely in their correlates, this reinforces the need to a deeper investigation on intra-individual variability above and beyond the normative view of mean values and heterogeneity among subjects. In addition, results found at the inter-individual level do not generalize to intra-individual level. The approach used in the mixed-effects location scale model showed to be very important in providing detailed information for a better understanding of correlates that best explain intra-individual sedentariness consistency/erraticism. Taken together, these findings provide evidence that a more complete map of children's patterns in sedentary behaviour will be highly important when designing intervention strategies to reduce their sedentariness and associated health hazards, and that within-child variance should not be neglected.

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Paper III

Overweight and obesity in Portuguese children: prevalence and correlates

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ABSTRACT

There are widespread differences in overweight/obesity prevalence in children, and understanding the reasons for this is very important. The present study aims: (I) to conduct a meta-analysis on overweight/obesity prevalence in Portuguese children; (II) to identify differences in biological and behavioural characteristics between normal-weight and overweight/obese children; and (III) to investigate the importance of individual- and school-level correlates of variation in children's BMI using multilevel modelling. A search was done for all published papers including Portuguese children during the last decade; further, 686 Portuguese children (9–11 years old) were sampled and their BMI, family income, maturity offset, nutritional habits, physical activity, sedentariness, sleep time, and school environment information were collected. Results showed a stabilization of overweight/obesity during the last decade, 30.6% (95%CI: 0.287–0.34) for boys, 28.4% (95%CI: 0.23–0.35) for girls, and 30.3% (95%CI: 0.27–0.34) for boys and girls together. Differences between weight groups were only found in individual-level biological traits. The multilevel analysis did not identify significant contributions of school-level variables to children's BMI variation. In conclusion, no increase was found in the prevalence of overweight/obesity among Portuguese children since 2000. Normal-weight and overweight/obese children only differ in individual-level characteristics, and school context variables were not related to variation in BMI.

Keywords: overweight; obesity; children; school; correlates

INTRODUCTION

Given the systematic rise in the prevalence of overweight/obesity in youth during the last decades [1,2], determining associated factors in children is necessary, especially to design efficient strategies targeting obesity/overweight reduction. Among Portuguese children, Bingham *et al*. [3] reported that being male, having been breastfed, having been born from mothers who did not smoke during pregnancy, engaging in little sedentary behaviour, performing at least 1 hour of moderate physical activity daily, and having parents with higher education levels and a healthy body mass index (BMI) were protective factors against childhood overweight/obesity. Strategies implemented to reduce overweight/obesity in children, especially at the school level, have reported inconclusive outcomes - some showed significant results [4,5], while others did not [6,7].

It has been suggested that overweight/obese children differ in their behavioural and dietary habits from those with healthy weight [8], namely that they tend to have a higher consumption of fat and a lower carbohydrate intake [9], engage in more sedentary activities (such as watching TV, movies, or playing video games) [10], and spend less time in moderate-to-vigorous daily physical activities [11], although this unhealthy profile is not always observed [12]. For example, Maier *et al.* [13] did not find differences between normal-weight and overweight children in total energy and macronutrient intake. In addition, when studying differences in physical activity and sedentary behaviour among Chinese youth, Wang *et al.* [14] reported no differences among normal-weight, overweight or obese youth.

The prevalence of overweight and obesity among Portuguese children may have increased during the last decades [15] leading to concern among public health authorities [16]. A better understanding of differences between normal-weight and overweight/obese children's lifestyle characteristics is needed to reduce the negative behavioural and health effects of excessive weight in childhood. Further, as children spend most of their daily time at school, where healthy behaviours are learned [17], the school environment may promote healthy habits that positively affect children's weight status [18].

In order to better understand the prevalence and correlates of overweight and obesity in Portuguese children, the present study aims to: (I) conduct a meta-analysis on overweight/obesity prevalence in 9 to 11 year old Portuguese children, (II) detect significant differences in behavioural characteristics among normal-weight and overweight/obese children; and (III) investigate the importance of individual- and school-level correlates on variation in children's BMI.

METHODS

To address this study's aims, we present the methodology in two parts: Part I addresses the meta-analysis and Part 2 focuses on aims II and III.

Part I: Meta-analysis of obesity prevalence among Portuguese children

Between June and July 2014, an online search was conducted using *Scopus, Pubmed* and *Scielo* databases to find all available articles reporting overweight and obesity prevalence in Portuguese children using the following keywords: *overweight*, *obese*, *obesity*, *children*, *youth, Portugal*, *Portuguese* and their respective translation to Portuguese by the first author. In addition, another search was done at the Faculty of Sport, University of Porto central library, and Portuguese Statistics databases with the same keywords. Valid papers, or Health Directorate Reports, were included if they: (I) were published between January 2000 and May 2014 because *International Obesity Task Force* (IOTF) cut-points were first published in 2000; (II) sampled Portuguese children aged 9 to 11 years [to be in agreement with the *International Study of Childhood Obesity, Lifestyle and the Environment* (ISCOLE) study sample age range, as mentioned below]; (III) used national samples; (IV) reported overweight/obesity prevalence; (V) used BMI to assess overweight/obesity; (VI) used IOTF [19] cut points to define overweight and obesity; and (VII) were published in English or Portuguese.

A meta-analysis of overweight/obesity prevalence was conducted using the Comprehensive Meta-Analysis v2.2.64 software [20]. Prevalence, 95% confidence intervals, Q-test and I^2 statistic were computed according to algorithms implemented in the software. Further, the software was also used to assess effect size heterogeneity as advocated by Borenstein *et al*. [21] and Beretvas [22], and fixed and random effects models were used.

Part II: Correlates of childhood overweight and obesity

Sample

The sample of the present study is part of ISCOLE, a research project conducted in 12 countries (Australia, Brazil, Canada, China, Colombia, Finland, India, Kenya, Portugal, South Africa, the United Kingdom, and the United States of America) from all major regions of the world. Its main aims are to determine the relationship between behaviours and obesity in a multi-national study of children aged 9 to 11 years, and to investigate the influence of higher-order characteristics such as behavioural settings, and physical, social and policy environments, on the observed relationship within and between countries [23]. Details regarding the ISCOLE study design and methodology were previously reported elsewhere by Katzmarzyk *et al*. [23].

The sample of the present study comprises 777 Portuguese children, aged 9 to 11 years, from 23 schools from the North of Portugal. In each school, after the project was approved by the Physical Education Department, school Principal and Parental Council, all 5th grade students were invited to enrol in ISCOLE, but only those children aged 9 to 11 years old were classified as eligible. From those, approximately 30 to 40 children per school were randomly selected (50% of each sex). Non-response was negligible (response rate was 95.7%), and missing information was at random (differences between subjects with missing information and those included were not statistically significant). The study protocol was approved by the University of Porto ethics committee, as well as by schools' directorate councils. Written informed consent was obtained from parents or legal guardians of all children.

Anthropometry

Height, sitting height and weight were measured according to standardized ISCOLE procedures and instrumentation [23]. Height was measured using a Seca 213 portable stadiometer (Hamburg, Germany) without shoes, with the head in the Frankfurt Plane, and sitting height was measured while seated on a table with legs hanging freely and arms resting on the thighs. Body mass was determined with a portable Tanita SC-240 scale (Arlington Heights, IL, USA), after all outer clothing, heavy pocket items and shoes were removed. Two measurements were taken on each child, and a third measurement was taken if the difference between the previous two was outside the permissible range for each measure and its replica (0.5 cm for height and sitting height, and 0.5 kg for weight). The mean value of each measured variable (closest two measurements) was used for analysis. BMI was computed using the standard formula [weight(kg)/height(m)^2], and subjects were classified as normal-weight, overweight and obese according to the IOTF cut-off points suggested by Cole *et al*. [19].

Family data

Information regarding family environmental characteristics was obtained from a questionnaire completed by parents or legal guardians (see ISCOLE Demographic and Family Health Questionnaire in Katzmarzyk *et al*. [23]). The questionnaire collected information on basic demographics, ethnicity, family health and socioeconomic factors. For the present study, we used information regarding familial socioeconomic status (SES) and parental BMI. SES was defined according to annual family income, ranging from <€ 6,000 to ≥€ 42,000, and subjects were classified in two categories (<€23,999; ≥€24,000).

Biological maturity

Using age, sex, sitting height, stature and body mass, a biological maturity estimate was obtained using the Mirwald *et al*. regression equations [24]. The set of equations, jointly labelled as maturity offset equations, estimates the timing to peak height velocity (PHV) occurrence. A positive (+) offset expresses the number of years a child is beyond PHV; a negative score (–) signifies the number of years a child is from PHV; a value of zero indicates that a child is presently experiencing his/her PHV.

Nutritional and behavioural habits

Information on diet and lifestyle was obtained from a questionnaire answered by each child [23], which includes questions about the frequency of consumption of different types of food in a typical week. Information related to fruits, vegetables, sweets, soft drinks and fast food consumption was assessed. Using principal components analysis, dietary scores were derived for each child from the children's Food Frequency Questionnaire [23] food groups as input variables (excluding fruit juices), expressing children's dietary patterns. Reported frequencies were converted into portions/week. Eigenvalues and a scree plot analysis were used as the criteria for deciding the number of components extracted. The two criteria led to similar conclusions, and two factors were chosen for each analysis. The components were then rotated with an orthogonal varimax transformation to force non-correlation of the components and to enhance the interpretation. The component scores computed for each subject for both dietary patterns were standardized to ensure normality. The two components were named "unhealthy food" (e.g., hamburgers, soft drink, fired food, *etc*.) and "healthy food" (e.g., vegetables and fruits). Time spent watching TV during the week was reported by children, and then categorized according to screen time recommendations (<2 hours/day and ≥2 hours/day). Children also reported whether or not if they had a TV available in their bedroom, as well as their main transportation method to/from school.

Physical activity, sedentary time and sleep

Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL, USA) were used to monitor physical activity, sedentary time and sleep. Children wore the accelerometer at their waist on an elasticized belt, placed on the right mid-axillary line 24 hours/day, for at least seven days, including two weekend days. To be eligible for this analysis, children had at least four days (including at least one weekend day) with a minimum of 10 hours of wear time per day. From the original sample of 777 children, 686 children fulfilled this condition. Accelerometer information was divided into daytime activities and nocturnal sleep time using an automated algorithm [25,26]. Non-wear time during the awake period was defined as any sequence of at least 20 consecutive minutes of zero activity counts [26].

Different activity phenotypes were determined using cut-points developed by Evenson *et al*. [27]. For the present study, mean moderate-to-vigorous physical activity (MVPA) and mean sedentary time were used, which were defined as greater than or equal to 574 activity counts and less than or equal to 25 activity counts using 15 seconds epochs, respectively.

The nocturnal sleep time for each participant was determined using a novel and fully-automated algorithm specifically developed for use in ISCOLE and other epidemiological studies employing a 24-hour waist-worn accelerometer protocol in children [25,26]. Mean sleep time across all days was used in the analyses.

School environment

Information concerning the school environment was obtained via a questionnaire (ISCOLE School Environment Questionnaire presented in Katzmarzyk *et al*. [23]) completed by the physical education teacher or the school principal. For the present study we primarily considered the following aspects of the school physical activity environment: the percentage of students participating in school sports or PA clubs; school promotion of active transportation (allowing children to bring their bicycles); student access to a gymnasium during school hours and outside school hours; student access to playgrounds during school hours; student access to sports equipment outside of school time; student access to a cafeteria at school; student access to food and drink vending machines; and student access to fast food restaurant close to school.

Statistical analysis

Differences in means and frequencies of biological and behavioural characteristics between groups were computed using Student-t and χ^2 tests. SPSS 20.0, and WinPeppi software [28] were used for these analyses. The extraction and identification of dietary patterns were performed in the SAS 9.3 (SAS Institute Inc., Cary, NC, USA, 2011).

To answer aim III and given data dependency, students nested within schools, a multilevel approach was used and the analysis was done in SuperMix software [29] allowing a simultaneous estimation of all model parameters using maximum likelihood procedures. A series of hierarchical nested models were fitted to explain variation in children's BMI using the Deviance statistic as a measurement of global fit [30]. Additionally, the relevance of predictors to explain variation in BMI was assessed with a pseudo-R^2 statistic, which is interpreted as a proportional reduction in variance for the parameter estimate resulting from the use of one model as compared to a previous one [30]. Modelling was done in a "stepwise" fashion as generally advocated [31,32]. Firstly, a null model (M_0) was fitted to the data to compute the intraclass correlation coefficient to estimate the variance accounted for by the school effects in BMI. Secondly, using child-level BMI predictors (sex, biological maturity, mother and father BMI, TV/PC use during weekdays, having a TV in the bedroom, diet categories, time spent in MVPA, sedentariness and sleeping), Model 1 (M_1) was fitted. Parental BMI, time spent in MVPA, sedentariness and sleeping were centred at the grand mean to facilitate the interpretation of parameter estimates. Thirdly, with the inclusion of school-level predictors, Model 2 (M_2) was fitted. Statistical significance was set at $p < 5\%$.

RESULTS

Prevalence of overweight/obesity among 9–11 year-old Portuguese children

Figure 1 presents a flow diagram illustrating the search process and the excluded studies in the meta-analysis. Only five studies fulfilled all inclusion criteria based on a close examination of abstracts and full texts. Further, this final list was checked by the first author against two recent systematic literature reviews concerning overweight and obesity in Portuguese children and adolescents [33,34]. Table 1 shows the available data using national samples. Studies were published between 2004 and 2013, and the sample sizes ranged from 405 to 3,584 subjects. The highest prevalence of overweight/obesity was for boys in 2009 [16], while girls in 2007 [35] had the lowest prevalence. A higher prevalence of overweight/obesity was found in boys in three [16,35,36] of the five studies, compared to girls. Taking boys and girls together, the prevalence of overweight/obesity ranged from 19% [36] to 35% [16] representing a moderate-to-high prevalence of Portuguese children with excess weight.

Figure 2 presents Forrest plots, fixed and random effects prevalence estimates and their 95% confidence intervals for overweight/obesity across time in boys, girls, and both sexes together. Although there is considerable evidence for heterogeneity in the prevalences for boys (Q-test = 11.371, $p = 0.023$, $I^2 = 64.823$), girls (Q-test = 56.564, $p < 0.001$, $I^2 = 92.928$), and both sexes together (Q-test = 25.770, $p < 0.001$, $I^2 = 84.478$), we nevertheless present fixed and random effects prevalence estimates (see Figure 2) although they are fairly similar. Across the time period, the prevalence estimate for overweight/obesity among boys is 0.306 (95%CI: 0.277–0.337), 0.284 (95%CI: 0.225–0.352) among girls and 0.303 (95%CI: 0.272–0.335) for both sexes. The funnel plot did not show evidence of publication bias; further, meta-regression analysis using study year as a moderator variable did not show any significant increase in overweight/obesity from 2002 till 2010 (boys, beta = 0.0096 ± 0.0132, $p \geq 0.05$;

girls, beta = 0.0220 ± 0.0127, $p \geq 0.05$; both sexes together, beta = 0.0178 ± 0.0916, $p \geq 0.05$).

Figure 1. Flow diagram of study selection for meta-analysis

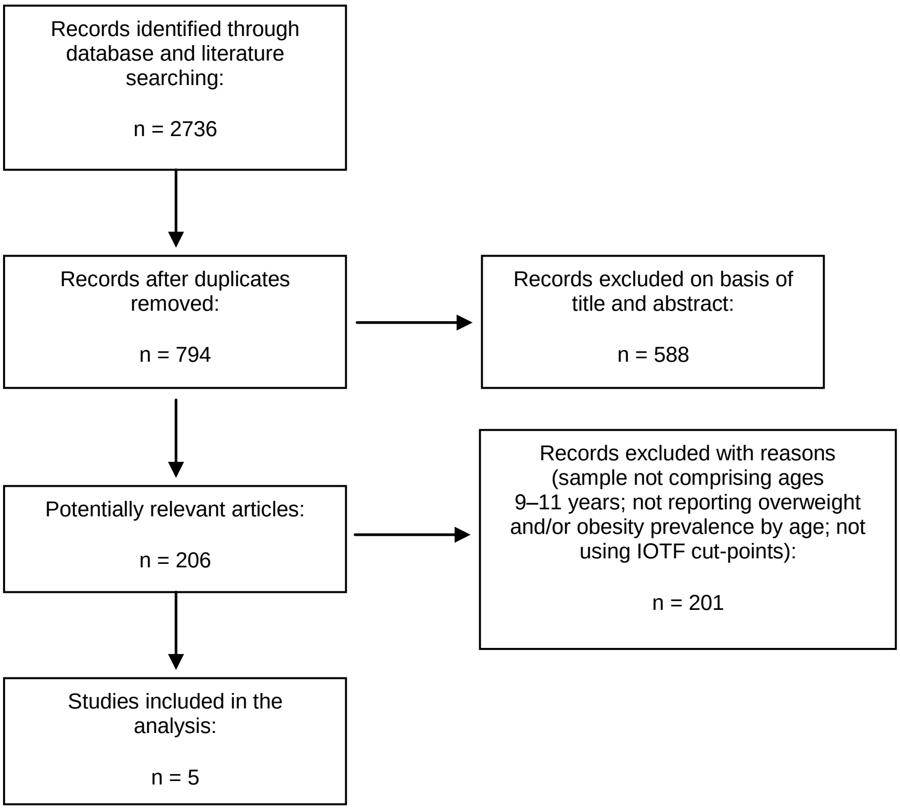
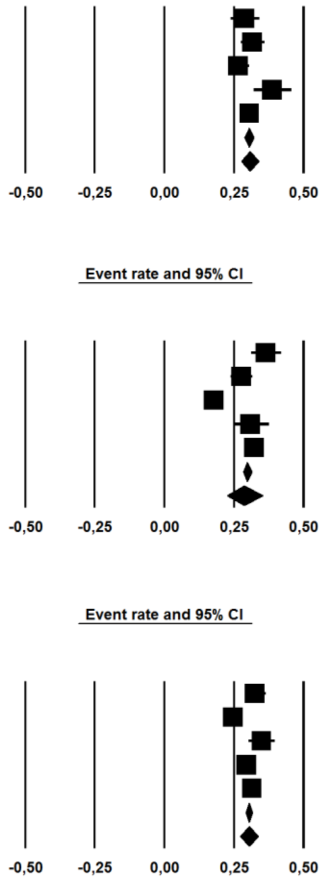


Table 1. Summary of overweight/obesity prevalence in 9–11 year-old Portuguese children used in the meta-analysis

Study	Study Year	Age Range	Sample Size	Prevalence of Overweight/Obesity		
				Boys	**Girls**	**Total**
Padez *et al.* [37]	2002/2003	9.5 years	631 (317 boys; 314 girls)	29%	36%	33%
Yngve *et al*. [35]	2003	11 years	1197 (552 boys; 645 girls)	27%	18%	22%
DGS [16]	2008	11 years	405 (204 boys; 201 girls)	39%	31%	35%
Sardinha [36]	2008	10 years	1001 (486 boys; 515 girls)	32%	28%	19%
Bingham *et al*. [3]	2009/2010	9–10 years	3584 (1685 boys; 1899 girls)	31%	32%	31%

Figure 2. Meta-analysis results for boys, girls and both sexes combined



Boys

Model	Study name	Event rate	Lower limit	Upper limit	Z-Value	p-Value
	Padez et al., 2005	0,287	0,240	0,339	-7,327	0,000
	Yngve et al., 2007	0,315	0,275	0,357	-7,963	0,000
	DGS, 2009	0,264	0,229	0,303	-10,598	0,000
	Sardinha et al., 2011	0,387	0,323	0,456	-3,193	0,001
	Bingham et al, 2013	0,305	0,284	0,327	-15,562	0,000
Fixed		0,304	0,288	0,320	-21,689	0,000
Random		0,306	0,277	0,337	-11,237	0,000

Girls

Model	Study name	Event rate	Lower limit	Upper limit	Z-Value	p-Value
	Padez et al., 2005	0,363	0,312	0,418	-4,790	0,000
	Yngve et al., 2007	0,276	0,239	0,316	-9,794	0,000
	DGS, 2009	0,177	0,149	0,208	-14,905	0,000
	Sardinha et al., 2011	0,308	0,248	0,376	-5,286	0,000
	Bingham et al, 2013	0,322	0,302	0,344	-15,139	0,000
Fixed		0,297	0,282	0,312	-23,272	0,000
Random		0,284	0,225	0,352	-5,793	0,000

Total

Model	Study name	Event rate	Lower limit	Upper limit	Z-Value	p-Value
	Padez et al., 2005	0,325	0,289	0,362	-8,605	0,000
	Yngve et al., 2007	0,246	0,223	0,272	-16,664	0,000
	DGS, 2009	0,348	0,303	0,396	-6,013	0,000
	Sardinha et al., 2011	0,295	0,267	0,324	-12,587	0,000
	Bingham et al, 2013	0,314	0,299	0,330	-21,694	0,000
Fixed		0,303	0,292	0,314	-31,483	0,000
Random		0,303	0,272	0,335	-11,001	0,000

Biological, behavioural and socio-demographic differences between normal-weight and overweight/obese children

Descriptive data comparing differences between normal-weight and overweight/obese children in biological, behavioural and socio-demographic traits are presented in Table 2. Except for biological traits (height, weight, and parental BMI), no statistically significant differences ($p > 0.05$) were found in any of the other variables between the two groups.

Table 2. Biological, behavioural and socio-demographic trait differences between normal-weight and overweight/obese children

	Normal-weight (n = 417)	**Overweight/Obese (n = 269)**	***t***	***p*-value**
Height (cm)	141.57 ± 6.39	145.73 ± 6.55	−8.403	<0.001
Weight (kg)	34.14 ± 4.59	47.79 ± 7.74	−28.584	<0.001
Biological maturity	−2.07 ± 0.85	−1.69 ± 0.90	−5.639	<0.001
Unhealthy diet (z-scores)	0.005 ± 1.00	−0.10 ± 0.91	1.424	0.155
Healthy diet (z-scores)	0.04 ± 0.98	−0.03 ± 1.03	0.945	0.345
MVPA (min)	57.34 ± 22.48	55.20 ± 20.54	1.291	0.197
Sedentary time (min)	553.26 ± 61.92	549.68 ± 61.90	0.754	0.451
Sleep time (hours)	8.26 ± 0.83	8.26 ± 0.90	-0.027	0.979
		Frequencies (%)	χ^2	***p*-value**
SES			0.119	0.730
<€23,999	78.4	79.6		
≥€24,000	21.6	20.4		
Mother BMI			11.744	0.001
Normal-weight	66.3	52.6		
Overweight/obese	33.7	47.4		
Father BMI			4.898	0.027
Normal-weight	41.3	32.2		
Overweight/obese	58.7	67.8		
Transport to/from school			0.339	0.561
Active	28.8	26.8		
Non-active	71.2	73.2		
TV bedroom			3.564	0.059
No	33.1	26.4		
Yes	66.9	73.6		
TV/school day			0.743	0.389
<2 hours	73.7	70.7		
≥2 hours	26.3	29.3		

Individual- and school-level correlates of BMI variation

Results from M_0, M_1 and M_2 are presented in Table 3. From M_0, the intraclass correlation coefficient is 0.0216 [0.25/(0.25 + 11.30)], meaning that only 2.2% of the total variance in BMI among schoolchildren is at the school level.

Results from M_1 (child-level predictors) show that boys (β = 6.22, p < 0.001) have, on average, higher BMI than girls; more mature children had higher BMI (β = 3.94, p < 0.001); fathers (β = 0.12, p < 0.001) and mothers (β = 0.15, p < 0.001) with higher BMIs have children with higher BMIs, suggesting familial aggregation. On the other hand, a higher healthy diet score (β = −0.24, p = 0.018) showed a negative effect on BMI, as well as MVPA (β = −0.02, p < 0.001). There was a significant reduction in Deviance from 3621.93 to 2332.03 (χ^2 = 1289.90, p < 0.001 from M_0 to M_1) showing the better fit of this model. Approximately 85% of the variance in BMI was explained by level-1 predictors. The final model, M_2, which adds school contextual predictors, showed that none were significantly associated with BMI. Although there was a reduction in Deviance from M_1 to M_2 (from 2332.03 to 2323.03), it was not significant (χ^2 = 9.02, p = 0.172). Further, as the intraclass correlation was small (2%), no significant associations in school predictors were expected.

DISCUSSION

Since this study was structured according to its three aims: (I) to conduct a meta-analysis on overweight/obesity prevalence in 9 to 11 year old Portuguese children; (II) to detect differences in behavioural characteristics among normal-weight and overweight/obese children, and (III) to investigate the importance of individual- and school-level correlates on children's BMI variation, the discussion will follow accordingly.

Prevalence of overweight/obesity in 9–11 year-old Portuguese children

There is compelling evidence that overweight/obesity in children is one of the most important public health problems worldwide. According to de Onis *et al*. [1], childhood obesity prevalence increased from 4.2% in 1990 to 6.7% in 2010, and it is expected to reach 9.1% in 2020. A recent Portuguese study [15] among 7–9 year-old children, using data from 1970, 1990 and 2002, reported mean increases in BMI during these time periods, with greater increases between 1992 and 2002. However, as the 1970 and 1992 studies did not report overweight/obesity prevalence, it was not possible to determine a trend although they hypothesized that the increase observed from 1992 to 2002 could be linked to increases in the prevalence of overweight/obesity observed in the last years. Our meta-analysis results suggest that the previous increases in obesity may have slowed or plateaued in Portugal. Our findings are in agreement with recent trends of stability or decrease in the prevalence of overweight/obesity in youth. For example, among Dutch, Moroccan and Surinamese South Asian ethnic groups in the Netherlands, aged 3–16 years, de Wilde *et al*. [38] found a stabilization, or even a decrease, in the prevalence of overweight/obesity between 1999-2011. Moreover, similar results were reported by Moss *et al*. [39] studying German children, especially from the year 2004 onward; further, they reported a decline in the prevalence of overweight from 8.4%–11.9% to 3.3%–5.4%, varying according to regions, with an absolute decrease of prevalence rates up to 3% for overweight and 1.8% for obesity. Additionally, among Danish children aged 7–14 years [40] (using self-reported values for height and weight), the prevalence of obesity did not increase significantly from 1995 to 2001 (2.3% to 2.4%), but the prevalence of overweight rose 10.9% to 14.4%). Among French children [41], aged 3–14 years, no significant difference was found in the prevalence of overweight/obesity from 1999 to 2007. Notwithstanding World Health Organization data showing increases in overweight/obesity in young populations [1], it seems that in some nations a levelling off in childhood overweight/obesity has been observed [42-44], which calls for a more reliable analysis about variations among countries. In our meta-analysis, the prevalence of overweight/obesity ranged across studies from 19% to 35%, with an overall estimate (from fixed and random effects) of approximately 30% (for boys and girls, together), which is higher than those reported from previous studies. Although one study relied on self-reported height and weight [35], its results were not different from the others.

Table 3. Multilevel modelling results: regression estimates (*β*), standard-errors (SE), and p-values for children and school characteristics influencing BMI variation

Parameters	Model 0			Model 1			Model 2		
	β	***SE***	***p-value***	*B*	***SE***	***p-value***	*β*	***SE***	***p-value***
Intercept	19.46	0.17	<0.001	23.81	0.41	<0.001	22.77	0.89	<0.001
Sex				6.22	0.35	<0.001	6.23	0.45	<0.001
Biological maturity				3.94	0.20	<0.001	3.96	0.24	<0.001
Mother BMI				0.15	0.02	<0.001	0.15	0.03	<0.001
Father BMI				0.12	0.03	<0.001	0.12	0.03	<0.001
TV weekdays				0.13	0.26	0.622	0.16	0.25	0.540
TV in bedroom				0.34	0.25	0.164	0.32	0.25	0.205
Unhealthy diet				−0.20	0.13	0.142	−0.20	0.12	0.098
Healthy diet				−0.24	0.10	0.018	−0.27	0.12	0.020
MVPA				−0.02	0.00	<0.001	−0.02	0.01	0.002
Sleep time				−0.19	0.12	0.102	−0.18	0.14	0.178
Sports/PA clubs							0.01	0.12	0.966
Incentive for active transportation (bike)							0.56	0.40	0.179
Playground access during school hours							0.24	0.67	0.722
Access to cafeteria							0.64	0.91	0.491
Access to fast food outside school							0.13	0.30	0.680
Access to sports equipment outside school hours							0.11	0.29	0.708
Model summary									
Deviance statistic	3621.93			2332.03			2323.03		
Number of estimated parameters	3			13			19		

The reason for the stability in youth overweight/obesity prevalence observed in some countries is not clearly understood, and the same occurs in the present study. Tambalis *et al*. [45] hypothesized that the obesity prevalence may have reached a race and/or country specific ceiling, implying that children with predisposition toward obesity are now obese and obesity prevalence will not increase systematically. Similarly, Olds *et al*. [43] suggested that the environment in developed countries may be saturated with unhealthy food and options for sedentariness that children with a predisposition to becoming overweight have become overweight, and the remaining children may be resilient to obesogenic environments. In the Portuguese context, there is no published evidence to support our results. However, available data on physical activity in this age range (9–11 years), showed that approximately 36% of children achieve the daily recommended levels of MVPA [46]. Additionally, there has been an observed increase in organized sports participation during the last years among Portuguese youth (increases up to 149% from 1996 to 2010) [47] which may also contribute to this stabilization, given the widely reported relationship between overweight/obesity and physical activity.

Biological, behavioural and socio-demographic differences between normal-weight and overweight/obese children

Our results demonstrate that normal-weight and overweight/obese children are significantly different in their biological traits in that overweight/obese children are taller, heavier and ahead in their maturation. This is expected since previous research has shown that early maturing youth usually are taller, heavier and have higher BMI than their later maturing peers [48]. There is also strong evidence showing familial aggregation in BMI, where children with parents with high BMI, tend to have high BMI values [49,50]. For example, among the Chinese Han population, Hu *et al*. [49] reported that children with overweight/obese parents had higher BMI, and Fuentes *et al.* [50], studying Finnish family's aggregation in BMI showed that when one or both parents were obese, children were more likely to be in the highest quartile of BMI.

Differences between weight groups in nutritional habits, physical activity levels, sedentariness, sleep time and SES were not statistically significant. There is no clear evidence that children with healthy weight differ from those with excess weight in varied sets of behaviours and demographic characteristics [12,13]. For example, with regards to nutritional habits, Yannakoulia *et al.* [12] using a 3-day food record to study food patterns between normal-weight and overweight children, reported no significant differences between groups. Additionally, Garaulet *et al*. [9], investigating the association between energy and nutrient intake with the prevalence of overweight and obesity, found that overweight boys derived a greater percentage of their energy from fat and less from carbohydrates as compared to normal-weight boys, whereas overweight girls consumed less carbohydrates than normal-weight ones. Furthermore, Storey *et al*. [51], investigating non-overweight, overweight and obese adolescent diets, showed that non-overweight students consumed significantly more carbohydrate and fibre, significantly less fat and high calorie beverages, and had a higher frequency of consuming breakfast and snacks compared to their overweight or obese peers. Among Portuguese children, no significant differences between weight groups in healthy and unhealthy diet consumption were found, and children from both groups had equal access to healthy and unhealthy foods. One possible explanation for these results may be related to characteristics of Portuguese schools, which offer a nutrient balanced lunch for children. Moreover, they have food policies and controls regarding snacking and fast food, allowing children from both groups to have equal access to healthy and unhealthy food. In addition, schools also have a national program called "education for health" that aims to teach children about healthy choices and healthy living.

Physical activity and sedentary time/behaviour are usually correlated with weight status where normal-weight children are more active and less sedentary [52,53]. However, in our data no differences were found in physical activity levels or time spent in sedentary behaviour. These results are in line with data reported by Wang *et al.* [14] in Chinese children, as well as with those from Maier *et al*. [13]. This last investigation showed that all overweight or obese children reached the recommendation of spending one hour per day in sportive activities which was similar to normal-weight children. In Portugal, physical education is mandatory (twice a week); in addition, all schools offer free sports club participation, and most children (independent of their SES) have access to private sports club which may explain our results.

There is evidence that short sleep duration is consistently associated with concurrent and future obesity [54], where overweight/obese children tend to spend less time sleeping than children of normal-weight [55-57]. This relationship may be explained by alterations in glucose metabolism, up-regulation of appetite, and decreased energy expenditure [58]. However, our results differ from previous studies in that no differences were found in sleep time between normal-weight and overweight/obese children meaning that sleep deprivation may not be a major risk factor for development of overweight/obesity among Portuguese children. However, our sample included both overweight and obese children, and different results may be obtained by focusing on obesity alone.

In our study SES was determined by annual household income, and it was not found to be different between children of different weight status. Available data do not consistently show a clear effect size and direction in the association between weight status and SES. Previous research suggests that in developed countries, children of low and medium SES are more likely to be obese than those of high SES [59], while others report that higher SES is positively associated with overweight and obesity in Chinese children [60]. Given the distribution of SES in our sample, without any noticeable income inequality, the null results were expected.

Individual- and school-level correlates of BMI variation

The third purpose of the present study was to investigate the importance of individual- and school-level correlates on variation in children's BMI. At the child level, most variables were significant predictors. Similar to our results, sex differences in BMI have been consistently reported, with boys having higher BMI. For example, Meigen *et al.* [61] studied secular trends in German children's and adolescents' BMI and showed a greater increase in boys. Moreover, among Chinese children, Song *et al*. [62] identified sex disparities in BMI-for-age z-score during a 15-year period in which girls were stable whereas a linear increase was observed in boys. Additionally, Ogden *et al*. [63], found a significant increase in obesity prevalence between 1999–2000 and 2009–2010 in American boys as contrasted with girls. Similarly, Skinner and Skelton [64] also reported a stabilization, with a non-significant increase, in the prevalence of obesity among US children from 1999 to 2012. In the Portuguese context, results differ between studies [65], and there is no clear trend for boys having higher BMI than girls.

It is evident that more mature children have higher BMIs [48], as early maturing children are taller and heavier than on time or late maturing peers, where early maturity affects weight relatively more than height [48]. As such, our results were expected since those closer to their PHV (higher maturity offset positive values) had higher BMI values. Further, the relationship between parental and offspring BMI was significant, *i.e.*, children whose parents report high BMI tend also to have high BMI values, which is a consistent finding in previous twin and family studies [49,50]. For example, two Portuguese family studies by Souza *et al*. [66] and de Chaves *et al.* [67] reported the presence of genetic factors explaining from 30% to 50% of the total variance in different body composition phenotypes, which agrees with our findings that parental BMI is positively correlated with their child's BMI.

Nutritional habits and physical activity levels are two behavioural phenotypes usually associated with BMI [53]. Our results firstly showed that a healthy diet was negatively associated with BMI, meaning that children with a higher healthy diet score had lower BMI values. Among Mexican children [68], food patterns characterized by a high intake of sugary cereals, sweetened beverages, industrial snacks, cakes, whole milk, and sweets were associated with higher risk of overweight/obesity. Moreover, accordingly to Maffeis [69], there is some evidence that obese children show a certain preference for a fatty diet. On the other hand, non-overweight students tend to consume significantly more carbohydrate and fibre, and significantly less fat and high calorie beverages [51]. With regard to physical activity levels, our results are also in line with previous research. For example, Bingham *et al*. [3] found that performing at least 1 hour of moderate physical activity every day is a protective factor against childhood overweight/obesity in Portuguese children. Furthermore, Janssen *et al*. [70], in a review paper examining associations between overweight, dietary and physical activity patterns in youth concluded that increasing physical activity participation was a relevant strategy to prevent and treat overweight and obesity.

School predictors were not significantly associated with children's BMI. It is possible that the low number of schools (23 schools) and the similarity observed across Portuguese school environments may explain the null results, since only 2% of the BMI variation was attributed to school-level differences. Pallan *et al*. [71] investigated interschool variation in BMI z-scores and also found low intracluster correlations - between 0.9% and 4.2%. Further, they reported that the only school-level variable associated with BMI z-score was time spent in physical education classes (minutes/week).

The present study has several limitations. Firstly, the national studies included in the meta-analysis did not include information from the Autonomous Regions of Madeira and Azores, and this may bias the estimates to an unknown degree. However, one study reported a 29.1% prevalence of overweight/obesity in 11 year-old Azorean children [72], and another [73] showed a prevalence of 31.3% and 25.2% among Azorean 10 year-old girls and boys, respectively, which falls within the range of the confidence interval of the overall prevalence across the decade. Additionally, among 9–11 year-old Madeiran children [74], the prevalence of overweight/obesity ranged from 14.7% to 17.1% for boys and 13.3% to 16.9% for girls, which are lower than the prevalences reported in studies involving mainland samples. Secondly, although we used subjective methods to determine nutritional habits and sedentary behaviour, this is current practice in previous studies cited in the present article. Thirdly, we use self-reported parental height and weight which are suitable proxies for their actual values, and this is usual procedure in epidemiological research [75]. Fourthly, although ISCOLE utilized a validated questionnaire to obtain information concerning TV watching, it is possible that children underestimated their actual TV time. Fifthly, the present study combined overweight and obese children for analyses which may have attenuated effect sizes. Note that our total sample size is 686 children, and the distribution of BMI groups is 60.8% (n = 417) normal-weight, 28.3% (n = 194) overweight, and 10.9% (n = 75) obese. Separate analyses were not performed due to the lack of statistical power; further, our parameter estimates would be less precise, and our conclusions less reliable. Notwithstanding these limitations, the study has several merits: (1)

the presentation of a meta-analysis of the prevalence of overweight/obesity in the last decade in Portuguese children within an important developmental transition period; (2) the use of an objective method to assess physical activity; (3) inclusion of objective information regarding sleep time; (4) use of standard methods and highly reliable data; and (5) the use of multilevel modelling to capture the complexity of nested information at children and school levels.

CONCLUSIONS

In summary, the present study showed that in the last decade, overweight/obesity among 10 year old Portuguese children was stable. Normal-weight and overweight/obese children differed in their biological traits, but not in behavioural or sociodemographic traits. School characteristics did not seem to play important roles in BMI variation because they explain approximately 2% of the total variance. On the other hand, child-level variables are important because they explain 85% of the total variance attributable to variables at the subject level. Taken together, this information should be carefully considered by families, school authorities and teaching staff, paediatricians, and planners of intervention studies when designing more efficient strategies to combat the obesity epidemic.

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AUTHOR CONTRIBUTIONS

Thayse Natacha Gomes collected the data, undertook the data analysis and interpretation, and led the writing of the article. Fernanda K. dos Santos collected the data and contributed to drafting the paper. Michele Souza and Sara Pereira collected the data. Peter T. Katzmarzyk conceptualized and designed the study and contributed to drafting the paper. José A. R. Maia organized and supervised data collection and management, undertook the meta-analysis, and contributed to drafting the paper. All authors read and approved the final manuscript.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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Paper IV

"Fat-but-active": Does physical activity play a significant role in metabolic syndrome risk among children of different BMI categories?

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ABSTRACT

Objective: Physical inactivity and adiposity have relevant roles in metabolic syndrome (MS) expression. Given the high prevalence of overweight/obesity and low levels of physical activity (PA) among Portuguese children, this study intends to explore the idea of "fat-but-active" by analysing differences in MS risk factors across four distinct BMI and PA groups.

Methods: The sample comprises 389 Portuguese children from both sexes, aged 9-11 years. BMI was computed from measurements of height and weight, and PA was assessed by an accelerometer for 7 days. Moderate-to-vigorous PA (MVPA) was used to classify children as active (≥60 min/day) or inactive (<60 min/day). Children were divided in four groups: normal-weight and active, normal-weight and inactive, overweight and active, overweight and inactive. A continuous MS score (zMS) was computed from measures of waist circumference, glucose, triglycerides, HDL-cholesterol and mean arterial blood pressure.

Results: There was a high prevalence of overweight (51.9%) among children, and only 35.2% were physically active. In general, the overweight and inactive group had the worst metabolic profile, while the normal-weight active group had the best. Except for glucose, differences ($p<0.05$) were found in the metabolic indicators and for zMS across groups, but they are mainly observed between BMI groups, but not between MVPA groups.

Conclusion: MVPA did not attenuate the MS risk factors in the overweight group, given that MS indicators do not differ in children of the same group when taking into account their MVPA levels. This is a significant result for public health, where strategies related to nutritional education as well as promoting PA should be used to reduce adiposity in children and decrease MS risk factors in this population.

Keywords: physical activity; BMI; weight categories; metabolic risk; children; lifestyle; Portugal.

INTRODUCTION

Although the metabolic syndrome (MS), defined as a cluster of three or more metabolic abnormalities such as abdominal obesity, high blood pressure, dyslipidemia and dysglycemia [1], is mostly an adult health hazard, there is evidence showing that it is also a health problem in the paediatric population [2]. The rise in MS prevalence in children parallels the worldwide increases in childhood obesity. For example, Saland [3] reported that in North America, Asia and Europe, the prevalence of MS in obese youth ranges from 18% to 50%, but in normal-weight youth the prevalence is 1% or less. Similarly, a recent review of MS prevalence in children from North America, Latin America, Europe, Asia, and Australasia (aged 2-19 years) reported that, in the general population, values ranged from 1.2% to 22.6%, with rates up to 60% in overweight/obese youth, revealing their increased metabolic risk [2]. As obesity and metabolic abnormalities track well from childhood and adolescence to adulthood [4,5], it is of foremost importance to reduce these risk factors as early in life as possible to minimize the incidence of cardiovascular diseases later in life.

There is some evidence that physically active children have a better metabolic profile than the less active [6], suggesting an inverse association between physical activity and MS [7]. Higher physical activity levels are associated with greater insulin sensitivity [8] and HDL-cholesterol [9], and lower levels of blood pressure [9], adiposity [10] and triglycerides [11]. Moderate-to-vigorous physical activity (MVPA) levels and patterns in all likelihood play an important role as a mediational path to a healthier body weight, thus attenuating the risk of developing MS in youth [7,12]. Furthermore, a recent review addressing the role of physical activity and cardiorespiratory fitness concluded that both are separately and independently associated with metabolic risk factors in children and adolescents [7]. In addition, Brambilla et al [12] pointed out that physical activity influences metabolic risk factors within body weight categories, where normal-weight subjects with low physical activity levels have higher metabolic risk than more active ones, and obese subjects with high levels of physical activity have a lower metabolic risk than those who are inactive.

The association between physical fitness and MS in children has been investigated, within the concept of “fat but fit”, where the role of body fatness and fitness levels on metabolic risk profiles has been explored [13,14]; however, the results have not always been in the same direction. There is some evidence that high fitness attenuates the effects of fatness on cardiovascular risk [13]; on the other hand there is also evidence that fatness may attenuate the benefits of fitness on cardiovascular risk [15]. Regarding physical activity, even with its known role on reducing cardiovascular risk factors, there is no clear evidence if it can attenuate the effect of fatness on cardiovascular risk factors in youth [11,12].

Given the high prevalence of overweight and obesity in Portuguese youth [16,17], as well as the moderate prevalence of MS [18-22] and the low percentage of children and adolescents that achieve the MVPA guidelines [23], it seems relevant to explore the idea of “fat-but-active” by analysing differences in MS risk factors across distinct body mass index (BMI) and physical activity groups.

METHODS

Sample

The sample is from an ancillary study of the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE), a research project conducted in 12 countries from all major world regions [24]. The ISCOLE sample is a two-level random sample of children aged 9-11 years old, from 23 schools from the Northern region of Portugal. Children were from the 5th grade and a sample of ≈30-40 children was randomly selected from each school (50% for each gender). The ISCOLE study was approved by the physical education department, the school principal and the parental council, from each school, before starting data collection.

From the 777 5th grade Portuguese children aged 9-11years old (mean age 10.0±0.23) taking part in ISCOLE, an opportunistic sub-sample comprising 389 children (219 girls, 170 boys) accepted to participate in an ancillary study to understand the relationship between physical activity, overweight/obesity and MS. All parents or legal guardians provided written consent for their child to take part in the study. Data were collected from September 2011 to January 2013. The study protocol was approved by the University of Porto ethics committee, as well as by the schools' directorate councils.

Anthropometry

Height, weight, waist circumference and sitting height measures were taken according to standardized ISCOLE procedures [24]. Each child was measured twice and, when necessary, a third measurement was taken if the difference between the previous two was outside the permissible range for each measure and its replica (0.5 cm for height, sitting height and waist circumference, and 0.5 kg for weight). The mean value of each measured variable was used for analysis.

Body mass index was calculated using the standard formula [weight(kg)/height(m)^2], and subjects were classified in two groups [normal-weight, and overweight (including obese)] according to the cut-off points from the World Health Organization (WHO), based on BMI z-scores (normal-weight: <+1SD; overweight/obese: ≥+1SD) [25].

Physical activity

Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL), attached on the right waist, were used to monitor physical activity. The devices were activated at midnight on the first day and data were recorded with sampling rate of 80Hz. Children were instructed to wear the accelerometer for at least 7 days (including two weekend days), 24 hours/day. The delivery, reception and information about accelerometer use were made personally.

Accelerometer information was divided into daytime activities and nocturnal sleep time using an automated algorithm [26,27]. Non-wear time during the awake period was defined as any sequence of at least 20 consecutive minutes of zero activity counts [26,27]. To be eligible for this analysis, children had to have at least 4 days (with at least one weekend day) with a minimum of 10 hours of wear time per day; all 389 children fulfilled this condition.

Although the accelerometer provides information related to different physical activity phenotypes, only MVPA was used in the present study. Mean MVPA, according to the cut points defined by Evenson et al [28] from valid days, was used to classify children into two groups, according to WHO recommendations [29]: active children (mean MVPA ≥60 minutes) and inactive children (mean MVPA <60 minutes). In addition, the frequency of days children meet the MVPA guidelines among valid days were computed. MVPA was defined as greater than 574 activity counts using 15 second epochs, which has been shown to classify children accurately into physical activity intensity categories [28,30].

Biological maturity

An estimate of biological maturity was obtained using the maturity offset method [31]. Using information on sex, age, and individual physical growth (sitting height, stature and body weight) this method estimates in decimal years the time from peak height velocity (PHV). A positive maturity offset indicates the number of years a child is beyond PHV; a negative maturity offset indicates the number of years before PHV.

Metabolic syndrome

Metabolic syndrome indicators included waist circumference (WC), mean arterial blood pressure (MAP), fasting glucose (GLU), triglycerides (TRI), and high-density lipoprotein cholesterol (HDL-C). Resting systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured using a digital Omron sphygmomanometer (5 Series™ Upper Arm Blood Pressure Monitor – BP742, England) after subjects had been at rest for at least 10 minutes [32]. Three measurements were taken with a 3-minute interval between successive measurements, and the mean value was used. The MAP was calculated as:

[(SBP-DBP)/3 + DBP]. Finger-stick blood samples were collected after 10-12 hours of fasting and GLU, TRI and HDL-C were analyzed with a LDX point of care analyser [33]. The blood collection was performed in a private room, by a trained technician, and blood analysis was done immediately at the same place of blood collection. All of these procedures (blood collection and blood analysis) took about 5-10 minutes.

A standardized MS score (zMS) was computed using MAP, WC, GLU, TRI, and HDL-C, as previously described [34]. Using a stepwise regression analysis, all MS indicators were adjusted for sex and biological maturity, and the maturity- and sex-standardized residuals (z-score) from each one were obtained. The zMS was derived by summing the continuously distributed MS indicators, with the HDL-C-z-score been previously multiplied by -1 (given the negative relationship between MS and HDL-C). A lower zMS is indicative of a better metabolic profile. Notwithstanding the many cut-points proposed to define MS in children, there is still no consensus regarding which indicators should be used and their respective values, varying from one criteria to another. This fact, in association with the relatively low prevalence of MS in general in youth, justifies the use of the zMS. As pointed out by Eisenmann [34], this low relative prevalence would require large sample sizes for association studies, limiting the power to detect any relationship between exposure factors (such as physical activity, BMI) and a dichotomous outcome (having, or not, MS based on defined cut-points). Further, several studies have used the zMS to represent the MS clustering components, with significant associations with physical activity [6,35].

Data analysis

Physical activity and BMI categories were used to determine the frequency of children classified in four groups (normal-weight and active; normal-weight and inactive; overweight and active; and overweight and inactive). Weight status/physical activity groups' mean differences in each of the individual MS indicators, as well for zMS, were analysed with ANCOVA (Analysis of Covariance), controlling for sex and biological maturity. A

Bonferroni adjusted multiple-comparison test was also used. All analyses were done in SPSS 20, and the significance level was set $p<0.05$.

RESULTS

Descriptive statistics are presented in Table 1. The average accelerometer valid days was ≈7; children had recorded about 910 minutes/day of waking wear time, and more than 95% of the sample had physical activity information for 6 or more days. On average, children’s daily MVPA is 55.5 minutes, and only 35.2% of them reached the recommendations, considering the mean value. Taking into account the daily time spent in MVPA, only 2.3% of the sample achieved the WHO guidelines on all valid days, and 18.5% did not reach the guidelines on any day. More than half of the sample was classified as overweight or obese.

Table 1. Descriptive statistics

VARIABLE		Means±sd or Percentages (%)
BMI (kg/m^2)		19.7±3.3
Maturity Offset (years relative to PHV)		-1.9±0.9
MVPA (min/day)		55.5±21.4
Average accelerometer valid days		6.8±0.6
Average awake wear-time (min)		910.0±53.5
Waist circumference (cm)		66.8±8.5
HDL (mg/dl)		52.5±13.0
Triglycerides (mg/dl)		76.4±54.6
Glucose (mg/dl)		89.7±6.8
SBP (mmHg)		107.3±10.4
DBP (mmHg)		61.6±7.3
MAP (mmHg)		76.8±7.4
Metabolic Syndrome z Score		0.0±2.8
BMI Classification (%)		
	Normal-weight	48.1%
	Overweight/Obese	51.9%
Physical Activity Level (mean/day) (%)		
	≥60min MVPA/day	35.2%
	<60min MVPA/day	64.8%
Valid accelerometer days (%)		
	4	1.3%
	5	3.3%
	6	9.8%
	7	85.6%
MVPA daily compliance (%)		
	No valid day	18.5%
	All valid days	2.3%

The frequency of children classified into the four groups was as follows: 17.2% in the normal-weight and active group; 30.8% in the normal-weight and inactive group; 18.0% in the overweight and active group; and 33.9% in the overweight and inactive group. Table 2 shows the ANCOVA results for individual risk factors, as well as the zMS, across the four BMI-physical activity groups. In general, the overweight and inactive group had the worst metabolic profile, while the normal-weight and active group had the best. Except for glucose, statistically significant differences ($p<0.05$) were found for the metabolic indicators and for the zMS across groups. Further, a linear and significant trend was found for MS indicators (except for glucose) and zMS, across groups. In general, significant differences are mainly observed between BMI groups (normal-weight versus overweight/obese), but not within groups (active versus inactive).

DISCUSSION

It has been suggested that high levels of physical activity can attenuate the risk of MS in youth. We explored this idea across four distinct BMI and physical activity groups with Portuguese children aged 9-11 years, but firstly produced important descriptive epidemiology information. We showed that a high percentage of Portuguese 9-11-year old children are overweight or obese, and also a high percentage does not reach the recommended levels of daily MVPA. These are apprehensive results, as the overweight prevalence differs somewhat from those found worldwide. For example, in a review of the overweight/obesity prevalence among Brazilian youth (aged between 2-19 years), a prevalence of overweight/obesity up to 28.2% was found [36], and Janssen et al [37] reported prevalences higher than 15% in several countries. Previous Portuguese studies, using children aged 10-11 years [16,17], reported that the prevalence of overweight and obesity ranged between 18.7%-30.4% and 5.8%-28.0%, which is lower than what we reported in the present study, and these differences can be related to the use of different cut-points (WHO, CDC, IOTF), as well as differences in sample characteristics (since these two previous studies used samples from Portugal mainland, and in the present study the sample came from the Porto region).

In any case, these results highlight the actual overweight/obesity trends in paediatric populations, namely among Western and developed countries [37]. The main hypothesized reasons for these changes in obesity are related to changes in nutrition and physical activity habits observed in the last decades, which are influenced by social, environmental, interpersonal, community, governmental and biological characteristics [38], and that are closely related to increases in metabolic risk factors in children and adolescents [39,40].

Although there is some disagreement concerning a physical activity decline across time or the presence of low physical activity levels among youth [41,42], our results are in line with those from Baptista et al [23], where they show that ≈64% of Portuguese children aged 10-11 years did not achieve the guidelines for daily MVPA. Additionally, a recent review reported that about 80.3% of 13-15 year-old worldwide do not reach the 60 min/day of MVPA [43] This scenario (low levels of physical activity and high prevalence of children with overweight or obesity) are hypothesized to be the result of children's adverse lifestyles, mostly characterized by increases in sedentary activities and over-consumption of unhealthy, energy-dense food [44].

The main aim of the present study was to investigate if differences in MS risk factors could be linked to different BMI and physical activity groups: normal-weight and active; normal-weight and inactive; overweight and active; overweight and inactive. As expected, the worst metabolic profile was found among the overweight and inactive group which makes them an important group to target for interventions, while the best metabolic profile was from the normal-weight and active children, implying that children with high physical activity levels and low BMI are less prone to develop MS.

Table 2. Differences in metabolic risk indicators and zMS across BMI-physical activity groups, controlling for sex and biological maturity

	Normal-Weight		**Overweight**					
Variables	**Active (n=67)**	**Inactive (n=120)**	**Active (n=70)**	**Inactive (n=132)**	**F**	***p*-value**	**Pairwise comparisons**	**p-value for linear trend**
WC (cm)	62.0±0.6	62.7±0.5	70.3±0.6	71.3±0.5	75.11	<0.001	NA<OA; NA<OI; NI<OA; NI<OI	<0.001
HDL-C (mg/dl)	55.4±1.6	54.5±1.2	52.1±1.6	49.4±1.1	3.99	0.008	NA>OI;NI>OI	0.002
TRI (mg/dl)	62.9±7.0	67.1±5.3	81.2±6.9	89.2±4.9	4.10	0.007	NA<OI;NI<OI	0.001
GLU (mg/dl)	89.5±0.9	89.5±0.7	90.5±0.9	89.6±0.6	0.33	0.806	------	0.712
SBP (mmHg)	104.3±1.2	106.0±0.9	107.3±1.2	110.0±0.8	5.40	0.001	NA<OI; NI<OI	<0.001
DBP (mmHg)	59.3±0.9	61.3±0.7	61.4±0.9	63.1±0.7	3.50	0.016	NA<OI	0.003
MAP (mmHg)	74.3±0.9	76.2±0.7	76.7±0.9	78.7±0.6	5.44	0.001	NA<OI; NI<OI	<0.001
zMS	-1.7±0.3	-1.1±0.3	0.8±0.3	1.5±0.3	26.55	<0.001	NA<OA; NA<OI; NI<OA; NI<OI	<0.001

WC = waist circumference; HDL-C = high-density lipoprotein cholesterol; TRI = triglycerides; GLU = glucose; SBP = systolic blood pressure; DBP = diastolic blood pressure; MAP = mean arterial blood pressure; zMS = metabolic syndrome z score; NA = normal-weight and physically active group; NI = normal-weight and inactive group; OA = overweight and physically active group; OI = overweight and inactive group

It has been suggested that physical activity [45] and adiposity [46] have relevant roles on the development of MS risk factors in children, but it is not clear if these traits act separately and independently or in conjunction. Regarding physical activity, a recent review by Guinhouya et al [47] reports that the impact of physical activity on MS appeared to be either independent of other factors, or mediated by adiposity in youth. There is some evidence that children with higher physical activity levels have a better metabolic risk profile than their peers with lower physical activity levels [6,7]. In this context, Ekelund et al [9], concluded that physical activity is inversely associated with metabolic risk, independent of cardiorespiratory fitness and adiposity. According to the authors, this result has several implications for public health, since increasing overall physical activity, such as through play, active transport, and involvement in sport have beneficial effects on children’s metabolic risk profile.

If on one hand, physical activity prevents the development of metabolic risk, it seems clear that adiposity acts in the opposite fashion [48], being a stronger predictor of metabolic risk in children than physical activity [11] or even physical fitness [49]. Additionally, previous studies [2,3] have also reported that among overweight/obese youth the prevalence of MS is higher than in normal-weight youth, highlighting the adverse role of adiposity in the development of cardio-metabolic risk factors, namely MS.

Few studies have demonstrated that physical activity can attenuate the negative association between adiposity and metabolic risk, where high levels of physical activity improves the MS profile among obese subjects [12,50], increasing muscle mass and thus having a direct effect on metabolic function, changing cardiovascular risk factors. However, in our study we did not find a significant physical activity effect on zMS, and no difference was observed within BMI groups (active versus inactive), meaning that physical activity does not attenuate the MS risk among normal-weight children or even among those who are overweight. This result can be related to two distinct factors. Firstly, it can reinforce the notion that adiposity is more strongly correlated with MS risk factors than physical activity. The second possible explanation for this result can be mean value of 60 minutes of MVPA used to classify subjects as active and inactive, as this cut-point may not be sufficient for preventing the clustering of risk factors in children [6,51].

Notwithstanding the importance of the present results, this study has several limitations that must be discussed. Firstly, although we have a somewhat small sample size and limitations in the number of cases in the four groups, we had enough power to detect differences [a posteriori power analysis showed that, with the exception of glucose (observed power=0.24) and DBP (observed power=0.62), the power was higher than 0.80 in all other variables]; it should be noted that available research linking MS and objectively measured physical activity have similar sample sizes as ours [52]. Secondly, the sample comes from one Portuguese region, and the results do not reflect all Portuguese children. Thirdly, this study has a cross-sectional design which does not allow for clearly determining if the role of physical activity in attenuating the BMI effects on MS risk changes over time. Despite the limitations, this investigation has several important strengths: firstly, the use of an objective method to estimate children's physical activity; secondly, the use of the accelerometer for a whole week; thirdly, the use of rigorous standard methods and trained personnel to collect high reliable data, and finally to explore the idea of "fat-but-active", cross-tabulating our sample into four groups according to BMI and physical activity levels.

CONCLUSIONS

In summary, MVPA did not attenuate the MS risk factors in overweight Portuguese children, given that MS indicators do not differ in children of the same weight group when taking into account their MVPA levels. On the other hand, weight category seems to be important link to MS, since the metabolic profile found in the normal-weight and active group was significantly better than the one found in overweight and active children. In our view this may represent a significant result in terms of public health to enhance interventions associated with nutritional education and consumption of healthy food which may help the reduction of children MS risk factors. Further, since sustained and systematic

MVPA acts on weight control, namely through adiposity reduction, strategies to improve physical activity must also be considered.

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Paper V

"Active and strong": physical activity, strength and metabolic risk in children

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ABSTRACT

Background: This study explored the roles of physical activity (PA) and muscular strength (MS) on metabolic risk (MR).

Methods: Sample comprises 378 Portuguese children (213 girls; 9-11 years). PA was assessed by accelerometry, and moderate-to-vigorous PA was used to classify children as active (≥60 min/day) or inactive (<60min/day). Static MS was assessed as the ratio of handgrip strength/body weight and used to classify children as having high (≥P50) or low (<P50) MS. Children were classified into four groups: active and high MS, active and low MS, inactive and high MS, inactive and low MS. A continuous MR score, adjusted for sex and biological maturation, was computed using five MR indicators.

Results: In general, the inactive and low MS group had the worst metabolic profile, and the active and high MS group had the best. Significant differences were found within PA groups for MR score: children classified as "active and high MS" and "inactive and high MS" have better metabolic profiles than "active and low MS" and "inactive and low MS", respectively.

Conclusions: Muscular strength has a relevant role in attenuating the role of physical inactivity on MR in children; further, an increased benefit was identified in children with high PA and high MS.

Keywords: physical activity; muscular strength; metabolic risk; children

BACKGROUND

There is a worldwide increase in the prevalence of metabolic syndrome in the paediatric population,[1] suggesting that it is not an exclusive adult health hazard. Among North American, Asian and European youth, the prevalence of metabolic syndrome in overweight/obese children ranges from 18% to 50%.[2] Further, in a recent review of metabolic syndrome prevalence in children and adolescents aged 2-19 years, values ranged from 1.2% to 22.6% in the general population, with rates up to 60% in overweight/obese youth.[1] Notwithstanding the role of genetics on the development of metabolic syndrome risk factors,[3] behaviours and traits such as low physical activity (PA) and low physical fitness (PF) levels are major elements in their emergence/development, especially in youth.[4]

Recent research has shown a negative relationship between PA levels and metabolic risk (MR) factors.[4] Since high PA levels are associated with greater insulin sensitivity,[5] high HDL-cholesterol levels,[6] low blood pressure,[6] lower body weight,[7] and lower triglyceride concentration,[7] active children tend to have a better metabolic profiles than their less active peers.[8]

Similarly, PF levels have also been inversely associated with MR in children and adolescents.[4,9-12] However, most of these studies have focused on the relationship between cardiorespiratory fitness and MR indicators. Recent evidence reveals the importance of muscular fitness in the prevention of chronic disease, as well as in the development of MR factors in both adults[13,14] and youth.[10]

Since the joint relationship of PA and muscular strength (MS) and MR has not been extensively explored in previous research, and due to the fact that PA and PF levels in youth have declined in the last decades,[15-18] it is important to understand if a better MS profile can attenuate the negative impact of low PA on MR in youth. As such, the aim of the present study is to explore the joint roles of PA and MS on MR factors in children.

METHODS

Participants

The sample is from an ancillary study of the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE), a research project conducted at sites from 12 countries from all major world regions.[19] The present sample comprises children aged 9-11 years old (5th grade), from 23 schools from the Northern region of Portugal. In each school, approximately 30-40 children were randomly selected (50% of each sex). The ISCOLE study was approved by the physical education department, the school principal, the parental council, and the school's directorate council as well as by the University of Porto ethics committee before starting data collection.

From the 777 5th grade Portuguese children taking part in ISCOLE, an opportunistic sub-sample comprising 378 children (213 girls) accepted to participate in an ancillary study to understand the relationship between PA, PF and MR. All parents or legal guardians provided written consent for their child to take part in the study. Data were collected from September 2011 to January 2013.

Procedures

Anthropometry. Height, weight, waist circumference and sitting height measures were taken according to standardized ISCOLE procedures and instrumentation.[19] Each child was measured twice and, when necessary, a third measurement was taken if the difference between the previous two was outside the permissible range for each measure and its replica (0.5 cm for height, sitting height and waist circumference, and 0.5 kg for weight). The mean value of each measured variable was used for analysis.

Physical activity. Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL) were used to monitor PA. Children were instructed to wear the accelerometer for at least 7 days (including two weekend days), 24 hours/day. Accelerometer data were divided into daytime activities and nocturnal sleep time using an automated algorithm.[20,21] Non-wear time during the awake period was defined as any sequence of at least 20 consecutive minutes of zero activity counts.[20,21] To be eligible for this analysis, children had to have at least 4 days (with at least one weekend day) with a minimum of 10 hours of wear time per day. Moderate-to-vigorous PA (MVPA) was used in the present study, which was determined according to the cut-points defined by Evenson et al[22] from valid days. Children were classified into two groups, according to World Health Organization (WHO) recommendations:[23] active children (mean MVPA ≥60 min/day) and inactive children (mean MVPA <60 min/day).

Static muscular strength. Static MS was assessed by the handgrip strength test, which was measured in a standing position using a digital hand dynamometer (Takei TKK 5401, Tokyo, Japan). Children were asked to squeeze the dynamometer with maximal force, using the preferred hand, holding it away from the body and with the arm extended. The result was recorded in kg. To remove the body size effect, grip strength was divided by body weight, and the ratio handgrip/weight was used in all analysis. Further, the 50th sex-specific percentile (P50) of this sample of the handgrip/weight ratio distribution was used to classify children as having high (≥P50) and low (<P50) MS.

Biological maturation. An estimate of biological maturity was obtained by the maturity offset method.[24] Using information on sex, age, and individual physical growth (sitting height, stature and body mass) this method estimates, in decimal years, the time from peak height velocity (PHV). A positive (+) maturity offset indicates the number of years a child is beyond PHV; a negative (-) maturity offset indicates the number of years before PHV.

Metabolic risk. Metabolic risk indicators included waist circumference, systolic (SBP) and diastolic (DBP) blood pressure, fasting glucose, triglycerides, and high-density lipoprotein cholesterol (HDL-C). Resting SBP and DBP were measured using a digital Omron sphygmomanometer (5 Series™ Upper Arm Blood Pressure Monitor – BP742, England) after subjects had been at rest for at least 10 minutes.[25] Three measurements were taken with a 3-minute interval between successive measurements, and the mean value was used. The mean arterial blood pressure (MAP) was calculated as: [(SBP-DBP)/3 + DBP]. Finger-stick blood samples were collected after 10-12 hours of fasting, and glucose, triglycerides and HDL-C were analysed with a Cholestech LDX (Cholestech Corporation, Hayward, CA, USA) point of care analyser[26].

A standardized MR score (zMR) was computed using MAP, waist circumference, glucose, triglycerides, and HDL-C, as previously advocated.[27] Prior to the computation of the zMR, all MR markers were adjusted for sex and biological maturity using a stepwise regression analysis, and the sum of the continuously distributed MR indicators (with the HDL-z-score multiplied by -1) was computed. A lower zMR is indicative of a better MR profile as previously suggested.[27]

Data analysis

Physical activity and MS categories were used to determine the frequency of children classified in four groups as follows: active and high MS (20.9%); active and low MS (14.6%); inactive and high MS (29.1%); inactive and low MS (35.4%). PA-MS group mean differences in each of the individual MR indicators, as well for zMR, were analysed with ANOVA (Analysis of Variance). A Bonferroni adjusted multiple-comparison test was also used. All analyses were done in SPSS 20, and the significance level was set at 5%.

RESULTS

Descriptive statistics are presented in Table 1. On average, children's daily MVPA is 55.5 minutes, and only 35.4% of them reached the international daily recommendations of ≥ 60 min/day. Their mean handgrip strength is 16.7 kg, and, in general, children are, on average, 2 years before their PHV.

Table 1. Descriptive statistics (means±standard deviation or percentage)

VARIABLES	Means±sd or Percentages (%)
Weight (kg)	41.1±9.2
Maturity Offset (years relative to PHV)	-1.9±0.9
MVPA (min/day)	55.5±21.3
Handgrip (kg)	16.7±3.6
Handgrip/weight ratio (kg/kg)	0.4±0.1
Accelerometer valid days	6.8±0.6
Waist circumference (cm)	66.9±8.5
HDL (mg/dl)	52.6±13.1
Triglycerides (mg/dl)	76.5±55.0
Glucose (mg/dl)	89.8±6.9
MAP (mmHg)	76.8±7.4
PA Level (mean/day) (%)	
≥60 min MVPA/day	35.4%
<60 min MVPA/day	64.6%

Table 2 shows the ANOVA results for individual risk factors, as well as the zMR, across the four PA-MS groups. In general, the inactive and low MS group had the worst metabolic profile, while the active and high MS group had the best. Except for glucose, statistically significant differences ($p<0.05$) were found for the MR indicators and for the zMR across groups. Further, significant differences were found within PA groups for zMR, where children classified as "active and high MS" and "inactive and high MS" have better metabolic profiles than their peers classified as "active and low MS" and "inactive and low MS", respectively.

Table 2: Differences in MR Indicators and zMR across PA-MS groups (mean±standard error)

	Active		**Inactive**				
Variables	**High MS (n=79)**	**Low MS (n=55)**	**High MS (n=110)**	**Low MS (n=134)**	**F**	***p*-value**	**Pairwise comparisons**
WC (cm)	63.0±0.7	71.3±1.2	62.7±0.6	70.8±0.7	36.810	<0.001	AHMS<ALMS; AHMS<ILMS; ALMS>IHMS; IHMS<ILMS
HDL-C (mg/dl)	56.5±1.6	52.0±1.4	55.2±1.3	48.3±1.0	8.989	<0.001	AHMS>ILMS; IHMS>ILMS
TRI (mg/dl)	64.3±3.6	78.1±10.9	71.9±3.3	86.9±5.5	3.162	0.025	AHMS<ILMS
GLU (mg/dl)	89.7±0.9	90.8±0.8	88.9±0.6	90.1±0.6	1.159	0.325	------
MAP (mmHg)	74.2±0.8	77.2±0.9	75.9±0.6	78.8±0.7	7.087	<0.001	AHMS<ILMS; IHMS<ILMS
ZMR	-0.9±0.3	0.3±0.3	-0.7±0.2	0.9±0.3	11.329	<0.001	AHMS<ALMS*; AHMS<ILMS; IHMS<ILMS

*p=0.051

WC = waist circumference; HDL-C = high-density lipoprotein cholesterol TRI = triglycerides; GLU = glucose; MAP = mean arterial blood pressure; zMR = metabolic risk z score; AHMS = active and high muscular strength group; ALMS = active and low muscular strength group; IHMS = inactive and high muscular strength group; ILMS = inactive and low muscular strength group

DISCUSSION

The present study examined the joint roles of MS and PA on MR factors, in a sample of Portuguese children aged 9-11 years. A first discussion note from our results is that a high percentage of children in this sample do not comply with the daily PA international recommendations. Only 35.4% (95%CI 31-40) of the children are, on average, engaged in at least 60 min/day of MVPA. These results are similar to another recent Portuguese study that reported that approximately 64% of 10-11 year-old children did not comply with the international guideline for daily MVPA.[28] Further, a recent literature review revealed that about 80.3% of youth aged 13-15 yrs worldwide do not achieve 60 min/day of MVPA.[15]

The main aim of the present study was to investigate if differences in MR exist across PA and MS groups in children. The "active and high MS" group showed the best metabolic profile, whereas the "inactive and low MS" group showed the worst, highlighting the need for more efficient intervention strategies targeting both physical inactivity and low muscular fitness. Given that these behaviours tend to track from childhood and adolescence to adulthood, interventions during childhood could have lifelong health impacts.[29-33]

Previous studies have identified independent roles of PA and MS on MR and adiposity[4-8,34-37] in youth. It is generally suggested that, on average, children with higher PA levels tend to have better metabolic profiles than those with lower levels,[4-8] and this association is independent of other factors, such as PF or adiposity.[6,34] Similarly, the independent role of MS in the prevention of chronic disease in adults,[38] as well as its positive role on MR indicators in both men and women have been shown.[13,39] Research in children has also reported a significant association between high MS and a better metabolic profile.[35] Additionally, the relationship between muscular and cardiorespiratory fitness with MR in the HELENA study[36] indicated that both PF indicators were independently linked to adolescents' MR. In a similar vein, the ACFIES study[37] also reported similar results, but the association between MS (assessed by the handgrip test) and MR was stronger and more consistent than the association between cardiorespiratory fitness and MR. The authors suggested that the cardiometabolic benefits of MS might be related to its association with body composition (a positive association with percentage of lean mass, and inverse association with percentage of body fat); however, this is not clear and future studies should be conducted to better clarify this association, namely using mediator and moderator variables in more complex statistical analysis where specific hypotheses can be tested. Furthermore, a relationship between MS in adolescence and premature mortality and cardiovascular disease in adulthood[40,41] has reinforced the notion of low MS "as an emerging risk factor for major causes of death in young adulthood" (p. 5).[40]

Limitations

Some limitations of the present study should be acknowledged. First, given the relatively small sample size, we were not able to investigate the relationship of MR and MS within BMI categories which could provide further insights that could be very helpful in designing/implementing interventions. However, a power analyses done in G*Power 3.0.10[42] given the following conditions: effect size = 0.20, α = 0.05, power = 0.90, and 4 groups, yielded an estimated total sample size of 360 subjects which is fairly close to our sample (n=378). Second, the cross-sectional nature of the present study does not allow any causation statements, and future research should use longitudinal research designs. Third, the sample comes from one Portuguese region, and the results cannot be generalized to all Portuguese children; however, in data not shown here, similar results were found in some characteristics between our sample and others from previous studies, namely in the prevalence of overweight/obesity[43] and socioeconomic status.[44] However, despite these limitations, this study has significant merits, such as the use of an objective method to measure children's PA over an entire week; the use of a robust method to measure MS; and the use of standardized methods and trained investigators to collect all data.

Conclusion

In conclusion, PA and MS are important predictors of low MR in children, meaning that children with high levels of PA and MS are less likely to have poor MR profiles. Further, high MS plays a relevant role in attenuating the effects of physical inactivity, suggesting that more attention should be given to MS development during childhood and adolescence. Taken together, our results are important in terms of child health when school strategies to reduce MR in youth are developed.

IMPLICATIONS FOR SCHOOL HEALTH

The findings of the present study, pointing out the important roles of PA and MS in reducing MR, are very important to be used by school administrators and physical education teachers when designing and implementing strategies (curricula program, interventions, policies) to increase PA and PF (namely MS) levels among children and youth. Since children spend about half of their awake time at school, this important educational context has to promote more efficient, joyful and highly successful activities linked to active and healthy lifestyles, aiming also to reduce comorbidities linked with physical inactivity and low PF, especially MR factors which are known to carry over their undesirable effects later in life.

Although children spend most of their school daily time in sitting positions attending classes, school administrators and physical education teachers have to provide them with varied and ample opportunities to be physically active and develop their MS, during recess time, where they can play freely, and in physical education classes, where they are engaged in structured physical activities/play, and organized sport planned at their level have important roles. Thus, school policies should provide students diversified playground areas, allowing their access to distinct/"challenging" equipment, to getting them involved in activities that could improve their PA and MS in a non-structured way. In addition, physical education classes should be planned with the purpose of also leading the child to adopt an active lifestyle, and also increasing their PF levels, improving, indirectly, their metabolic profile.

The role of school environment in reducing MR via increasing PA and PF in children is mandatory. In this context, it is known, for example, that overweight/obese children tend to have a worse metabolic profile, and also spend less time in MVPA and are more unfit than their normal-weight peers. Thus, improving PA and PF among this high-risk group could reduce their adverse MR profile. Further, it seems to us that improving MS among overweight/obese children could be much easier than improving their overall PA levels. As such, the role of the school context in decreasing the MR in this group is highlighted, especially during physical education classes, where teachers could implement diversified strategies to improve the strength of children and, as a consequence, contribute to lowering their risk of developing undesirable MR.

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Paper VI

Relationship between sedentariness and moderate-to-vigorous physical activity in youth. A multivariate multilevel study

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Submitted

ABSTRACT

This study aimed to jointly analyse moderate-to-vigorous physical activity (MVPA) and sedentariness (Sed), and their correlates, in children within their school contexts, using a multivariate multilevel approach in a sample of 499 Portuguese children (284 girls) from 23 schools. Overall, schools explained a small amount of the total variance in both MVPA (5.6%) and Sed (3.6%), and a correlation coefficient of -0.46 was found between MVPA and Sed at the child level. Number of siblings and family socioeconomic status (SES) were significantly associated with both Sed (SES: β=3.921±1.329; siblings: β=-6.658±3.119) and MPVA (SES: β=-1.599±0.422; siblings: β=2.971±0.980), but with opposite signs; further, a significant negative association was observed for both of them with sleep time (Sed: β=-22.866±2.935; MVPA: β=-1.868±0.923). BMI (β=-4.744±1.911) and sex (β=22.314±3.508) were only associated with MVPA. None of the school covariates were statistically significant in their joint effects to simultaneously explain Sed and MVPA. These results suggest that although MVPA and Sed may be different constructs, they are correlated and this should be taken into account when designing strategies to reduce children's Sed and increase their MVPA. In addition, the small effect of the school context on this relationship highlights the important roles of child and family characteristics.

Keywords: physical activity; sedentariness; children; multivariate multilevel approach

INTRODUCTION

It is now widely accepted that moderate-to-vigorous physical activity (MVPA) positively affects child and adolescent health (Steele et al., 2008). In contrast, sedentariness (Sed) is considered a potential risk factor for chronic disease in youth, as it is linked to the increased prevalence of overweight/obesity (King et al., 2011) and metabolic risk factors (Steele et al., 2008).

Traditionally, sedentary behaviour was often considered as a lack of physical activity (PA) (Marshall & Welk, 2008). As such, PA and Sed are sometimes viewed as opposite sides of the same coin (Owen et al., 2000), supporting the “displacement hypothesis” which assumes that inactive behaviours may prevent more active ones (Biddle et al., 2004; British Heart Foundation, 2000). However, it has more recently been suggested that sedentary behaviour is a different construct from that of PA, i.e., they are two independent traits not expressing themselves in different “parts” of a continuum (Biddle et al., 2004; Katzmarzyk, 2010; Pate et al., 2011); apparently both have similar (King et al., 2011) or different correlates (Van der Horst et al., 2007) with distinct effect sizes and direction (Marshall et al., 2002; Pate et al., 2011). Further, these two behaviours can coexist (Owen et al., 2000), reinforcing the idea that the relationship between PA and Sed is far from being clear, with previous studies reporting conflicting results (Biddle et al., 2004; Marshall et al., 2002; Tammelin et al., 2007).

Both Sed and PA are linked with a wide array of biological, social, behavioural and environmental correlates (King et al., 2011; Van der Horst et al., 2007). In this context, it has been suggested that sex (King et al., 2011), weight status (King et al., 2011), maturity (Sherar et al., 2007), socioeconomic status (SES) (Pate et al., 2011; Van der Horst et al., 2007), siblings’ influence (Atkin et al., 2013), and sleep time (Stone et al., 2013) are related to time spent in these behaviours. Additionally, since children spend a considerable amount of their awake time at school, the school context is thought to provide many opportunities for children to be physically active and to avoid extended sedentary periods given its social and physical environments, such as campus size or playground areas, sports equipment and sporting facilities, recess periods, lunch breaks, and physical education classes (Cradock et al., 2007; Sallis et al., 2001; Wechsler et al., 2000). Taken together, both child-level and school characteristics may explain a proportion of the variance in PA and Sed at the population level. Therefore, the aim of this study was to use a multivariate multilevel approach (Snijders & Bosker, 2012), to jointly analyse MVPA and Sed as well as their correlates in children within their school contexts.

METHODS

Sample

The sample of the present study comes from the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE), a research project conducted in 12 countries from all major regions of the world, with the purpose to determine the relationship between lifestyle behaviours and obesity, and to investigate the influence of higher-order characteristics such as behavioural settings, and physical, social and policy environments, on the observed relationships within and between countries (Katzmarzyk et al., 2013).

A total of 777 Portuguese children, aged 9-11 years, from 23 schools from the North of Portugal, were enrolled in the ISCOLE project. In each school, after the project was approved by the physical education department, school principal and parental council, all 5th grade students were invited to take part in ISCOLE, and those that were aged 9-11 years were eligible to participate after informed consent was obtained from parents or legal guardians. From those, approximately 30 to 40 children were randomly selected per school (50% of each sex), and the response rate was 95.7%. After the inclusion criteria (children with valid accelerometer data and with no missing information on all other variables used in this study), the final sample comprised 499 children (284 girls).

Data were collected from September 2011 to January 2013, and all assessments were done during a full week per school. The study protocol was approved by the University of Porto ethics committee, as well as by each school's directorate councils.

Outcome variables

Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL, USA) were used to monitor PA and sedentary time. Children were asked to wear the accelerometer at their waist on an elasticized belt placed on the right mid-axillary line 24 hours/day, for at least 7 days, including two weekend days. To be eligible for this study, children had to have at least 4 days (from which at least one of them was a weekend day) with a minimum of 10 hours of awake wear time per day. Accelerometer information was divided into daytime activities and nocturnal sleep time using an automated algorithm (Barreira et al., 2014; Tudor-Locke et al., 2014). Non-wear time during the awake period was defined as any sequence of at least 20 consecutive minutes of zero activity counts (Barreira et al., 2014).

Using cut-points advocated by Evenson et al. (2008), different activity phenotypes were determined. For the present study, mean MVPA and mean sedentary time were used, which were defined as being greater than or equal to 574 activity counts and less than or equal to 25 activity counts using 15 second epochs, respectively.

Predictor variables

Child level

Anthropometry

Height, sitting height, and weight were measured according to standardized ISCOLE procedures and instrumentation (Katzmarzyk et al., 2013). Each child was measured twice, and a third measurement was taken if the difference between the previous two was outside the permissible range for each measure and its replica (0.5 cm for height and sitting height; 0.5 kg for weight). The mean value of each variable was used for the analysis.

The body mass index (BMI) was computed with the standardized formula [weight (kg)/height (m)^2], and using the World Health Organization (WHO) (de Onis et al., 2007) cut-points, children were classified as normal-weight or overweight/obese.

Biological maturation

Using information on sex, age, and physical growth characteristics (sitting height, leg length, stature, and body mass), an estimate of biological maturity was computed using the maturity offset method (Mirwald et al., 2002). This method uses specific regression equations for boys and girls and estimates, in decimal years, the timing to peak height velocity (PHV) occurrence. A positive maturity offset expresses the number of years a child is beyond PHV; while a negative maturity offset means the number of years a child is before the PHV; a value of zero indicates that a child is experiencing his/her PHV.

Sleep time

Using the accelerometer information, the nocturnal sleep time for each participant was determined using a novel and fully-automated algorithm specifically developed for use in ISCOLE and epidemiological studies employing a 24-hour waist-worn accelerometer protocol in children (Barreira et al., 2014; Tudor-Locke et al., 2014). The mean sleep time across all days was used in the analysis

Family characteristics

Basic demographic characteristics were obtained via a questionnaire, completed by parents or legal guardians [ISCOLE Demographic and Family Health Questionnaire (Katzmarzyk et al., 2013)], which also provides information regarding ethnicity, family health and socioeconomic factors. For the present study, we only used information about family SES and number of siblings. Socioeconomic status was determined by asking parents about the family annual income. The answer was split in eight categories, ranging from <€6000 to ≥42000, where category 1 represents the lowest family income, and the category 8 represents the highest; for data analysis, these categories were centred at category 4. Parents were also asked about family size, informing the number of siblings the child enrolled in the project has.

School level

Information concerning to school environment was obtained via a questionnaire [ISCOLE School Environment Questionnaire (Katzmarzyk et al., 2013)] completed by the physical education teacher or school principal. For the present study the following school environment factors were considered: the school size (defined according to the number of students); the percentage of students participating in school sports or PA clubs; students' access to outdoor facilities outside of school hours (0=no; 1=yes); and students' access to playground equipment during school hours (0=no; 1=yes).

Data analysis

Exploratory analysis, descriptive statistics, and t-test to compare differences between boys and girls were done on SPSS 21. Snijders & Bosker (2012) have described several advantages of the multivariate approach (i.e., analysing multiple dependent variables jointly): (1) the possibility to obtain conclusions about the correlations between the dependent variables (in our case Sed and MVPA), notably the extent to which the unexplained correlations depend on child-level traits and school context variables; (2) a statistical increase in efficiency for tests of specific effects on any single dependent variable given the multivariate nature of the data structure; (3) the ability to test whether the effect of any exploratory variable is similar or different across the multiple dependent variables; (4) avoid the capitalization on chance due to systematic tests being carried out on single dependent variables, which does not happen when a multivariate analysis is done (Tabachnick & Fidell, 2007). Given the multivariate and clustered structure of our data, as shown in Fig 1, we utilized a three-level model: the two dependent variables Sed and MVPA are at level-1, children are at level-2, and schools are at level-3.

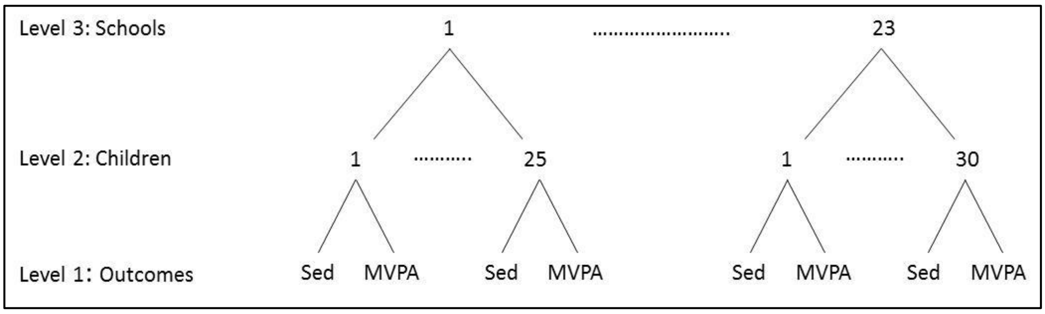

Fig 1. Multivariate multilevel structure of outcome variables (Sed and MVPA) at level 1, nested within children at level 2, nested within schools at level 3

All analyses were done in SuperMix software v.1 (Hedeker et al., 2008), using full maximum likelihood estimation; the explicit formulation of this type of model and estimation details are described elsewhere (Snijders & Bosker, 2012). As has been previously advocated (Snijders & Bosker, 2012), we used a three step approach allowing only intercepts to randomly vary. In step 1 (baseline model, Model 1) the variances and covariances were jointly estimated for Sed and MVPA. These estimates allow us to calculate two important pieces of information: (i) how much of the total variation in Sed and MVPA is explained at the child and school levels; (ii) what is the size and direction of the correlations between Sed and MVPA for children within schools, and between schools. In step 2 (Model 2) we included child-level predictors, and in step 3 (Model 3) we added school context variables. Final decisions about the best fitting solution were made according to deviance and corresponding Likelihood-ratio (LR) tests in nested models of increasing complexity. A more complex model fits better than a previous one if the difference in their respective deviances is statistically significant by the LR test. This is done using a Chi-square statistic with degrees of freedom equal to the difference in estimated parameters between the two models. For ease of presentation, we display the results for Models 1 to 3 according to Snijders & Bosker (2012).

RESULTS

Tables 1 and 2 present descriptive statistics (mean±SD and percentage) for child- and school-level variables. There is a high frequency (44.1%) of overweight/obese children. On average, children are about 2 years from their estimated PHV, and girls are more mature than boys. Children sleep about 8 hours·day^{-1}, and have 1 sibling on average. A total of 49.5% of the children live in a family with an annual family income below €12000.

More than 90% of the schools in the sample have children engaged in sports participation or PA clubs. About half of the schools allow the students to have access to sports equipment outside school hours, but only 8.7% of them allow their students to have access to playground equipment during school hours. The mean number of students per school is 782±309, ranging from 239 to 1589.

Table 1. Descriptive statistics for variables at the child level (level 1)

Variables		Child-level variables (mean±SD or percentage)		
		Boys	Girls	Total
BMI (kg·m^{-2})		19.24±3.29	19.32±3.37	19.28±3.33
Maturity Offset (years to PHV)		-2.77±0.42*	-1.24±0.51	-1.90±0.90
Sleep time (hours·day^{-1})		8.22±0.95	8.36±0.82	8.30±0.88
Number of siblings		0.97±0.79	0.98±0.86	0.97±0.83
BMI (classification)				
	Normal-weight	51.6%	59.2%	55.9%
	Overweight/obese	48.4%	40.8%	44.1%
SES				
	<€6000	14.9%	22.2%	19.0%
	€6000 - €11999	32.6%	28.9%	30.5%
	€12000 - €17999	20.0%	17.3%	18.4%
	€18000 - €23999	11.2%	9.5%	10.2%
	€24000 - €29999	7.9%	7.4%	7.6%
	€30000 - €35999	5.1%	5.6%	5.4%
	€36000 - €41999	2.8%	3.9%	3.4%
	≥42000	5.6%	5.3%	5.4%

*p<0.05

The multivariate multilevel modelling results are presented in Tables 3, 4 and 5. Model 1 (see Table 3) is the starting point and shows that, on average, these 10 year old children have 560±4 min·day^{-1} of Sed, together with 56±1 min·day^{-1} of MVPA. Schools explain a small amount of the total variation of both Sed (3.6%), and MVPA (5.6%). The major portion of the variance in Sed and MVPA is at the child level. Further, at the child level (within-schools), the covariance between Sed and MVPA is significant (σ_{CL}=-595.32±65.87, $p<0.05$) which translates to a negative correlation coefficient within child as ρ_{CL}=-0.46.

Table 2. Descriptive statistics for variables at the school level (level 2)

		School-level variables (mean±SD or percentage)
Number of students (mean±SD)		782±309
Children participation in sports or PA clubs		
	Not available	4.3%
	Less than 10%	4.3%
	10-24%	34.8%
	25-49%	13%
	≥50%	43.5%
Student's access to equipment outside school hours		
	No	47.8%
	Yes	52.2%
Student's access to playground equipment during school hour		
	No	91.3%
	Yes	8.7%

Table 3. Model 1 main results [parameter estimates, standard errors (SE) and deviance] for both Sed and MVPA

	Sed		MVPA		Covariance	
	Estimate	SE	Estimate	SE	Estimate	SE
			Fixed Effects			
Intercept	559.647*	3.789	55.682*	1.452		
			Random Effects			
Between-schools (school level)						
Variance	144.588	96.377	26.682**	14.203		
Covariance					-22.752	28.245
Correlation					-0.366	
Within-schools (children level)						
Variance	3841.645*	248.815	446.050*	28.897		
Covariance					-595.317*	65.867
Correlation					-0.455	
Deviance				9912.6458		

*p<0.05; **p<0.10
School explained variance for Sed = [144.588/(144.588+3841.645)] = 3.6%; and for MVPA = [26.682/(26.682+446.050)] = 5.6%

Model 2 (Table 4) includes a set of child characteristics (BMI category, maturity offset, SES, number of siblings, sex and sleep time). Number of siblings and SES were associated with both Sed and MPVA, but with opposite signs: children with higher SES are more sedentary (β=3.921±1.329, p=0.03) but spend less time in MVPA (β=-1.599±0.422, p<0.001); those with more siblings tend to be less sedentary (β=-6.658±3.119, p=0.03) but are more physically active (β=2.971±0.980, p=0.002). Children who sleep more spend less time in both Sed (β=-22.866±2.935, p<0.001) and MVPA (β=-1.868±0.923, p=0.043). BMI and sex were only associated with MVPA such that overweight/obese children are less physically active than their normal-weight peers (β=-4.744±1.911, p=0.013), and boys are more physically active than girls (β=22.314±3.508, p<0.001). The child level negative correlation (within schools) remained unchanged relative to the previous model ρ_{CL}=-0.45. This model fits better than the previous one given the reduction in deviance (Δ=267.3843, 12 df, p<0.001).

Table 4. Model 2 [parameter estimates, standard errors (SE) and deviance] including child-level predictors for both Sed and MVPA

	Sed		MVPA		Covariance	
	Estimate	SE	Estimate	SE	Estimate	SE
			Fixed Effects			
Intercept	597.375*	10.660	44.804*	3.474		
Overweight/obese	-4.830	6.080	-4.744*	1.911		
Maturity offset	9.348	6.337	0.507	1.993		
Socioeconomic status	3.921*	1.329	-1.599*	0.422		
Number of siblings	-6.658*	3.119	2.971*	0.980		
Sex	-17.625	11.187	22.314*	3.508		
Sleep time	-22.866*	2.935	-1.868*	0.923		
			Random Effects			
Between-schools (school level)						
Variance	119.781	79.881	30.786*	13.566		
Covariance					-40.718	26.702
Correlation					-0.671	
Within-schools (child level)						
Variance	3196.876*	207.036	310.504*	20.121		
Covariance					-448.753*	50.059
Correlation					-0.450	
Deviance				9645.2615		

*p<0.05

The final model (Model 3) included school level covariates (Table 5). None of the covariates were observed to be statistically significant in their joint effects to simultaneously explain Sed and MVPA ($p>0.05$). Further, the small non-significant reduction in deviance (Δ=6.4047, 8 df, p=0.112) confirmed that the previous model (Model 2), being more parsimonious, fits the data better.

Table 5. Model 3 [parameter estimates, standard errors (SE) and deviance] including child- and school-level predictors for both Sed and MVPA

	Sed		MVPA		Covariance	
	Estimate	SE	Estimate	SE	Estimate	SE
			Fixed Effects			
Intercept	607.198*	19.202	40.807*	6.832		
BMI categories	-4.941	6.080	-4.730*	1.906		
Maturity offset	9.031	6.369	0.388	1.994		
Socioeconomic status	3.992*	1.342	-1.636*	0.423		
Number of siblings	-6.684*	3.135	3.002*	0.981		
Sex	-18.128	11.226	22.092*	3.510		
Sleep time	-22.760*	2.943	-1.845*	0.920		
Students involvement in PA or sports Clubs	-1.083	3.198	-0.654	1.195		
Students access to outdoor facilities	-6.014	8.023	1.244	3.024		
Students access to playground equipment	5.461	12.527	-5.024	4.749		
School size	-0.004	0.012	0.007	0.005		
			Random Effects			
Between-schools (school level)						
Variance	108.143	76.353	22.286*	11.008		
Covariance					-33.052	23.486
Correlation					-0.673	
Within-schools (child level)						
Variance	3197.661*	207.078	310.275*	20.103		
Covariance					-449.748*	50.062
Correlation					-0.452	
Deviance				9638.8568		

*p<0.05

DISCUSSION

Using a multivariate multilevel approach, the present study aimed to jointly analyse MVPA and Sed as well as their correlates in Portuguese children within their school contexts.

The major finding of the present study is the negative correlation between MVPA and Sed at the child level, suggesting that children with higher levels of MVPA tend to also have lower levels of Sed even after adjusting for a set of covariates at the child and school levels. The relationship between MVPA and Sed is not always clear, and although they have been seen as two different individual traits (Biddle et al., 2004; Katzmarzyk, 2010; Pate et al., 2011), the co-existence of both in children can occur, suggesting that highly physically active children can be less sedentary than their low physically active peers. A recent review by Leech et al (2014), aiming to identify clustering patterns of diet, PA and sedentary behaviour in children/adolescents reported that both cluster patterns, high PA/high sedentary behaviour and high PA/low sedentary behaviour, are observed in youth, supporting Owens et al.'s (2000) account that PA and Sed can sometimes compete with each other and can sometimes coexist. Further, Marshall et al. (2002) studied the interrelationship between sedentary behaviours and PA (expressed in METs) in youth aged 11-15 years, and reported a positive correlation between these traits. It was also found that, among boys, 40% of them self-reported more sedentary behaviour, and 94% of those more sedentary boys participated in double the recommended PA guidelines necessary for health. On the contrary, Tammelin et al (2007) reported a negative association between PA and TV viewing and computer use, where the highest proportion of physically inactive individuals were observed among those who watched TV for at least 4 hours·day^{-1} (14% in girls, 13% in boys). These authors also reported that those who spent at least 4 hours·day^{-1} watching TV (prevalence ratio: 1.5 for boys and 2.5 for girls) or using the computer/playing video games for more than 2 hours·day^{-1} (prevalence ratio: 1.4 for boys and 2.2 for girls) were more likely to be physically inactive than those who watched TV or used the computer/played video games for less than 1 hour·day^{-1}, respectively. Using PA and Sed pattern data from a sample of 10-11 year-old children, Jago et al (2010) identified three distinct clusters that were subjectively labelled as "high active/low sedentary", "low active/moderate sedentary", and "high active/high sedentary", highlighting that the presence of one behaviour does not exclude the presence of the other. The negative correlation found in the present study can be related to the fact that, since the 24h day is finite, children that spend higher amounts of the time in MVPA or Sed have less available time to engage in the other behaviour. Further, this result also highlights that both Sed and MVPA should be considered when strategy planning and program implementation are prepared to reduce health risks in children.

When MVPA and Sed are investigated as single/individual outcomes they sometimes share similar correlates. For example, SES, number of siblings, and sleep time were common correlates in our multivariate model. With respect to SES and number of siblings, the associations were in opposite directions: children with higher SES are more sedentary and less physically active, while those with more siblings are less sedentary but more physically active. There seems to be no consensus about the association between SES and PA or Sed (Pate et al., 2011; Tandon et al., 2012; Van der Horst et al., 2007) For example, Tandon et al (2012), studying children aged 6-11 years, reported that lower SES home environments provide more opportunities for sedentary behaviour and fewer for PA, while Atkin et al (2013) found an increase in Sed time over one year among children (mean age: 10.2 years) with higher SES. The results for PA are similarly diverse. For example, Newton et al (2011) found that lower SES African American boys spend more time in MVPA compared to middle SES African American and lower SES Caucasian children. Among British adolescents aged 11-12 years, followed for 5 years, Brodersen et al (2007) reported no significant association between boys' PA and SES, but among girls those from lower SES were less active. Our findings indicate that children from higher income families tend to have greater Sed and lower MVPA. This can be related to the fact that these children are more likely to have more access to media entertainment (such as TV, computer, games etc) for use during their leisure time, reducing available time to spend in PA and thus increasing time spent in sedentary activities.

The role of siblings as a correlate of children's PA and sedentary behaviours is not clear. For example, Tandon et al. (2012) reported that children, on average, tend to spend more days per week watching TV/DVD's with siblings than participating in PA. However, it was also demonstrated that the presence of more children at home is highly related to more MVPA overall and at home, as well as more overall Sed at home but less screen time (Tandon et al., 2014). As to the results of our study, a possible explanation may be that those children with less sedentary/more active siblings tend also share such behaviours, becoming less sedentary/more active too.

It has generally been suggested that sleep time, PA and Sed are linked, such that children with more sleep time, or those with a better sleep efficacy, tend to be more physically active and less sedentary (Stone et al., 2013). Our results, however, are not in total agreement with this trend. Our results indicate that children who sleep more are less sedentary, in accordance with previous data; however, these children also tend to be less active. Apparently this is in line with previous findings in children reported by Pesonen et al. (2011), where for each standard deviation (SD) unit increase in PA during the day, a decrease of 0.30 SD in sleep duration, and a decrease by 0.16 SD in sleep efficiency was observed. These results can be possibly explained by the fact that, as reported above, the hours of the day are finite and sleeping more reduces the time available to engage in others activities such as MVPA or Sed (Olds et al., 2012). In addition, it has also been suggested that PA promotes better sleep rather than more sleeping hours per se (Pesonen et al., 2011).

The simultaneous modelling of MVPA and Sed also revealed that, when their predictors were jointly analysed, sex and BMI are only associated with MVPA, but not with Sed. Sex differences in both Sed and MVPA have been previously reported, where boys are generally more physically active and less sedentary than girls (Van der Horst et al., 2007), mostly explained by the fact that during leisure time girls largely tend to get involved in more sedentary activities (such as reading, listening to music, socializing with peers), while boys tend to engage in more physical activities (such as sports or competitive games) (Blatchford et al., 2003). Our results, however, found no significant sex differences for Sed when simultaneously analysed with MVPA, meaning that the relationship between Sed and sex may be different when Sed is analysed as a single outcome and when the analysis is done examining the co-occurrence of MVPA and Sed at the child level.

Studies focusing on the association between PA and weight status in children generally report a negative relationship between excess weight and PA (King et al., 2011), probably because overweight/obese children have poorer motor skill proficiency (Okely et al., 2004), lower physical perceived competence and peer acceptability in sports (Seabra et al., 2013) than their normal-weight peers, and this cluster of “factors” may contribute to their lower MVPA levels. However, we did not find a significant association between BMI and Sed, indicating that probably the most relevant variable related to excess weight is PA. This also suggests that both normal-weight and overweight children tend to spend similar amounts of time in sedentary activities.

At the school level, no statistically significant covariance/correlation was observed because of we have only 23 schools; however, the correlation coefficient is -0.67, and suggests that MVPA and Sed tend to cluster at the school level. In other words, more active and less sedentary children tend to study at the same school. Although we were not able to identify any specific school-level covariate that could be related to the co-occurrence of these behaviours, results indicate that such covariates exist. It is possible that the number of schools (23 schools) and the reduced variance observed across schools, among school-level covariates, may be a possible explanatory factor for the non-significant covariance/correlation at the school level. In any case, our results highlights the role of biological and demographical characteristics in regulating the clustering of PA and Sed in children, as well as the relevance of the school environment as an important agent to promote active and healthy lifestyles among children, promoting policies to teach children how be active and reduce their Sed during their leisure time. An example of the school as a venue to increase children PA can be found in the study by Straker and Abbott (2013), where they report that 10-12 year old children from New Zealand and Australia spend 36 minutes less in Sed, 56 minutes less in light activity, 4 minutes more in moderate activity, and 2 minutes more in vigorous activity at school when compared to non-school time.

This paper has several limitations which should be discussed. First, the cross-sectional design does not allow for causal interpretation of the results. Second, the fact that the correlation between MVPA and Sed was only significant at the level of the child, and not at the level of the school, may indicate that individual variables may be more important than school context predictors. However, it is a well-known fact in multilevel modelling that first level predictors usually explain more of the total variance in any outcome variable than higher-order levels. If we had more schools we would probably be more likely to show that the correlation between these two traits, MVPA and Sed, was also significant at this level. Third, the sample comes from only one Portuguese region, and results cannot be generalized to other areas, notwithstanding the fact that, in data not shown here, overweight/obesity prevalence (Sardinha et al., 2011) and SES distribution (Fundação Francisco Manuel dos Santos, 2013) compares with previous studies. There are also several strengths in this study that deserve to be mentioned: (1) the use of a multivariate multilevel analysis to identify the joint "determinants" of MVPA and Sed at individual and school-levels, as well as their variance and covariance; (2) the use of an objective method to estimate MVPA and Sed, and (3) the use of highly standardized methods for data collection and highly reliable information.

In conclusion, this study showed that there is a negative correlation between MVPA and Sed, i.e., that although MVPA and Sed are two different traits that can occur in children, they are correlated. A relevant set of covariates at the child-level was significantly associated with both MVPA and Sed, such as number of siblings, SES, and sleep time, reinforcing the suggestion that they have similar correlates, although not always equal in magnitude and direction. In addition, the significant role of BMI and sex was only reported for MVPA. School environment explains a small amount of the total variation of both Sed (3.6%), and MVPA (5.6%), and none of the school-level covariates were statistically significant in explain simultaneously the variation in Sed and MVPA; however the correlation coefficient at school-level of -0.67 suggests that Sed and MVPA tend to cluster at the school level. This is relevant information that should not be neglected when planning strategies to promote PA and reduce Sed in youth.

PERSPECTIVE

Findings of the present study point out the significant negative correlation between Sed and MVPA at both child- and school-levels. This is relevant information that should be carefully considered by families, school authorities, teaching staff, paediatricians, and planners of intervention studies when designing and implementing strategies to reduce Sed and increase MVPA in youth. Since the role of biological and family characteristics on these variables, active home environments, where children are reinforced to be active in their leisure time, and also where sedentary activities are not the most prominent activities, can be helpful in the promotion of health behaviours. There is also the need for parental involvement in controlling the amount of time that children spend in sedentary behaviours (such as screen time), promoting adequate environmental settings allowing for better sleep quantity and quality, and encouraging their engagement in PA, alone or in peers (i.e., with friends or siblings). Furthermore, schools have a very important role to play in promoting/disseminating active and healthy lifestyles in children, helping them in health choices related to their activities during leisure time; further, they are expected to provide ample opportunities to be joyfully active during the time spent at school, namely during recess time and physical education classes.

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Paper VII

Are BMI and sedentariness correlated? A multilevel study in children

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ABSTRACT

The purpose of this research was to investigate the relationship between body mass index (BMI) and sedentariness (Sed) in children, and to examine the influence of child and school correlates on their variation. Sample comprises 580 children (337 girls, 9-11 years). Sedentariness was assessed with accelerometer and BMI was computed. Child- and school-level covariates were analysed using multilevel models. No significant correlation between Sed and BMI was found. School context explains 5% and 1.5% of the total variance in Sed and BMI, respectively. At the child level, only moderate-to-vigorous physical activity was associated with both Sed (β=-0.02±0.002) and BMI (β=-0.005±0.002). Sleep time is related to Sed (β=-0.42±0.04), while sex (β=1.97±0.13), biological maturity (β=1.25±0.07), media in bedroom (β=0.26±0.08), and healthy (β=-0.09±0.03) and unhealthy (β=-0.07±0.04) diet scores were associated with BMI. Any school-level covariate was related to BMI, but access to cafeteria (β=-0.97±=0.25), playground equipment (β=-0.67±0.20), and restaurants (β=0.16±0.08) were related to Sed. In conclusion, Sed and BMI were not correlated. Further, they have different correlates, while children's traits seem to play more relevant roles on their differences in Sed and BMI than the school milieu. This information should be taken into account when strategies to reduce Sed and BMI are implemented.

Keywords: BMI; sedentariness; children; multilevel analysis

INTRODUCTION

Drastic increases in prevalence of youth overweight/obesity [1] and associated co-morbidities [2] have been reported in past decades. A systematic review [1] from 1980 to 2013 indicated that prevalence of childhood overweight/obesity in developed countries increased from 16.2% to 22.6% and from 16.9% to 23.8% in boys and girls, respectively; in developing countries this increase was from 8.1% to 12.9% in boys, and from 8.4% to 13.4% in girls. Although still high, this rise appears to be stabilizing in some countries [3]. Since overweight/obesity tends to track into adulthood [4], increasing risk for cardiovascular diseases and co-morbidities [5], excess weight in youth remains a major public health problem.

Sedentariness (Sed), an emerging potential risk factor for obesity [6], has been linked to increasing body mass index (BMI) [7]. Compelling evidence suggests that youth spend large proportions of awake time in sedentary behaviours [8]. Several interventions indicate that decreasing sedentary time can contribute to weight reduction in children [9-11].

BMI and Sed seemingly share common biological and environmental correlates [12-14], with that sex [13,15], physical activity levels [16-18], maturity status [19], sleep time [20,21], family environment (namely siblings' influence and availability of electronic media) [22-24], nutritional habits [25,26], and time spent at school [27,28] are possible determinants of Sed and/or BMI. Since children spend most of their awake time at school, school has a relevant role in children's Sed and BMI variation, offering ample opportunities for physical activity [29] and healthy eating [30].

The available evidence supports the hypothesis that Sed and BMI are correlated in youth. The present study aims to (1) study the relationship between BMI and Sed in Portuguese children, and (2) investigate the importance of child and school correlates in BMI and Sed variation.

METHODS

Sample

The present study sample is from the International Study of Childhood Obesity, Lifestyle and the Environment (ISCOLE), research conducted in 12 countries from all major regions of the world, to determine the relationship between lifestyle behaviours and obesity and to investigate the influence of factors including behavioural settings and physical, social and policy environments, on observed relationships within and between countries [31].

Portuguese children (n=777), aged 9-11 years, from 23 schools in northern Portugal, enrolled in the ISCOLE project. The study protocol was approved by the University of Porto ethics committee and by each school's directorate councils. After the project was approved by the Physical Education Department, school Principal and Parental Council, 5th grade students, 9-11 years old) were invited to participate [31]. Written informed consent was obtained from by parents or legal guardians, and assent from children. Approximately 30 to 40 children were randomly selected per school (50% of each sex); response rate was 95.7%.

Outcome variables

Our first outcome variable was Sed, objectively measured using Actigraph GT3X+ accelerometers (ActiGraph, Pensacola, FL, USA). Children were instructed to wear the accelerometer at their waist on an elasticized belt placed on the right mid-axillary line 24 hours/day, for at least 7 days. For eligibility, children should have at least 4 days (from which at least one was a weekend day) with a minimum of 10 hours of daily wear time. Accelerometer information was divided into daytime activities and nocturnal sleep time using an algorithm [32,33]. Non-wear time during awake periods was defined as any sequence of at least 20 consecutive minutes of zero activity counts [33]. Mean week sedentary time (minutes·d^{-1}) was defined as ≤25 activity counts per 15 second epoch [34].

Our second outcome variable was BMI. Height and weight were measured according to standardized ISCOLE procedures and instrumentation [31]; BMI was computed using the standardized formula [weight(kg)/height(m)^2].

Predictor variables

<u>Child level</u>

Predictor variables at child level included biological maturation, moderate-to-vigorous physical activity (MVPA), sleep time, dietary patterns, and family demographics. Biological maturation was computed using maturity offset sex-specific regression equations [35]. This method estimates the timing to peak height velocity (PHV) occurrence in decimal years. A positive (+) maturity offset is the number of years a child is beyond PHV; while a negative (-) maturity offset is the number of years a child is before the PHV; a zero value indicates that a child is experiencing his/her PHV.

Weekly mean MVPA (minutes·d^{-1}) and sleep time (hours·d^{-1}) were estimated by accelerometry. MVPA was defined as activities greater or equal to 574 activity counts per 15 second epoch [34]; nocturnal sleep time for participants was determined using a novel and fully-automated algorithm specifically developed for use in ISCOLE and epidemiological studies employing 24-hour waist-worn accelerometry [32,33].

Information on dietary patterns was obtained from questionnaires [31], completed on the same day as anthropometric measures. The questionnaire included frequency of consumption of different foods in a typical week, specifically fruits, vegetables, sweets, soft drinks, other foods and fast food consumption. Reported frequencies were converted into portions/week for principal components analysis. The component scores computed for each subject for two dietary patterns were standardized to ensure normality. These two patterns were designated as “unhealthy” (positive loadings for hamburgers, soft drink, fried food, etc.) and “healthy” (positive loadings for vegetables, fruits, etc.).

School level

The ISCOLE School Environment Questionnaire [31] was completed by the physical education teacher or school principal. Environmental aspects of the school were considered: students' access to (i) outdoor facilities outside of school hours; (ii) playground equipment during school hours; (iii) cafeteria at school; (iv) fast food restaurants close to schools; and (v) drink vending machines. In addition, information regarding the daily time each child spends at school, and a "mean week school time" was calculated.

Data were collected from September 2011 to January 2013, and all assessments were completed with a single week at each school by trained personnel from the Kinanthropometry Laboratory of the Faculty of Sport (University of Porto) following certification by the ISCOLE Coordinating Center. After the inclusion criteria (children with accelerometer valid data for ≥4 days and with no missing information in any other variables), the final sample included 580 children (337 girls).

Data analysis

For exploratory and descriptive statistics SPSS 21 was used. To address our first aim, a multivariate multilevel model as suggested by Goldstein [36] and Snijders and Bosker [37], for situations where at least two outcome variables are used at the same time – in our case Sed and BMI, was employed. These and others [38,39] have outlined the main reasons for such an approach, specifically increased power, reduction in capitalization by chance with systematic testing, and ability to model correlations (at school and child levels) between outcome variables.

If the correlations at child and school level in our joint outcomes are not statistically significant, then a univariate outcome analysis (BMI separated from Sed) using child and school covariates within a multilevel approach was conducted. A three step analytical approach was used [37,38]. In step 1 (our Null model) only an intercept term and variances (at school and child levels) were estimated. These variance estimates allowed us to calculate the total variation in Sed and in BMI explained at the school level. In step 2 (Model 1), child-level predictors were included, and in step 3 (Model 2) school context variables were added. Final decisions were made according to deviance and corresponding Likelihood-ratio (LR) tests in nested models of increasing complexity. A more complex model fits better than a previous one if the difference in their respective deviances is statistically significant by the LR test, according to a Chi-square statistic with degrees of freedom equal to the difference in estimated parameters between the two models. Since BMI is kg/m^2, and Sed is $min \cdot day^{-1}$, to make all fixed effects coefficients as well as variances comparable of BMI with Sed, we standardized BMI and Sed, i.e., expressed in z-scores. All multilevel (multivariate and univariate) analyses were done in SuperMix software v.1 [40]. Explicit formulation of these types of models and estimation details are described elsewhere [36,37].

RESULTS

Descriptive statistics (mean±SD and percentage) are presented in Table 1. On average, children spent about 9½ hours/day or about 1/3 of the day in sedentary activities, < one hour/day in MVPA, and slept about 8 $hours \cdot night^{-1}$. They were about 2 years from PHV and spent ≈ 7 hours/day at school. Significant differences among sexes were observed for sedentary time, MVPA, maturation, sleep, and dietary pattern scores. Compared to boys, girls are ahead in their maturity status, spend more time in sedentary activities and sleep, and less time on MVPA, have higher healthy diet scores and lower unhealthy diet scores. Children had about 1 sibling on average; almost 81% had media in their bedroom.

Almost 96% of schools reported that students had access to a cafeteria, and 69.6% reported that students had access to fast food restaurants close to school. However, only 26.1% reported that students had access to vending machines. About half of the schools allowed students access to sports equipment outside school hours, only 8.7% of them allowed students access to playground equipment during school hours.

On average, a 10 year old child spends 560±4 minutes·day^{-1} in sedentary behaviour and has a BMI of 19.3±0.2 kg·m^{-2} (Table 2). Schools explain small amounts of the total variation of Sed (4.9%) and BMI (1.5%). The major portion of the total variance in Sed and BMI is at the child level. Interestingly, at the child level, the covariance between Sed and BMI is not significant ($\sigma_{SED,\ BMI}$=-1.91±0.83, p=0.826), translating to a small and negative correlation coefficient ($\rho_{SED,\ BMI}$ =-0.01).

Since no statistically significant covariance/correlation was observed between Sed and BMI at the child level, a separate multilevel analysis for each outcome variable using values transformed into z-scores was computed. Predictor variables from both child and school levels were used.

The null model (Table 3) estimated that school-level effects from intraclass correlation coefficients were 0.050 and 0.015 for Sed and BMI, respectively; 5% of variance in Sed and 1.5% of variance in BMI among children are explained by school effects; 95% of Sed variance and 98.5% of BMI variance are explained by child-level characteristics.

Results from Model 1 (table 3) indicate that sleep time was significantly associated with Sed - children who sleep more were less sedentary (β=-0.43, SE=0.04, p<0.001); no significant effect of sleep time on BMI was observed. Sex, biological maturity, bedroom media availability and dietary patterns correlated with BMI, but not Sed: boys (β=1.97, SE=0.13, p<0.001), children advanced in biological maturity (β=1.25, SE=0.07, p<0.001), and those with media in bedroom (β=0.26, SE=0.08, p=0.002) had higher BMI than girls, than later maturing children, and those without media in bedroom, respectively. Children with a higher healthy diet score (β=-0.09, SE=0.03, p=0.009) tended to have lower BMI, also true for unhealthy diet score, but marginally significant (β=-0.07, SE=0.04, p=0.054). MVPA was the only variable significantly correlated with both Sed and BMI, where higher MVPA involvement was negatively related to both (for Sed: β=-0.02, SE=0.002, p<0.001; for BMI: β=-0.004, SE=0.002, p=0.011); the effect of MVPA is greater in reduction of Sed compared to BMI.

Table 1. Descriptive statistics for variables at the child and school level (level 1)

Child-level variables (mean±SD or percentage)	Boys	Girls	Total
BMI (kg·m^{-2})	19.2±3.3	19.3±3.4	19.3±3.4
Sedentary mean (min·day^{-1})	544±66*	572±59	560±63
MVPA (min·day^{-1})	67±23*	46±15	55±22
Maturity Offset (years to PHV)	-2.78±0.42*	-1.25±0.53	-1.89±0.90
Sleep time (hours·day^{-1})	8.2±0.9*	8.4±0.8	8.3±0.9
Number of siblings	0.98±0.80	0.96±0.82	0.96±0.82
Healthy diet score	-0.09±1.00*	0.11±0.99	0.03±1.00
Unhealthy diet score	0.21±1.21*	-0.21±0.68	-0.04±0.96
Time at school (hours·day^{-1})	6.8±0.4	6.8±0.4	6.8±0.4
Media at bedroom			
No	18.1%	19.9%	19.1%
Yes	81.9%	80.1%	80.9%
School-level variables (percentage)			
Student's access to cafeteria			
No			4.3%
Yes			95.7
Student's access to fast food restaurant			
No			30.4%
Yes			69.6%
Student's access to drink vending machines			
No			73.9%
Yes			26.1%
Student's access to playground equipment during school hours			
No			91.3%
Yes			8.7%
Student's access to sports equipment outside school hours			
No			47.8%
Yes			52.2%

*p<0.05

Table 2. Null model main results [parameter estimates, standard errors (SE) and deviance] for both Sed and BMI

	Sed		BMI		Covariance	
	Estimate	SE	Estimate	SE	Estimate	SE
Fixed Effects						
Intercept	559.68*	3.93	19.26*	0.16		
Random Effects						
School level						
Variance	196.81*	104.24	0.17^{ns}	0.18		
Covariance (σ_{SL})					1.68^{ns}	3.07
Correlation (ρ_{SL})					0.29^{ns}	
Child level						
Variance	3818.05*	228.71	11.03*	0.66		
Covariance (σ_{CL})					-1.91^{ns}	0.83
Correlation (ρ_{CL})					-0.01^{ns}	
Deviance			9493.6327			

*p<0.05; ns=not significant
School explained variance for Sed = [196.807/(196.807+3818.052)] = 4.9%; and for BMI = [0.166/(0.166+11.029)] = 1.5%

Table 3. Results summary of modelling Sed and BMI: estimates[¥](standard-errors)

	Sed			BMI		
Parameters	Null Model	Model 1	Model 2	Null Model	Model 1	Model 2
			Fixed Effects			
Intercept	-0.008[§](0.062)	0.083(0.14)	1.05(0.28)*	-0.004[‡](0.05)	1.34(0.13)*	1.11(0.27)*
Sex		0.06(0.13)	0.05(0.13)		1.97(0.13)*	1.97(0.13)*
Siblings		-0.03(0.04)	-0.02(0.04)		-0.01(0.04)	-0.02(0.04)
Maturity offset		0.07(0.07)	0.07(0.07)		1.25(0.07)*	1.25(0.07)*
Media bedroom		0.01(0.09)	0.01(0.09)		0.26(0.08)*	0.26(0.08)*
MVPA		-0.02(0.002)*	-0.02(0.002)*		-0.004(0.002)*	-0.005(0.002)*
Sleep time		-0.43(0.04)*	-0.42(0.04)*		-0.02(0.04)	-0.02(0.04)
Healthy diet score		0.02(0.03)	0.03(0.03)		-0.09(0.03)*	-0.09(0.03)*
Unhealthy diet score		-0.03(0.04)	-0.03(0.04)		-0.07(0.04)**	-0.07(0.04)*
Time at school		-0.06(0.1)	-0.08(0.09)		0.13(0.08)**	0.09(0.09)
Access to cafeteria			-0.97(0.25)*			0.24(0.24)
Access to fast food restaurant			0.16(0.08)*			0.04(0.08)
Access to drink vending machine			-0.11(0.09)			0.02(0.09)
Access to playground equipment			-0.67(0.20)*			0.08(0.19)
Access to sport equipment outside school hour			-0.12(0.08)			-0.06(0.08)
			Random Effects			
Between-school variance	0.05(0.03)	0.03(0.02)	0.007(0.007)	0.02(0.02)	0.003(0.007)	0.003(0.007)
Within-school (child) variance	0.95(0.06)	0.63(0.04)	0.63(0.04)	0.98(0.06)	0.60(0.04)	0.59(0.04)
			Model Summary			
Deviance	1634.4114	1396.9434	1374.8328	1643.2667	1342.2034	1340.4157
Number of estimated parameters	3	12	17	3	12	17

¥: all estimates are expressed as z-scores; §=559.68 $min \cdot day^{-1}$ in the original metric; ‡=19.26 $kg \cdot m^{-2}$ in the original metric; *p<0.05; **p<0.10

Difference in deviance from the null model to model 1 (for sedentariness: Δ=237.468, 9 degrees of freedom; for BMI: Δ=301.0633, 9 degrees of freedom), was statistically significant, therefore model 1 fits better than the null model in explaining variance of each outcome variable. Further, from model 1 the proportion of variance in Sed and BMI explained by children's characteristics was 48% for Sed, 80% for BMI.

Children with access to a cafeteria (β=-0.97, SE=0.25, p<0.001), and those with access to playground equipment (β=-0.67, SE=0.20, p<0.001) were less sedentary; children with access to fast food restaurants close to school (β=0.16, SE=0.08, p=0.037) were more sedentary. However, none of the school-level covariates was related to BMI.

Difference in deviance from model 1 to model 2 (for Sed: Δ= 22.1015, 5 df; for BMI: Δ=1.7866, 5 df), was only significant for Sed. For Sed, model 2 fits better than model 1; but for BMI, model 1 is the best. Approximately 86% of the original 5% of the between-school variance in Sed was attributed to students having access to cafeteria, playground equipment during school hours and fast food restaurants close to school.

DISCUSSION

The relationship between BMI and Sed in a sample of Portuguese children, and child- and school-level correlates using both multivariate and univariate multilevel models were explored. Results from the multivariate model were that correlations between BMI and Sed were low and not statistically significant. Relationships between Sed and BMI reported in youth suggest that greater Sed (especially time watching TV) is associated with increased body weight [7]. However, in our study, this relationship was not found. Carandente et al [41] found positive correlations between BMI and time spent in sedentary activities, and between time spent in sedentariness and food consumption; the more hours 8-10-year old children spent watching TV, the more likely they consumed snacks and beverages. Watching TV and other sedentary behaviours may stimulate eating and increased energy intake, thus affecting body weight [11]. Results suggest that the relationship between body weight and Sed may be indirect, with eating behaviours and energy intake mediating this relationship.

Our univariate multilevel analysis indicated that from all of the child-level predictors, only MVPA was significantly associated with both Sed and BMI. Previous research offered similar results regarding BMI; at least 1 hour of daily MVPA was shown to be protective against overweight/obesity in Portuguese children [16]. Others [17] found that increased MVPA reduced BMI z-score over 3 years in overweight/obese children. In examining relationships between overweight, diet and physical activity patterns in youth, Jansen et al [42] concluded that increasing involvement in physical activity is a relevant strategy to prevent/treat excess weight. Previous studies have reported similar MVPA and Sed results [18], and as Epstein and Roemmich [43] note, engagement in physical activity “usually involves choosing exercise over a concurrent and powerful competing sedentary behaviour” (p. 103). Since we found stronger effects of MVPA in reducing Sed, than BMI, other factors beyond behavioural, namely genetic, have key roles in increasing/decreasing BMI [44].

Sleep time is relevant to children’s health [20,21,45,46]. However, associations between sleep and Sed are inconclusive [20,46]. No relationship was found in Taiwanese adolescents [46] between time spent watching TV/using the computer and sleep; however, among Belgium students [20] those spending more time in sedentary activities spent less time in bed on weekdays. In our data the negative association between sleep and Sed suggests that since hours of the day are limited, sleeping more reduces sedentary time available [47]. Although there is evidence of an association between short sleep duration and obesity in youth [21,45], this was not observed in our study.

Other predictors at the child level (except number of siblings and time spent at school), sex, biological maturation, bedroom media availability and diet were only associated with BMI, similarly with previous reports. Among Spanish youth [48], significantly higher prevalence of obesity was observed for boys.

Also, the relationship between BMI and maturity status seems to be clear, more mature children tend to be taller and heavier [19].

Screen time is frequently researched in children, usually negatively associated with BMI [22]; and having a TV in the bedroom increases risk of overweight/obesity [23,24]. Our results reinforce this – on average, children with bedroom media had higher BMI than those without. While unclear, several mechanisms possibly contribute: reduced energy expenditure while watching TV, increased dietary intake through snacking, and increased exposure to media promoting food consumption. Higher healthy diet pattern scores were negatively associated with BMI. Differences in diet are not always observed between normal-weight and overweight children [25,26]. However, normal-weight children may consume significantly more carbohydrate and fibre, and less fat and high calorie beverages compared to overweight peers [49].

The school environment, widely recognized as promoting active and health lifestyles among children/adolescents, offers mandatory/extracurricular activities and policies reducing sedentary time [29]. In the present study, 5% of variance in Sed was explained by school environment. Playground areas [29] provide opportunities to engage in physical activity during recess and reduce sedentary time. Promoting healthy eating and access to healthy food at school affect weight gain and control [30]. However, no relationship between students' access to cafeteria or fast food close to school and BMI was found, but a significant association between these two predictors and Sed was observed.

School "effects" on children's BMI was only 1.5%. Pallan et al [50] similarly found low intraclass correlations (i.e., variance attributable to school effects), varying from 0.9% to 4.2%. Low school-level variation in Sed has previously been reported [51]. Relatively low numbers of schools (23), and low variance across Portuguese school environments, may explain low school effects on Sed, and very low results on BMI.

There are study limitations: 1) its cross-sectional nature does not allow cause-effect interpretations; 2) the number of schools and low variance of schools' contextual characteristics limit identification of school-level traits on

BMI and Sed; 3) one Portuguese regional population limits generalization, although overweight/obesity prevalence [52] and socioeconomic status distribution [53] compares with previous studies; 4) diet as a mediated variable in the relationship between Sed and BMI in multilevel models was not used; 5) since all children had at least 10 hours of awake wear time, with mean accelerometer use value of 15.17±0.86 hours, we did not adjust physical activity and Sed for wear time because this effect is not significant (data not shown). Strengths were: 1) multivariate multilevel analysis identifying relationships between Sed and BMI, with multilevel modelling to understand complex nested information at child and school levels; 2) objective methods to estimate Sed, MVPA and sleep time; 3) Standardized data collection methods; 4) reliable child- and school-level information.

CONCLUSIONS

Sed and BMI were not significantly correlated, but MVPA was significantly associated with both. However, correlations were different and should be considered since strategies to reduce Sed or BMI may act through different pathways. Low variance at school level for both BMI and Sed reinforce suggestions that although children spend considerable awake time at school, individual variables play more relevant roles in differences between Sed and BMI than school. School policies promoting active and healthy habits play important roles in reducing sedentary time, making wise nutritional choices, and controlling body weight.

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CHAPTER IV

General Overview and Conclusions

GENERAL OVERVIEW

The main purposes of this thesis were to unravel possible patterns of individual differences in sedentariness, obesity and physical activity in Portuguese children, and to investigate the explanatory influence of individual, familial and environmental characteristics on these patterns. The effort in responding to these aims was presented in the previous chapters and the key outcomes are summarised in Table 1. Further, a general interpretation of the main findings and implications, as well as the limitations of the present thesis, suggestions for future research and an overall summary of the main conclusions are provided in this chapter.

Table 1: Summary of the main conclusions of the papers.

Paper I

Correlates of sedentary time in children: a multilevel modelling approach

- About 6.0% of the total variance in sedentary time is explained by school effects, and 94% is explained by children's characteristics.

- Among normal-weight children, boys, children who sleep more, and those with lower family income are less sedentary. For overweight/obese children, being a boy, having more siblings, and sleeping more are associated with lower mean sedentary time.

- None of the school variables were correlated with children's sedentariness.

Paper II

Why are children different in their daily sedentariness? An approach based on the mixed-effects location scale model

- No significant differences were found between Tuesday, Wednesday and Thursday (relative to Monday) for sedentary hours, but children were more sedentary on Friday and less so on Saturday and Sunday.

- There is significant variation between subjects across days and also within subjects.

- Boys are less sedentary and more heterogeneous than girls.

- Subjects differ in how consistent/erratic they were in sedentary behaviour.

- Girls, overweight/obese children and "late maturers" showed a lower erraticism in their sedentariness.

Paper III

Overweight and obesity in Portuguese children: prevalence and correlates

- During the last decade, the prevalence of overweight/obesity of Portuguese children ranged from 19% to 35%.

- The increase in the prevalence of overweight/obesity among Portuguese children in the last decade slowed or plateaued.

- Normal-weight and overweight/obese children differ in their mean biological traits (height, weight, and parental BMI), but not in behavioural and socio-demographic characteristics.

- School explains only 2.2% of schoolchildren's variance in BMI.

- Boys, more mature children, and those whose parents have higher BMIs tend to have higher BMI. On the other hand, children with higher healthy diet scores and those with higher moderate–to-vigorous physical activity levels have lower BMI.

- None of the school-level variables were correlated with children's BMI.

Paper IV

"Fat-but-active": Does physical activity play a significant role in metabolic syndrome risk among children of different BMI categories?

- The percentage of children who complied, on average, with the WHO guidelines for mean daily physical activity was 35.2%. However, only 2.3% of children spent at least 60 min/day on all accelerometer valid days, while 18.5% did not reach the guidelines on any day.

- More than half of the sample (51.9%) was classified as overweight or obese.

- The overweight and inactive group had the worst metabolic profile, while the normal-weight and active group had the best.

- Significant differences in metabolic risk were mainly observed between BMI groups (normal-weight versus overweight/obese), but not within groups (active versus inactive).

Paper V

"Active and strong": physical activity, strength and metabolic risk in children

- In general, the inactive and low muscular strength group had the worst metabolic profile, while the active and high muscular strength group had the best.

- Significant differences were found within physical activity groups for metabolic risk: "active and high muscular strength" and "inactive and high muscular strength" have better metabolic profiles than their peers "active and low muscular strength" and "inactive and low muscular strength", respectively.

Paper VI

Relationship between sedentariness and moderate-to-vigorous physical activity in youth. A multivariate multilevel study

- Sedentariness and moderate-to-vigorous physical activity were negatively correlated within children.

- Number of siblings, socioeconomic status, and sleep time were significantly correlated with variance in both sedentariness and moderate-to-vigorous physical activity.

- Overweight/obese children and girls tend to be less physically active, but no BMI or sex effects were observed for sedentariness.

- None of the school-level covariates were statistically significant to simultaneously explain variance in sedentariness and moderate-to-vigorous physical activity.

Paper VII

Are BMI and sedentariness correlated? A multilevel study in children

- No significant correlation between BMI and sedentariness was observed in children.

- School-level variables explain 5% and 1.5% of the schoolchildren's variance in sedentariness and BMI, respectively.

- Moderate-to-vigorous physical activity was the only predictor significantly correlated with both sedentariness and BMI, with a greater effect on sedentariness as compared to BMI.

- Sleep time was negatively associated with sedentariness, but not with BMI.

- The other variables correlated with sedentariness were sex, biological maturity, bedroom media availability, and diet.

- School-level covariates (access to cafeteria, to playground equipment, and to fast-food restaurant near school) were correlated with children's sedentariness, but none of the school variables were correlated with children's BMI.

In order to present a coherent path in the general overview/discussion of the main findings, they are tied around three "clusters": prevalence and correlates of physical activity and sedentariness (papers 1, 2 and 3); relationship between physical activity, physical fitness and overweight/obesity with metabolic risk (papers 4 and 5); and the correlation between physical activity, sedentariness and BMI (papers 6 and 7).

The first "cluster" comprises papers that explored the prevalence of time spent in moderate-to-vigorous physical activity and sedentariness, overweight/obesity in children, as well as their multifaceted correlates. It was found that about half of the children were overweight/obese, less than 40% complied with the recommended daily PA guidelines (spending at least 60 min/day in moderate-to-vigorous physical activity), and, on average, spend one third of their day in sedentary activities. This scenario, apparently seen "everywhere" is also of worldwide concern, since low levels of physical activity (Guthold et al., 2010; Hallal et al., 2012; Janssen et al., 2005; Nilsson et al., 2009), increases in sedentariness (Atkin et al., 2014; Ekelund et al., 2012; Guthold et al., 2010; Pate et al., 2011) and excess weight (Due et al., 2009; Lobstein et al., 2004; Ng et al., 2014; Yngve et al., 2008) have been observed in the paediatric population in developed and developing countries from all continents.

One possible explanation for these results is related to changes in the western lifestyle observed in the last decades (Yusuf et al., 2001), which is characterized by increases in consumption of energy-dense foods in association with unparalleled rises in time spent in sedentary behaviours, especially during leisure time and transportation; systematic decreases in moderate-to-high vigorous physical activities affects children's and adolescents' health, having consequences in their later life (Berenson & Srnivasan, 2005; Deshmukh-Taskar et al., 2006; Ortega et al., 2013).

Since children's health status and behaviours are influenced by different sets of environmental characteristics, namely in their homes, neighbourhoods and schools, markers of these different contexts undoubtedly exert disparate effects on them. In other words, individual (genetic, biological and behavioural), familial, and environmental characteristics act in concert, in children, so that they tend to manifest clear inter-individual differences in their physical activity, sedentariness and BMI (Ferreira et al., 2007; Van der Horst, Oenema, et al., 2007; Van der Horst, Paw, et al., 2007). This is the main reason why we used the ecological model as a lens to investigate the role of the variables originating from different levels to study their "influences" on the main outcome traits, i.e., our focus was on the examination of correlates from different sources: biological, familial and school.

Briefly, results show that among Portuguese children, gender is significantly associated with physical activity, sedentariness and BMI - boys are more physically active, spend less time in sedentary behaviours, but have higher BMI. Sleep time and family demographics, namely socioeconomic status and number of siblings, are covariates shared by both physical activity and sedentariness, but not always with opposite signs: children with more siblings and those from higher socioeconomic status are more active and less sedentary, while those who sleep more are less active and also less sedentary. In addition, biological maturation, parental BMI, having media in the bedroom, and nutritional habits were only correlated with BMI. Interestingly, results from other papers (paper 6 and 7) may suggest that the three traits may be related to each other: BMI was a significant predictor of moderate-to-vigorous physical activity (but not for sedentariness) in paper 6, while in paper 7 the inverse relationship was observed where physical activity was a predictor of BMI and also a predictor of sedentariness.

Boys spend more time in moderate-to-vigorous physical activity and less time in sedentary behaviours (Van der Horst, Paw, et al., 2007), which can be related to cultural differences as girls generally tend to get involved in more sedentary activities during their leisure time (Blatchford et al., 2003); in addition, boys are stimulated to get more involved in physical activities/sports than girls, and even when girls take part in physical activity/sports, their options are usually for less competitive and less intense activities (Seabra et al., 2008). Since physical activity and sedentariness are complex traits (Caspersen et al., 1985), their correlates usually originate from different sources (levels); so, the familial environment has been pointed out as a significant determinant, and indicators like home socioeconomic status, and number/influence of children/siblings at home are significantly interconnected to them (Brodersen et al., 2007; Tandon et al., 2014; Tandon et al., 2012).

In the last years there is a renewed interest in a more extensive understanding of the connection between sleep time and health in youth (Chen et al., 2006; Patel & Hu, 2008; Van den Bulck, 2004). Results suggest that better sleep efficacy is related to higher physical activity levels and less sedentariness, while sleep duration is apparently linked to weight gain (Patel & Hu, 2008; Stone et al., 2013). Our results are not in complete accordance with such views given that no significant relationship was observed between sleep and BMI, and higher sleep time with higher sedentariness. There are two possible explanations for this finding: firstly, since the number of daily hours are limited, sleeping more reduces the time available to be physically active or sedentary (Olds et al., 2012); secondly, sleep efficacy, rather than sleep duration, can probably be more associated with higher physical activity levels and lower levels of sedentariness (Stone et al., 2013). However, we did not explore this relationship in our data. Finally, available data indicate that boys usually have higher BMI than girls (Serra-Majem et al., 2006); early maturing children are taller and heavier than their latter-maturing peers (Malina et al., 2004); children with TV in bedroom are at greater risk of overweight/obesity (Adachi-Mejia et al., 2007; Delmas et al., 2007); normal-weight children tend to consume less fat and high calorie beverages than their overweight peers (Storey et al., 2012); and there is familial aggregation in BMI, where parental BMI can be related to offspring BMI (Fuentes et al., 2002; Hu et al., 2013). This is the trend in the Portuguese children we studied.

Since children spend a large amount of their awake time at school, it is expected that the school environment and policies could act in concert to promote active and healthy habits which could lead to increases in physical activity levels, decreases in sedentariness, and help in reducing/controlling body weight (Cradock et al., 2007; Ridgers et al., 2007; Sallis et al., 2001; Wechsler et al., 2000). Furthermore, given the known role of the environment on the manifestation of our main outcomes, and the urge to investigate children in their living contexts, the school environment was our prime target as a major element in children's physical activity, sedentariness, and BMI. Surprisingly, the explained variance by different markers of the school environment, in each of these traits was low, from 1.5% (for BMI) to 6.0% (for sedentariness). In addition, only in sedentariness we were able to find significant school-level correlates, but not for physical activity or BMI, suggesting that individual correlates are more important, namely biological and behavioural, and the family environment. Notwithstanding the fact that most of the time children are at school is spent in sedentary behaviours (such as sitting, reading, talking), which can partially explain the higher sedentariness variance at the school level, it is more likely that the options they make during their leisure time regarding physical activity (be active or be sedentary), and food consumption at home or with parents, have more impact on their activity/sedentariness and weight gain.

The second "cluster" of issues (papers 4 and 5) tackled by this thesis addressed the complexities of the relationship between physical activity, physical fitness and obesity in the expression of metabolic risk in children and adolescents. This link has previously been studied and these variables seem to be closely related to the development of metabolic abnormalities (Saland, 2007; Steele et al., 2008; Tailor et al., 2010), although there is no consensus if they act separately or in concert (Bridger, 2009; Ruiz & Ortega, 2009; Steene-Johannessen et al., 2009). Since our results showed a high prevalence of low physical activity levels, high overweight/obesity and sedentary time, we tried to explore the joint role of these traits on the expression of metabolic risk factors.

When obesity and physical activity were analysed together in the expression of metabolic risk factors, we found that “normal-weight and active” children had a better metabolic profile than those with “overweight/obesity and inactive”. However, physical activity did not attenuate the negative role of excess weight on the development of metabolic risk, since significant differences were only observed between BMI groups (normal-weight vs overweight/obese) but not within BMI groups (active vs inactive). This reinforces the significant role of excess weight in the development of metabolic abnormalities, and that obesity may be more highly associated with metabolic disorders than lower physical activity. Moreover, when the joint role of physical activity and muscular strength on metabolic risk was studied, children classified as “active and with high muscular strength” had better metabolic profile than those classified as “inactive and with low muscular strength”. In this case, muscular strength seems to attenuate the negative role of low physical activity levels in the development of metabolic risk, since significant differences within physical activity groups (high muscular strength vs low muscular strength) were observed. Children classified as having high muscular strength, independently of their physical activity levels, showed a better metabolic profile than their peers classified as having low muscular strength.

The role of physical activity on metabolic syndrome in the paediatric population has been partly credited to be either independent of other factors or mediated by adiposity (Ekelund et al., 2007; Guinhouya et al., 2011). In addition, available data concerning the putative role of muscular strength on metabolic risk in youth showed an association between high muscular strength with a better metabolic profile (Steene-Johannessen et al., 2009), but we did not find evidence to suggest that it acts in conjunction with physical activity or adiposity. On the other hand, regarding adiposity, it has been pointed out as a stronger predictor of metabolic risk in children when compared to physical activity or physical fitness (Eisenmann, 2007; Ekelund et al., 2006). Taking together, our results suggest that there is a relationship between these variables, which should be considered when planning strategies to reduce the negative impact of excess weight on youth health. In this case, increasing physical activity and muscular strength in children having excess weight is a relevant strategy to reduce their metabolic risk, since these variables could act in mediating/reducing the undesirable effect of each other. Especially in the school context, this information should be of relevance for school principals, councils and physical education teachers when designing and implementing their annual plans to increase children's physical activity and physical fitness by both structured and non-structured physical activities/play.

Finally, we tackled the commonality of correlates for physical activity, sedentariness and BMI. Our third and last results' "cluster" is related to the papers aiming to determine if physical activity, sedentariness and BMI are correlated, or not, with each other (papers 6 and 7). To the best of our knowledge, these are the first studies where these variables were jointly analysed.

Regarding the correlation between physical activity and sedentariness, a negative and significant correlation between them was found, meaning that increasing one, the other decreases, and vice-versa. Although these behaviours are expressed as different constructs, and that they can co-occur in the same person (Leech et al., 2014; Marshall et al., 2002), it has also been suggested that increasing time spent in sedentariness leads to decreases in physical activity levels (Tammelin et al., 2007). It is possible that the negative correlation observed may be related to the fact that day is limited to 24 h, and increasing time spending in one activity, implies in the reduction of time available to be engaged in other activity.

Moreover, the correlation between sedentariness and BMI was not significant, meaning that the current idea that increasing time in sedentariness leads to increases in BMI may not be necessarily true or it may be mediated by another biological or behavioural variable. In this context, it has been suggested that food intake can be a significant mediator of the relationship between children's BMI and TV viewing (Epstein et al., 1995; Fuller-Tyszkiewicz et al., 2012), since when watching TV children are more likely to consume snacks and beverages (Carandente et al., 2009). We did not address this hypothesis in our study, but we can only speculate about it. However, the possibility of the existence of mediating factors in this relationship, namely food consumption, showed to be relevant for parents and educators roles, which can act to help children to change habits related to their food choices.

Taking all these results together (from the three clusters), it seems obvious that using the ecological model to better unravel the importance of links between children's physical activity, sedentariness and BMI was a relevant choice - the main assumption of the ecological model is that behaviours are influenced by various factors from intrapersonal, interpersonal, organizational, community and public policy levels (Sallis et al., 2008). In addition, since alterations in one level of the ecological model can affect the other levels and may influence the outcomes, directly or indirectly (Bronfenbrenner, 1979), it is important to understand how all ecological environments act in concert to regulate children's physical activity, sedentariness and obesity, and thus the use of multilevel models are the most adequate approach to generate this knowledge.

Apart from the significance of the ecological model, it has inherent weaknesses: (1) the lack of specificity about the most important hypothesized influence; (2) the lack of information about "how the broader levels of influence operate or how variables interact across levels" (Sallis et al., 2008, p. 480), and (3) its silence about putative mechanisms that express and regulate such variety in behaviours in children, adolescents and adults (i.e., across the lifespan). For example, we were not able to investigate interactions across levels or even the degree of relevance of them in each level not because we could not do it, but because we felt a substantial lack of thinking/hypothesizing about them. Nonetheless, the ecological model is a consistent and highly regarded approach to persons in their contexts, especially when intervention strategies are planned to improve child health (decreasing sedentariness and overweight/obese prevalence, and increasing physical activity levels), because interventions tend to be more effective when operating at different levels (Sallis et al., 2008). As clearly noted by Sallis et al. (2008, p. 482) the challenge for researchers is “to be creative and persistent in using ecological models to generate evidence on the roles of behavioural influences at multiple levels, and on the effectiveness of multi-level interventions on health behaviors, and to translate that evidence into improved health.”

LIMITATIONS

Notwithstanding the relevance of the present results, this thesis has limitations that should be addressed. The first limitation is related to the study design. Cross-sectional studies do not allow causal interpretations of the links among physical activity, sedentariness and BMI. However, the recent years witnessed a plethora of multi-national studies (Moreno et al., 2008; Riddoch et al., 2005) having a cross-sectional design, and even so they provided relevant information in terms of policy making, public health and education. It would be an enormous task to conduct a longitudinal study involving 12 countries, although this idea was also in the minds of the coordinating group at Pennington Biomedical Research Center.

The second limitation concerns the sample size and the fact that it comes from only one Portuguese region, which does not allow the generalization to all Portuguese children. Yet, in the very beginning, it was very clear to all sites around the world that it was not a purpose of the ISCOLE to have representative data of each country, but to gather multi-country information from different levels, increasing its heterogeneity as well as power. However, since in the Portuguese context schools are quite similar in their environments and policies, the use of only 23 schools may have limited the identification of the school roles on children physical activity, sedentariness and BMI.

The third limitation concerns the fact that sedentariness is a very complex trait, usually involving a wide array of behaviours. The objective measure of sedentariness (accelerometry) we employed does not provide extensive information about the types, frequency and duration of activities children are involved with. Although a questionnaire was also applied to all children, it may not provide sensible information about all sedentary behaviours children are involved with or even the moment when they occurred. The use of direct observation of children’s activities, or the use of diaries, would be of relevance, although it would probably be unfeasible with the present sample size across all the sites.

The fourth limitation concerns the fact that we circumscribed our sample age to 9-11 years which does not allow any inference to adolescence or early childhood nor was this intended.

The fifth limitation concerns the small sample size in the ancillary study. However, costs related to analytical data (blood analysis) prohibited a larger sample size. Further, we did not explore the relationship of other physical fitness components with metabolic risk, as this would probably provide “interesting” results.

IMPLICATIONS AND OPPORTUNITIES FOR FUTURE RESEARCHES

Implications

We are convinced that the results of this thesis have educational as well as policy making implications concerning physical activity/exercise/sports participation, sedentariness and obesity.

Firstly, we showed that at 10 years of age a large proportion of Portuguese children have excess weight, do not comply with the physical activity guidelines, and spend great portions of their day in sedentary activities. This information calls for families, schools and public health authorities' attention/concern in order to change this scenario. The first call is towards schools, since the school context is also an important agent to promote/develop policies to increase children's physical activity/physical exercise/sports participation to reduce their time in sedentary behaviours and thus help in weight control; further, physical education classes should be more interesting/challenging providing ample opportunities for individual success in all tasks, and playground equipment should always be available for children to use.

Secondly, although the relationship between physical activity, sedentariness and BMI have been well explored, we showed that they have different determinants and this is relevant information that should be taken into account when planning intervention strategies to promote active and healthy lifestyles in children.

Thirdly, children differ in their sedentariness from each other, as well as in their patterns, meaning that individual differences have to be considered when designing intervention programs to reduce sedentariness.

Fourthly, the family environment plays an important role on children's behaviour and health. With respect to overweight/obesity, we found that children whose parents have higher BMI also tend to have high BMI, i.e., there is familiality in this trait. This calls for careful paediatrician interventions so that they pay closer attention to these children in order to prevent their future obesity status.

Fifthly, the Portuguese school environment does not seem to explain a large proportion of children’s physical activity, sedentariness, and BMI variance, and several of their correlates did not show any significant association. However, schools have prominent roles on children physical activity, sedentariness and obesity, by developing and promoting active and healthy lifestyles helping them to make “better” choices in the use of their time outside school as well as in their nutritional choices.

Sixthly, physical activity, physical fitness and BMI are associated with children’s metabolic risk, acting separately or in association with each other. Obesity appears to be the strongest correlate of metabolic risk, reinforcing the need to reduce this risk during childhood, and also in later life. Further, high levels of physical activity and high levels of muscular strength were also associated with decreasing metabolic risk. Given that muscular strength attenuates the negative role of low levels of physical activity on metabolic risk, this information should be carefully used by paediatricians and physical education teachers.

Seventhly, and lastly, physical education teachers, school authorities, paediatricians and health care professionals have to have in mind that physical activity and sedentariness, although correlated, are not different sides of the same coin, but they co-occur in the same child, i.e., a given child can spend sufficient time in moderate-to-vigorous physical activity but, at the same time, he/she can also spend a high proportion of his/her time in sedentary activities. Since both behaviours seem to be related to overweight/obesity development in children, as well as in their co-morbidities, interventions should be designed to act in both behaviours. In addition, since BMI and sedentariness are probably not correlated, suggesting that their link may be mediated by other covariates, this calls again to school policies regarding children healthy eating habits.

Opportunities for future researches

From the large set of data collected in the ISCOLE-Portugal, as well as in its ancillary study, only a small portion was explored. Further, we have not yet considered the joint analysis with other European countries (UK and Finland), or Brazil, and less so with the other 11 countries of the ISCOLE project.

Below we provide a short list of questions that will be addressed in a near future considering only the Portuguese data set:

<u>Questions related to the prevalence and correlates of physical activity, sedentariness and overweight/obesity:</u>

- What is the proportion of time children are physically active, considering their total physical activity? Does it vary substantially within subjects? If so, which are the main predictors and why?
- Are children different in their daily physical activity? What is the variance within- and between-subjects in physical activity over a week?
- Do moderate-to-vigorous physical activity and sedentariness vary along the day? Is there a pattern in this variation? Is this pattern different between school-days and weekend day? Which variables are associated with these patterns?
- Does the neighbourhood environment play a significant role in children's physical activity, sedentariness and BMI?
- At the home environment, is parental support a significant predictor of children's physical activity and sedentariness? Does the availability of sports equipment explain the expression of children's physical activity and sedentariness? Does the quantity and quality of food availability act as suitable predictors of children's BMI?

<u>Questions related to the relationship between physical activity, physical fitness, obesity and its co-morbidities:</u>

- Can physical fitness, namely muscular strength, attenuate the role of obesity in the expression of metabolic risk?

- Is sedentariness related to metabolic risk? Can sedentariness increase the risk for metabolic risk within BMI groups? On the other hand, can sedentariness decrease the role of physical activity in the reduction of metabolic risk?
- Which can be more efficient to reduce the negative role of obesity on metabolic risk: low levels of sedentariness, high levels of physical activity, or high levels of physical fitness? Do these covariates act independently or in conjunction?
- Is screen time a significant predictor of metabolic risk?

<u>Questions related to the correlation between physical activity, sedentariness and BMI</u>

- Are physical activity and BMI correlated? Are their covariates similar? Is this link mediated by other variables?
- Which variables mediate the relationship between sedentariness and BMI? Do nutritional habits play relevant roles in this relationship?
- Does the correlation between physical activity and BMI, and sedentariness and BMI, differ between sex and/or socioeconomic status? Further, is the correlation between physical activity and sedentariness similar among weight groups?

CONCLUSIONS

Using data from ISCOLE, from distinct levels, this cross-sectional study tried to unravel the relationships between physical activity, sedentariness and BMI in children. Since study design has a hierarchical foundation, multilevel models were used to come to terms with the complexities of the data structure. In addition, this is the first study to examine between- and within-subjects variation in sedentariness using mixed-effects location scale model, and also to jointly analyse physical activity and sedentariness, and sedentariness and BMI, as well as their correlates in children within their school contexts.

Here are the main conclusions:

- ISCOLE Portuguese children showed high prevalence of overweight/obesity, low levels of moderate-to-vigorous physical activity, and high amount of time spent in sedentary behaviours.

- There is a little school-level variability in children’s sedentariness, moderate-to-vigorous physical activity, and BMI.

- Variance in sedentariness, moderate-to-vigorous physical activity, and BMI can be explained by child-level characteristics (biological, behavioural, environmental), but none of school context variables explain moderate-to-vigorous physical activity and BMI variance.

- Normal-weight and overweight/obese children differ in their sedentariness correlates, and also showed differences in their biological traits.

- There is a significant erraticism of sedentariness in children along an entire week, meaning that children differ in their sedentariness patterns according to the days of the week.

- A relationship between moderate-to-vigorous physical activity, muscular strength, and BMI may exist in the expression of metabolic risk in children.

- Moderate-to-vigorous physical activity and sedentariness are negatively correlated meaning that although they are different constructs, increasing time spent in one decreases the time spent in the other.

- No significant correlation was observed between sedentariness and BMI.

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